Ospedali Galliera di Genova

Cited in Index Medicus/MEDLINE, BIOSIS Previews, SCOPUS

PATHOLOGICA

JOURNAL OF THE ITALIAN SOCIETY OF ANATOMIC PATHOLOGY AND DIAGNOSTIC CYTOPATHOLOGY,
ITALIAN DIVISION OF THE INTERNATIONAL ACADEMY OF PATHOLOGY

PROCEEDINGS OF TRIENNIAL CONGRESS SIAPeC-IAP

October 12th-15th, 2022

SIAPeC CASE REPORT AWARD 2022 - BREAST EDITION

SIAPEC-IAP
Società Italiana di Anatomia Patologica e Citopatologia Diagnostica,
Divisione Italiana della International Academy of Pathology

01
VOL. 115 Supplement 1
FEBRUARY 2023

PACINI EDITORE MEDICINA

Periodico bimestrale - Aut. Trib. di Genova n. 75 del 22/06/1949 - ISSN: 1591-951X (Online)

Ospedali Galliera di Genova

Cited in Index Medicus/MEDLINE, BIOSIS Previews, SCOPUS

PATHOLOGICA

JOURNAL OF THE ITALIAN SOCIETY OF ANATOMIC PATHOLOGY AND DIAGNOSTIC CYTOPATHOLOGY, ITALIAN DIVISION OF THE INTERNATIONAL ACADEMY OF PATHOLOGY

SIAPEC-IAP
Società Italiana di Anatomia Patologica e Citopatologia Diagnostica, Divisione Italiana della International Academy of Pathology

01
Vol. 115 Supplement 1
February 2023

PACINI EDITORE MEDICINA

Publisher
Pacini Editore S.r.l.
Via Gherardesca, 1
56121 Pisa, Italy
Tel. +39 050 313011
Fax +39 050 3130300
info@pacinieditore.it
www.pacinimedicina.it

C O N T E N T S

PATHOLOGICA 2023;115(Suppl.1):S1-S110;
https://doi.org/10.32074/1591-951X-862

PROCEEDINGS OF TRIENNIAL CONGRESS SIAPeC-IAP 2022

October 12th-15th, 2022

SIAPeC CASE REPORT AWARDS 2022 - Breast edition

Mercoledì 12 Ottobre 2022

Sala Mantegna 13.45-14.55

PATOLOGIA MAMMARIA 1

Moderatore: I. Castellano

ID 800
ROLE OF E3 LIGASE PRAJA2 IN HUMAN BREAST CANCER. MORPHOLOGICAL, IMMUNOPHENOTYPIC AND MOLECULAR STUDY. BREAST PATHOLOGY

F. Puccio[1], M. Fuggi[1], D. Russo[1], P. Isernia[1], A. Feliciello[2], L. Rinaldi[2], L. Insabato[1]

[1] Dept of Advanced Biomedical Science, University of Naples, Federico II; [2] Dept of Molecular Medicine and Medical Biotechnology, University of Naples, Federico II

Objectives: Breast cancer is the most common malignancy in adult women, including many histological subtypes, each with a different prognosis based on biological and clinical features. Identifying novel biomarkers for early cancer detection and further mechanisms underlying breast cancer growth and progression will help to treat this aggressive tumor. Downregulation of Praja2 is associated with a marked upregulation of tyrosine kinase receptors, with consequent activation of MAPK signal transduction. We aim to evaluate the expression of Praja2 in breast cancer.
Materials and methods: We retrieved from our database 60 breast cancers. Data regarding the histologic type, grading and molecular status of the neoplasm were collected. In each case Praja2 expression by tumor and nonneoplastic parenchyma was assessed. Based on the level of expression of the protein we defined a scoring system ranging from 0 (absence of expression) to 3 (maximum expression). All the cases were stratified into "low-expression" (score 0,1) and "high-expression" (score 2,3). Moreover, cervical carcinoma-derived cells (HeLa) were cultured in Dulbecco modified Eagle's medium, containing 10% fetal bovine serum in an atmosphere of 5% CO2. Western blot analysis was performed using an anti-EGFR antibody (1:1000) and anti praja2 antibody (1:1000).
Results: Here we report that Praja2 expression is decreased in breast cancer. In particular, on immunohistochemistry, it was downregulated in more than 50% of the tumors analyzed. The variability in protein expression depended on receptor profile and molecular classification. In particular in triple-negative tumors Praja2 levels were generally higher (3+ score) compared to other phenotypical and molecular classes. Finally, in mammary tumors where praja2 was downregulated EGFR was upregulated.
Conclusions: Better addressing praja2 function in breast tissue could be a relevant molecular mechanism to better understand the pathogenesis of breast cancer, also providing a new biomarker and a possible therapeutic target for one of the most lethal tumors of our century.

ID 807
UNUSUAL AND DIFFICULT BREAST LESIONS: RADIOLOGIC AND PATHOLOGIC CORRELATIONS

A. D'Amuri[1], M. Capodieci[2]

[1] Anatomic Pathology Unit,"A. Perrino" Hospital, ASL Brindisi, Brindisi, Italy; [2] Department of Breast Radiology, ASL Brindisi, Brindisi, Italy

Objectives: The presentation of unusual breast lesions is quite challenging, knowing that a wide array of benign and malignant lesions may be encountered in practice. Familiarity with their radiologic appearances as well as the pathologic findings are important since the detection of such lesions can impact their clinical management.
Materials and methods: Cases search was conducted through the archives of the electronic files of the Pathology and Radiology Unit in 2022. We identified two patients who underwent breast core needle biopsies.
Results: The 1st patient was 53 years old and had history of breast carcinoma. In 2018, breast right inferior quadrantectomy was deemed consistent with invasive ductal carcinoma. In February 2022, at ultrasonography super-fluid collection with an underlying irregular, hypoechoic area with striking posterior acoustic shadowing simulating malignancy was discovered in the inferior sector of the right breast. A core needle biopsy was performed and routine and immunohistochemical (CD68 +; CKAE1/AE3 and S-100 -) examination of the lesion were considered with inflammation and fibrosis surrounding foreign material composed of homogenous, basophilic and irregular pieces of material (oxidize cellulose). The 2nd patient was 48 years old and had history of bilateral mastectomy for invasive lobular carcinoma. In May 2022, at ultrasonography and mammography discovered a sub-scarring nodule of left superior external quadrant of the breast. The lesion was located between breast implant and thoracic wall and the biopsy was very difficult to perform. Histological examination showed a desmoplastic stroma with a dense infiltrate of inflammatory cells that has entrapped few neoplastic cells with a typical "*indian file*" growth pattern. Immunohistochemical analyses showed staining for CK19, ER and PR receptors. The diagnosis was invasive lobular carcinoma.
Conclusions: Radiologic-pathologic correlation is crucial in the diagnosis of the spectrum of breast diseases. The relation of the underlying pathology of a lesion explains its imaging appearance. Close collaboration between radiologists and pathologists is copacetic and greatly influences clinical decisions and management.

ID 814
INTER-PLATFORM REPRODUCIBILITY OF MISMATCH REPAIR STATUS ASSESSMENT IN BREAST CANCER

M. Ivanova[1], E. Sajjadi[1], K. Venetis[1], G. Bonizzi[1], P. Rafaniello[2], U. Malapelle[3], E. Guerini-Rocco[1], M. Fassan[4], C. Scatena[5], N. Fusco[1]

[1] IEO, European Institute of Oncology IRCCS, University of Milan, Italy; [2] Fondazione IRCCS Ca' Granda Ospedale Policlinico, Milan, Italy; [3] University Federico II, Naples, Italy; [4] Veneto Institute of Oncology IRCCS, University of Padua, Italy

[5] Pisa University Hospital, University of Pisa, Italy

Objectives: Mismatch repair (MMR) status assessment is clinically relevant in breast cancer (BC) for prognostication and therapy selection, although MMR deficiency (dMMR) is rare (~3%) in these tumors. The reference method for MMR testing depends on IHC for four MMR proteins and/or microsatellite instability investigation. These tests have been developed for colorectal and endometrial carcinomas screening and therefore there are no BC-specific guidelines. We sought to characterize the reproducibility of MMR testing using the three most common IHC platforms available in Italian pathology laboratories.

Materials: A total of 1,023 BC samples were subjected to IHC using antibodies against MLH1, PMS2, MSH2, MSH6 on three different platforms, i.e. Leica BOND-III (Platform A), Ventana Benchmark Ultra (Platform B), Dako Omnis (Platform C). Cases were classified as dMMR based on the complete loss of immunoreactivity in at least one of the MMR markers. Differences in the IHC results were assessed using Chi-squared tests; concordance was calculated by Cohen's (dual) and Fleiss (triple) κ coefficients.

Results: dMMR was observed in 42 (4.1%), 70 (6.8%), and 75 (7.3%) cases, according to platforms A, B, and C, respectively. Platforms A-B agreement was fair (n = 20; 2.0% cases; 93.4%; κ 0.32), A-C was moderate (n = 29; 2.8% cases; 94.4%; κ 0.46), akin B-C (n = 25; 2.4% cases; κ 0.29). Taken together, all platforms showed a poor agreement (n = 17; 1,7% cases; Fleiss' κ -0.2). The largest discrepancy was observed in triple-negative BCs. The best single-marker agreement was on PMS2, while the lowest on MSH1.

Conclusions: The low interplatform reproducibility of MMR status assessment by IHC demonstrates the need for harmonization studies specifically dedicated to BC, with the aim of developing tumor-specific recommendations and guidelines.

ID 828
CLINICO-PATHOLOGICAL CORRELATIONS AND PROGNOSTIC IMPACT OF TRIPLE NEGATIVE BREAST CANCER LAR SUBTYPE

L. Serrao[1], J. Metovic[2], F. Sibona[1], C. Tamperi[1], A. A. Ricci[1], G. Orlando[2], L. Bertero[1], I. Castellano[1]

[1] Department of Medical Sciences, University of Turin, Turin, Italy; [2] Department of Oncology, University of Turin, Turin, Italy

Objectives: The present study aims to identify the clinico-pathological and molecular features of a series of triple negative breast cancers (TNBC) with emphasis on AR-positive cases, defined as Luminal Androgen Receptor (LAR) subtype.

Materials and methods: Hematoxylin and eosin glass-slides were reviewed and representative tumor block for subsequent molecular and immunohistochemical (IHC) analyses was selected. Molecular data were obtained from 42 patients, by means of NanoString technology employing Breast Cancer 360™ Panel. AR, FOXA1, Ki-67, CK 5/6, FGFR4 and GCDFP-15 were analyzed by IHC. Clinico-pathological data were also collected.

Results: Surrogate LAR categorization by IHC analyses did not coincide with molecular data, as within 15 AR+ cases, 8/15 (53%) were confirmed at molecular level. The rest were categorized as BLIA (19/42, 45,3%, 5 AR+), BLIS (11/42, 26,2%, 2 AR+) and MES (4/42, 9,5%).

When comparing 8 LAR and 34 non-LAR tumors, we observed that LAR were characterized by low grade and low Ki-67, higher FOXA1 expression, absence of CK 5/6, and more frequent apocrine histotype being GCDFP-15 more commonly expressed in this tumor subtype. No significant difference was found regarding FGFR4 immunoexpression. At molecular level, among other genes, we observed upregulation of *AR, FOXA1, FGFR4, MLPH, ERBB2* and downregulation of *FOXC1, GABRP, ASPM, MCM2, FZ9D* in LAR category compared to non-LAR ($p < 0.01$).

In cases bearing AR+ IHC status compared to AR-, we observed, among others, upregulation of *AR, FOXA1, FGFR4, SIDT1, AGR2* and downregulation of *FOXC1, DSC2, KRT6B, CKS1B, NDC80* ($p < 0.01$).

Conclusions: This study characterized LAR tumors from clinico-pathological and molecular perspective, offering closer insights regarding their biology and tumor microenvironment. Accurate distinction of these neoplasms is important, as these patients, being resistant to chemotherapy, may benefit from tailored-therapeutical management. Specifically, overexpression of AR and additional biomarkers such as FGFR4 in this subtype, may represent future directions in treatment approach.

ID 830
PROGNOSTIC AND BIOLOGICAL ROLE OF NDRG1 IN INVASIVE BREAST CARCINOMA AND IN TNBC

L. Schirosi[1], C. Saponaro[1], M. Sonnessa[1], S. De Summa, R. D'Aprile[1], S. Bravaccini[2], F. Pirini[2], M. Salzet[3], F.A. Zito[1], D.Vergara[4].

[1] IRCCS Istituto tumori "Giovanni Paolo II", Bari, Italy; [2] Biosciences Laboratory, IRCCS Istituto Scientifico Romagnolo

per lo Studio e la Cura dei Tumori (IRST), Meldola, Forlì-Cesena, Italy; [3] Laboratoire Protéomique, Réponse Inflammatoire Spectrométrie de Masse (PRISM), Inserm U1192, University of Lille, CHU Lille, F-59000 Lille, France; [4] Department of Biological and Environmental Sciences and Technologies (DiSTeBA), University of Salento, Lecce, Italy

Objectives: N-myc downstream regulated gene 1 (NDRG1) is a protein that plays a central role in various biological processes including invasion, differentiation and metabolism. Its overexpression is an indicator of poor prognosis in various tumor types, including breast cancer (BC). In BC, NDRG1 regulates lipid metabolism and vesicle transport and it is regulated under stress conditions.
Methods: We used liquid chromatography-mass spectrometry (LC-MS/MS) and RNAseq analysis to identify the proteomic and transcriptomic profiles of 12 formalin-fixed paraffin-embedded (FFPE) BC samples. Further, NDRG1 expression was evaluated by immunohistochemistry on 212 primary BC samples [140 luminal BCs and 72 triple negative cancers (TNBCs)]. Statistical evaluations were performed with the Prism version 5.00 software package, with the statistical significance set at $p < 0.05$.
Results: NDRG1 protein and transcript resulted overexpressed in TNBCs compared with Luminal samples in the 12 FFPE. In 212 primary BC, NDRG1 was also more expressed in TNBC compared with the Luminal phenotype ($p = 0.0001$), confirming the LC-MS/MS and RNAseq Results: Spearman correlation showed an inverse correlation between NDRG1 and hormonal receptors ($p < 0.0001$, $r = -0.44$) and a direct correlation with Ki67 ($p < 0.0001$, $r = 0.50$). Kaplan- Meier curves revealed that BC patients with high respect to low NDRG1 expression had a worse disease free survival (DFS) ($p = 0.011$). Further, TNBC patients with high NDRG1expression had the worst DFS with respect to Luminal tumours ($p = 0.0029$).
Conclusions: This study shows a pivotal role of NDRG1 during cancer progression in invasive BC and in particular in the subgroup of TNBC. Furthermore, NDRG1 impacts on the survival of patients assuming a potential role as clinical marker.

ID 841
COMPARISON OF NGS AND RTQPCR FOR PIK3CA TESTING IN HR+/HER2- BREAST CANCER ON METASTATIC AND MATCHED PRIMARY TUMOR SAMPLES

K. Venetis[1], F. Pepe[2], E. Sajjadi[1], M. Ivanova[1], D. Vacirca[1], A. Rappa[1], M. Barberis[1], E. Guerini-Rocco[1], U. Malapelle[2], N. Fusco[1]

[1] IEO, European Institute of Oncology IRCCS, Milan, Italy; [2] University of Naples Federico II, Naples, Italy.

Objectives: Activation of the phosphatidylinositol-3-kinase (PI3K) pathway via *PIK3CA* mutations occurs in around 40% of hormone receptor-positive (HR+)/ human epidermal growth factor receptor-2-negative (HER2-) advanced breast cancers. Accurate detection of such somatic alterations is crucial considering their clinical actionability. Here, we sought to assess the concordance rate for *PIK3CA* molecular analysis between different technical platforms in metastatic and matched primary tumors.
Materials and methods: From our Institutional registry, n = 16 metastatic HR+/HER2- breast cancers, which were found to harbor *PIK3CA* mutations via next-generation sequencing (NGS) assays [custom panel and Oncomine Comprehensive Assay (OCA) v3 (Thermo Fisher Scientific, Waltham, MA, USA)], were selected. Matched primary tumors (n = 8) were subjected to *PIK3CA* testing with NGS. The analytical performance between NGS and a semi-closed RTqPCR (EasyPGX®, Diatech Pharmacogenetics, Italy) was assessed in metastatic and primary tumors.
Results: Overall, upfront NGS detected *PIK3CA* mutations in exons 7, 9, and 20. A concordance rate of 100.0% was observed between primary and metastatic tumors. The analysis of primary tumors (n = 8) and 13/16 (81.3%) metastatic samples with RTqPCR revealed a concordance of 42.9%. Analytical performance comparison showed that the two technologies were concordant in 7/13 metastatic cases (53.8%). Interestingly, visual inspection of the RTqPCR raw data increased the concordance to 76.9%. In primary tumors, consensus was observed in 5 (62.5%) cases.
Conclusions: The concordance rate analysis shows that upfront *PIK3CA* molecular testing with NGS appears to be more efficacious compared with RTqPCR. Primary tissues reflect *PIK3CA* mutational status when tested with NGS. RTqPCR is simple with short turnaround time, however, trained personnel are required for accurate results interpretation.

ID 872
ATYPICAL AND EPITHELIOID CELL MYOFIBROBLASTOMA OF THE BREAST: A POTENTIAL PITFALL OF MALIGNANCY

S. Poidomani[1], G.M. Vecchio[1], L. Salvatorelli[1], G. Angelico[2], G. Attanasio[1], G. Magro[1].

[1] Department of Medical and Surgical Sciences and Advanced Technologies, G.F. Ingrassia, University of Catania, Catania, Italy., [2] Pathology Unit, Cannizzaro Hospital, Catania, Italy

Objectives: Myofibroblastoma (MFB) of the breast is a benign spindle cell tumor showing fibroblastic e myofibroblastic differentiation. Over the past decades the spectrum of this tumor has been expanded by the recognition of numerous variants, including the atypical and the epithelioid cell variants. The recognition of these latter two variants is crucial to prevent a misdiagnosis of malignancy, when dealing with small biopsies.

Materials and Methods: We collected from the archives of our institution three cases of atypical MFB and 5 cases of epithelioid cell MFB. Five cases occurred in male patients and three cases in female patients. For each single case the clinico-radiologic features were available. In addition, core biopsies were available in 4 cases (one atypical MFB and 3 epithelioid cell MFBs).
Results: All tumors had well-circumscribed borders and mainly consisted of medium- to large-sized epithelioid cells (> 70% of the entire lesion) and, to a lesser extent, of round and spindled cells. The neoplastic cells exhibited several patterns, including single cell files, alveolar and nested patterns. Moderate to focally severe nuclear atypia was seen in all the cases labeled as "*atypical MFB*". Low mitotic activity (< 2 mitoses x 10 HPF) and neither atypical mitoses nor necrosis were observed were negative. Neoplastic cells were positive to vimentin, and desmin and variably to a-ASMA, CD34, CD10, bcl-2, CD99, ER, PR, and AR; epithelial markers were negative.
Conclusions: Atypical and epithelioid-cell MFB is a potential pitfall of malignancy, especially on core biopsy. Atypical MFB needs to be distinguished from a high grade sarcoma, while epithelioid cell MFB often mimics invasive lobular carcinoma. The absence of infiltrative margins, the low mitotic activity and the lack of epithelial markers are helpful for achieving the correct diagnosis.

Mercoledì 12 Ottobre 2022

Sala A 13.45 - 14.55

MISCELLANEA 1

Moderatori: C. Mescoli, S. Pizzolitto

ID 865
INCIDENTAL NEOPLASIA IN EXPLANTED LUNGS FROM LUNG TRANSPLANT RECIPIENTS: MORPHOLOGY AND PROGNOSIS

F. Lunardi[1], V. Verzeletti[1], F. Pezzuto[1], F. Fortarezza[1], A. Kilitci[1], M. Schiavon[1], M. Loy[1], F. Rea[1], F. Calabrese[1]

[1] Department of Cardiac, Thoracic, Vascular Sciences and Public Health, University of Padova, Padova, Italy.

Objectives: Lung transplantation (LTx) represents the only therapy for advanced lung diseases, but undiagnosed neoplasms are found in 0.5% to 2.4% of explanted lungs. One of the most common indications for LTx is idiopathic pulmonary fibrosis (IPF) that represents the most frequent native disease associated with neoplasms. The main goal of the study was to evaluate the prevalence of incidental lung cancer in explanted lungs from IPF recipients mainly focusing on their morphological/molecular characteristics and prognostic impact.
Materials and methods: All patients undergoing LTx for interstitial lung disease in our Centre from 1995 to May 2022 were retrieved and clinical, radiological and pathological review was performed only in cases with available full clinical/radiological data. According to the most recent international guidelines, 45 patients were confirmed as IPF. Lung tissue slides were carefully re-evaluated to detect and grade dysplasia or neoplasia according to the 2021 WHO. Overall survival (using Kaplan-Meier analysis) of IPF patients with invasive neoplasia was estimated and compared to that of other IPF recipients.
Results: 5(11%) of patients showed dysplasia/in situ carcinoma and 5(11%) an invasive neoplasia, unrecognized before transplantation. The invasive tumors were all lung adenocarcinomas with a prevalent acinar pattern in 4(80%) cases, grade 2, with a mean of 3 mitoses/HPF, without STAS features. Poorly differentiated-solid pattern (grade 3) was found in one case (20%). Pleural invasion was seen in 4(80%) cases and peribronchial lymph node metastases in 3(60%). EGFR was wild type in all adenocarcinomas. IPF recipients with invasive neoplasia showed a lower overall survival than those with or without dysplasia/neoplasia.
Conclusions: A high prevalence of unrecognized invasive tumor was found in explanted IPF lungs. Careful pathological examination of explanted lungs in IPF patients is critical as it can majorly influence their immunosuppressive regimens, surveillance and overall prognosis.A large molecular profiling of invasive and non invasive lung tumors is ongoing and will be presented at the conference.

ID 938
BAFF AND BAFFR EXPRESSION IN AHR HEART TRANSPLANTATION: NEW PROMISING MARKERS?

LA De Nicola[1],L Bardhi[1],E Sorrentino[1],G Ventrella[1],M Vannucchi[1],C Defraia[1],F Rossi[1],S Bernazzali[2],D Tudose[1],C Bellan[1]

[1]Medical Biotechnology,University of SIENA; [2]UOC Cardiochirurgia, AOUS

Objection: Modern cardiac transplant success is related to the ability to diagnose acute rejection by performing endomyocardial biopsy (EBM).The diagnosis of acute cellular rejection is made by identifying specific histological features(inflammatory infiltrate and myocyte damage)with good correlation between clinical and histology.On the other hand, patients with acute humoral reaction (AHR) span a rather wide clinical range, from individuals with clinically manifest

graft dysfunction to others completely asymptomatic. In the latter, the role of EBM in order to diagnose AHR is of particular importance. Using the current histologic and immunophenotypic (anti-CD4 and anti-CD68 antibodies) criteria, cases persist in which there are an incongruence between the clinic and the histology. With the expanding use of anti-C4d antibody by transplant pathologists, several shortcomings of C4d were identified, which appear to be a less-sensitive marker than initially thought. B cell activating factor (BAFF) belonging to TNF family cytokine enhances B-cell proliferation and differentiation. Recently the roles of BAFF in transplantation immunity and the induction of transplantation immune tolerance have attracted attentions.
Materials and methods: To investigate the role of BAFF and its receptor in AHR in heart transplantation, we conducted a single center retrospective study analyzing by IHC the expression of BAFF and BAFFR in EBM samples of 8 patients with AHR then compared them with a control group of 8 patients who did not experience AHR. We also investigate the expression of anti-CD68 and anti-CD16 antibodies to better define the role of macrophages M1-M2 polarization.
Results: BAFF and BAFFR antibodies were positive in cytoplasm of endothelial cells in all the EBM with AHR without correlation with C4d expression, which resulted positive only in 2 out of 8 patients. We observed a significant M2 macrophages polarization in patients with AHR in contrast to the control group.
Conclusions: BAFF and BAFFR expression in endothelial cells in addition to M2 macrophages infiltration are associated to AHR and could be, in association with C4d, promising biomarkers to predict heart AHR.

ID 940
FIBRILLARY GLOMERULONEPHRITIS: A CASE SERIES WITH A CRUCIAL ROLE OF DNAJB9 AS A DIAGNOSTIC MARKER

C. Pitino[1], A. Gambella[1], A. Barreca[2], A. Nocifora[2], M. Giarin[2], L. Biancone[1,3], D. Roccatello[4], M. Papotti[5], P. Cassoni[1]

[1] Department of Medical Sciences, University of Turin, Turin, Italy; [2] City of Health and Science Hospital, Turin, Italy; [3] Division of Nephrology dialysis and transplantation, City of Health and Science Hospital, Turin, Italy; [4] Nephrology and Dialysis Unit, San Giovanni Bosco Hospital, Turin, Italy; [5] Department of Oncology, University of Turin, Turin, Italy

Objectives: Fibrillary glomerulonephritis (FGN) is a rare glomerular disease characterized by a challenging diagnostic workup. The final diagnosis ultimately necessitated the ultrastructural identification of the 20 nm-thick randomly-oriented fibrillar deposit, thus requiring dedicated laboratory facilities, pathologist expertise, and prolonged diagnostic turn-around time. Recently, the immunohistochemical expression of DNAJB9 emerged as a putative FGN diagnostic marker. This study aims to assess the expression of DNAJB9 in a large series of FGN cases and its role as a diagnostic marker of FGN.
Materials and methods: 74 cases of FGN diagnosed between January 1992 and May 2022 were retrospectively retrieved from the Pathology Unit of the University of Turin. As a control group, 98 cases representing the most common diagnostic differentials of FGN (including 21 cases of immunotactoid glomerulopathy) were also collected. We reviewed the histopathological, immunofluorescent, and ultrastructural features of all cases to confirm the diagnosis, and then evaluated the immunohistochemical expression of DNAJB9 (Rabbit Polyclonal, ThermoFisher) and correlated it with the pathologic and clinical findings.
Results: 73 of the 74 cases of FGN resulted positive to DNAJB9 with a moderate-to-intense glomerular expression, including cases presenting combined diseases (such as one case of combined FNG and ANCA-related glomerulonephritis). In addition to the glomerular expression, 34 cases resulted positive to DNAJB9 also in the tubules, 5 cases in small vessels, and 6 cases in both tubules and arteries. None of the control cases resulted positive to DNAJB9.
Conclusions We confirmed the role of DNAJB9 as a diagnostic marker of FGN, even in early cases with tiny deposits. Its adoption in the clinical routine will allow a faster, more feasible, low-cost and accurate diagnosis of FGN.

Mercoledì 12 Ottobre 2022

Sala B 13.45 - 14.55

PATOLOGIA APPARATO DIGERENTE 1

Moderatore: C. Luchini

ID 744
INTRADUCTAL TUBULOPAPILLARY NEOPLASM (ITPN) OF THE PANCREAS: A DISTINCT ENTITY AMONG PANCREATIC TUMORS. PANCREATIC PATHOLOGY

G. Paolino[1], I. Esposito[2], S.M. Hong[3], O. Basturk[4], P. Mattiolo[1], T. Kaneko[5], N. Veronese[6], A. Scarpa[1,7], V. Adsay[8], C. Luchini[1,7*]

[1] Department of Diagnostics and Public Health, Section of Pathology, University and Hospital Trust of Verona, Verona, Italy; [2] Institute of Pathology, University Hospital of Duesseldorf, Duesseldorf, Germany; [3] Department of Pathology, Asan Medical Center, University of Ulsan College of Medicine, Seoul, South Korea; [4] Department of Pathology, Memorial Sloan Kettering Cancer Center, New York, USA; [5] Department of Hepato-Biliary-Pancreatic Medicine, NTT Medical

Center, Tokyo, Japan; [6] Department of Internal Medicine, University of Palermo, Palermo, Italy; [7] ARC-Net Research Center, University and Hospital Trust of Verona, Verona, Italy; [8] Department of Pathology, Koç University Hospital and Koç University Research Center for Translational Medicine (KUTTAM), Istanbul, Turkey.

Objectives: Intraductal tubulopapillary neoplasm (ITPN) of the pancreas is a recently recognized entity. We aimed to determine its main features with a systematic review and an integrated statistical approach.
Materials and methods: PubMed, SCOPUS and Embase were searched for studies on pancreatic ITPN. Clinicopathological, immunohistochemical (IHC), and molecular data were collected (128 patients). A survival analysis and a comparative analysis of the molecular alterations of ITPN with those of pancreatic ductal adenocarcinoma (PDAC) and intraductal papillary mucinous neoplasm (IPMN) from reference cohorts (International Cancer Genome Consortium-ICGC dataset and The Cancer Genome Atlas-TCGA program) were conducted.
Results: Clinicopathological data: the head is the main site; associated adenocarcinoma was reported in 60% of cases; nodal metastases were rare. IHC data: MUC1 (> 90%) and MUC6 (70%) were the most expressed mucins. ITPN lacked the intestinal marker MUC2 and the IPMN marker MUC5AC. Molecular features: *KRAS*, *TP53*,*CDKN2A*, *SMAD4*, *GNAS, RNF43* were less altered in ITPN than in PDAC/IPMN ($p < 0.001$); *MCL* amplifications, *FGFR2* fusions, *PI3KCA* mutations were common ($p < 0.001$). Survival analysis: "pure" branch duct ITPN showed the lowest risk of recurrence.
Conclusions: ITPN is a distinct entity with specific characteristics. Its recognition is fundamental for its clinical implications and for the enrichment of potential targets for precision oncology.

ID 749
MOLECULAR SUBTYPING OF GASTROESOPHAGEAL DYSPLASIA HETEROGENEITY ACCORDING TO TCGA/ACRG CLASSES

V. Angerilli[1], G. Pennelli[1], F. Galuppini[1], S. Realdon[2], A. Fantin[2], E. Savarino[3], F. Farinati[3], L. Mastracci[4], C. Luchini[5], M. Fassan[1,2]

[1] Department of Medicine (DIMED), Surgical Pathology & Cytopathology Unit, University of Padua, Padua, Italy; [2] Istituto Oncologico Veneto - IOV-IRCCS, Padua, Italy; [3] Department of Surgery, Oncology and Gastroenterology, University of Padua, Padua, Italy; [4] Department of Surgical Science and Integrated Diagnostics (DISC), University of Genova, Genova 5 Department of Pathology and Diagnostics, University and Hospital Trust of Verona, Verona, Italy

Objectives: Gastric adenocarcinoma has recently been classified into several subtypes on the basis of molecular profiling, which has been successfully reproduced by immunohistochemistry (IHC) and in situ hybridization (ISH). The aim of this study is to classify gastric and gastroesophageal dysplasia according to TCGA/ACRG classes.
Materials and methods: A series of 73 gastroesophageal dysplastic lesions (37 gastric dysplasia and 36 gastroesophageal [GEJ] junction dysplasia; 44 low-grade dysplasia and 29 high-grade dysplasia) was investigated for mismatch repair proteins status, E-Cadherin, p53 and EBER expression, to reproduce The Cancer Genome Atlas (TCGA) and Asian Cancer Research Group (ACRG) molecular clustering.
Results: Overall, the dysplastic lesions were classified as follows: according to TCGA classification, EBV 0/73 (0%), MSI, 6/73 (8.2%), GS, 4/73 (5.5%), CIN 63/73 (86.3%); according to ACRG molecular subtyping, MSI 6/73 (8.2%), MSS/EMT, 4/73 (5.5%), MSS/TP53-, 33/73 (45.2%), MSS/TP53+, 30/73 (41.1%). A positive association was found between MSS/TP53- and GEJ dysplasia ($p = 0.0004$), between MSS/TP53+ and LG dysplasia ($p = 0.001$) and between MSS/TP53+ and gastric dysplasia ($p = 0.0018$).
Conclusions: Gastroesophageal dysplastic lesions proved to be heterogenous in terms of TCGA/ACRG classes, but with a different distribution from that of cancers, with no EBV positive cases, an increasing presence of mismatch repair deficiency from low grade to high grade lesions, and a prevalence of p53 aberrations in GEJ dysplasia. The present study also demonstrated that a routine-feasible approach based on IHC/ISH classification can be applied to the wide spectrum of gastroesophageal precursor lesions.

ID 771
THE IMMUNE LANDSCAPE OF SMALL BOWEL ADENOCARCINOMAS REVEALS A SITE-OF-ORIGIN-DEPENDENT HETEROGENEITY

Luca Dal Santo[1,2], Valentina Angerilli[1], Edoardo Maffi[1], Valerio Pellegrini[1], Roberta Salmaso[1], Giada Munari[3], Gianmaria Pennelli[1], Alessandro Vanoli[4], Claudio Luchini[5], Matteo Fassan[1,3]

[1] Department of Medicine, University of Padua, Padua, Italy; [2] Department of Pathology, Angelo Hospital, Mestre, Italy; [3] Veneto Institute of Oncology IOV-IRCCS, Padua, Italy; [4] Department of Molecular Medicine, University of Pavia; Anatomic Pathology, Fondazione IRCCS San Matteo Hospital; [5] Department of Diagnostics and Public Health, University and Hospital Trust of Verona, Verona, Italy; ARC-Net Research Center, University and Hospital Trust of Verona, Verona, Italy

Objectives: Small bowel adenocarcinomas (SBAs) are frequently grouped under the same therapeutic umbrella despite their anatomically distinct microenvironment. Since immune checkpoint blockade emerged as a promising approach, information on MMR status, tumor immune infiltration and PD-L1 expression is critical to identify patient subgroups

potentially suitable for checkpoint inhibitors.
Materials and methods: Intraepithelial and stromal tumor infiltrating lymphocytes (TILs) were evaluated in 153 resected SBAs and compared to clinicopathological variables, MMR status and PD-L1 expression. Based on tumor location, 84 (54.9%) ampullary adenocarcinomas, 24 (15.7%) duodenal non-ampullary adenocarcinomas, 16 (10.4%) jejunal adenocarcinomas and 29 (19.0%) ileal adenocarcinomas were included in the study.
Results: MMRp prevalence was statistically higher in ampullary than in other SBAs. Intraepithelial CD3+, CD4+, CD8+, CD20+, and FOXP3+ TILs density and PD-L1 expression were significantly higher in MMRd compared to MMRp SBAs. The prevalence of high PD-L1 expression (CPS ≥1) and high Intraepithelial CD3+ TILs density (> 2/HPF) showed an anatomical distribution along the small bowel and allowed to classify SBAs into four phenotypes: PD-L1 high/TILs high, PD-L1 high/TILs low, PD-L1 low/TILs low and PD-L1 low/TILs high. The prevalence of the PD-L1 high/TILs highphenotype was significantly higher in duodenal non-ampullary (66.7%) and jejunal-ileal tumors (66.7%) compared to the ampullary adenocarcinomas sub-group (39.3%).
Conclusions: The heterogeneous SBAs' immune landscape reported in our study suggests that highly variable patterns of response to immunotherapy may be encountered in the clinical setting for these malignancies.

ID 804
LIVER DISEASE PROGRESSION IN HISTOLOGICALLY DIAGNOSED NASH: A 14-YEAR FOLLOW-UP COHORT STUDY

De Bartolo D[1], Alessandra Mangia[2], Samantha Sarcognato[1], Diana Sacchi[1], Valeria Piazzolla[2], Stefano Brignola[1], Maffeis V[1], Ivana Cataldo[1], Vito Attino[3], Maria Guido[1,4]

[1] Pathology Department AULSS2, Treviso Hospital, Treviso, Italy; [2] Liver Unit, IRCCS "Casa Sollievo della Sofferenza", San Giovanni Rotondo, Italy; [3] Pathology Department, IRCCS "Casa Sollievo della Sofferenza", San Giovanni Rotondo, Italy; [4] Department of Medicine DIMED, University of Padova, Padova, Italy.

Objectives: Nonalcoholic steatohepatitis (NASH) causes severe liver fibrosis and cirrhosis. Only a few studies assessed fibrosis progression by histology and evaluated related predictors in untreated patients with a histological diagnosis of NASH. This study aims to investigate the proportion of patients with NASH who progress to an advanced stage of liver disease and to identify predictors of disease progression after a long follow up.
Materials and Methods: For 14 (9-19) years, we followed up a cohort of Italian patients with histological diagnosis of NASH. Disease progression was defined as evidence of cirrhosis in patients without cirrhosis at baseline, evidence of de novo occurrence of cirrhosis complications, histological established worsening of 1 stage of fibrosis or increase of liver stiffness by transient elastography (TE) in patients rejecting a second liver biopsy. Variables were compared in patients with or without evidence of progression.
Results: A total of 74 patients with histological diagnosis of NASH not related to concomitant medications were enrolled. Of them, 10 were lost to follow up and 5 excluded for different reasons. Overall, 59 patients, 21 of whom received a second liver biopsy, completed their follow up. Liver disease progression was observed in 28.8%. In the 21 patients with paired liver biopsy, fibrosis progressed in 28.5%. Diabetes and severe portal inflammation were key predictors for fibrosis progression (OR = 1.09, 95% CI 1.03-1.15, p = 0.004 and OR = 58.8, 95% CI 1.47-225.3, p = 0.03, respectively).
Conclusions: Our results reinforce the evidence that NASH progresses in about 30% of patients in the absence of pharmacologic treatment. In well-characterized patients with risk factors and persistently increased liver enzymes levels, liver biopsy retains a prognostic value, as severe portal inflammation is able to predict disease progression.

ID 808
HEPATOCELLULAR CARCINOMA EXPRESSING AFP WITHOUT ELEVATED SERUM LEVEL: DEFINITION OFA NOVEL SUBSET

C. Rossi[1], V. Ravetta[2], L.A. Stivala[1], S. Rossi[2], M. Paulli[1,2], A. Vanoli[1,2]

[1] Department of Molecular Medicine, University of Pavia, Pavia, Italy; [2] IRCCS San Matteo Hospital Foundation, Pavia, Italy

Objectives: Alpha-fetoprotein (AFP) is an oncofetal protein expressed by germinal and non-germinal tumors. In hepatocellular carcinomas (HCCs) AFP serum elevation is associated with adverse features, such as high histologic grade, proliferation molecular class, and prognosis. Clinico-pathologic and molecular data regarding HCCs with positive immunohistochemical staining for AFP in absence of a prognostically significant serum elevation (iAFP-HCCs) are lacking.
Materials and methods: Patients with a diagnosis of very early or early HCC (BCLC 0-1) who underwent surgical resection or ablation therapy (with pre-treatment biopsy) for iAFP-HCCs were compared to those with HCCs with serum elevation (≥400ng/ml) of AFP (sAFP-HCCs) for clinico-pathologic and immunohistochemical features and relapse-free survival (RFS). Previously treated HCCs were excluded. Immunohistochemistry for AFP, Ki67, and glutamine synthetase (GS), a transcriptional target of ß-catenin, was performed.

Results: Fourteen iAFP-HCCs were compared to eleven sAFP-HCCs. No significant difference was found in terms of patient age and gender, HCC etiology, as well as tumor size, multifocality, histologic grade, mitotic count, vascular invasion, macrotrabecular pattern and Ki67 proliferation index. Diffuse GS immunoreactivity was more frequently found in iAFP-HCCs (58%) in comparison with sAFP-HCCs (22%) although it still failed to reach statistical significance ($p = 0.184$). RFS did not show statistical significant difference between the two groups, with a median RFS of 9.5 months for sAFP-HCCs and of 18.7 months for iAFP-HCCs ($p = 0.702$).
Conclusions: From our preliminary results, iAFP-HCCs seem to share several characteristics with clinically and histologically aggressive sAFP-HCCs, although molecularly they might be a more heterogeneous class of tumors. A direct comparison of iAFP-HCCs with HCCs completely negative for AFP will be needed to a complete characterization of this tumor subset. AFP immunohistochemical staining might integrate radiology and biochemistry in the risk stratification of HCC patients without AFP serum elevation.

ID 811
ACINAR CYSTIC TRANSFORMATION OF THE PANCREAS: HISTO-MOLECULAR ANALYSIS FOR UNRAVELLING ITS NATURE

P. Mattiolo[1], O. Basturk[2], A. Mafficini[1], O. Kerem[2], R. T. Lawlor[1], S. Hong[3], L. A. Brosens[4], V.Adsay[5], A. Scarpa[1], C. Luchini[1]

[1] Department of Diagnostics and Public Health, Section of Pathology, University and Hospital Trust of Verona, Verona, Italy; [2] Department of Pathology, Memorial Sloan Kettering Cancer Center, New York, USA; [3] Department of Pathology, Asan Medical Center, Seoul, South Korea; [4] Department of Pathology, University Medical Center Utrecht, Utrecht,The Netherlands; [5] Department of Pathology, Koç University Hospital, Istanbul,Turkey

Background & objective
Acinar cystic transformation (ACT) of the pancreas is a poorly understood and rare entity among pancreatic cystic lesions. This study aims at clarifying their real nature.
Methods
The study cohort includes 25 pancreatic ACT, representing the largest series of ACT in the literature. Here we provide their clinicopathological characterization along with molecular profiling by next-generation sequencing (NGS).
Results: ACT were more common in female patients, and arose more frequently in body-tail region, with a mean size of about 4 cm. At the best of follow-up, all patients were alive and free of disease. Histologically, all cysts were lined by a typical acinar epithelium, sometimes intermingled with columnar or ductal-like epithelium. Cell atypia, necrosis, mitoses, and associated invasive carcinoma were absent in all cases. Three cases showed a patchy distribution in the pancreatic gland, and two cases were associated to ductuloinsular complexes with centroacinar microcysts. Two ACT showed very peculiar histologic features: one showed a distinctive microcystic pattern, and another harbored foci of low-grade dysplasia in the areas lined by ductal-like epithelium. NGS detected the presence of two pathogenic / likely-pathogenic mutations in two different cases: *KRAS*, c.34G > C, p.G12R, and *SMO*, c.1685G > A, p.R562Q.
Conclusion
Overall considered, our findings indicate that ACT is as a heterogeneous entity. It seems to encompass lesions with different possible pathogenesis, which includes the evolution from a centroacinarmicrocyst, and malformative, obstructive, or neoplastic origins. The potential presence of driver mutations call for a careful management of ACT patients, taking into account also surgical resection and active imaging surveillance / life-long follow-up.

ID 818
IMPACT OF HISTOCHEMICAL STAINS ON THE ASSESSMENT OF FIBROSIS IN CHRONIC LIVER DISEASES

Crudele GDL[1], De Bartolo D[2], Sarcognato S[2], Sacchi D[2], Baciorri F[2], Cataldo I[2], Guido M1,[1,2]
[1] Department of Medicine-DIMED, University of Padova, Padova, Italy; [2] Pathology Department AULSS2, Treviso Hospital, Treviso, Italy

Objectives: In chronic liver diseases, the stage of fibrosis is the main factor defining patient's prognosis and therapeutic options. Different histochemical stains are available to assess fibrosis in liver biopsy. The aim of this study was to investigate whether the use of different histochemical staining affects the histological evaluation of liver fibrosis.
Material and methods: Studied cases consisted of 40 liver biopsies: 20 chronic viral hepatitis and 20 non-alcoholic steatohepatitis matched for sex, age, and stage of the disease. In each case, 4 consecutive 3 µm-thick sections were cut and stained with Van Gieson, Masson's trichrome, and sirius red picrate. The first and last sections were stained using the same method (Van Gieson), to exclude that any difference in the evaluation of fibrosis was due to the different level of depth in the tissue. Fibrosis staging was performed by using validated scores. The diagnostic agreement among different section thicknesses and different staining methods was assessed by calculating the Fleiss' kappa.
Results: No disagreement was observed in staging chronic viral hepatitis, among different stainings (Fleiss' $\kappa = 1$ in both groups). However, in sub-staging stage 1-NASH cases, we found 3 discrepancies, with a Fleiss' $\kappa = 0.88$ (almost perfect agreement).

In evaluating sinusoidal fibrosis, we observed 12 discordant cases among different staining techniques, with a Fleiss' κ = 0.47 (moderate agreement), both in chronic viral hepatitis and NASH patients. In both groups, the discordant cases were characterized by more severe fibrosis detected with the siriusred staining (7 chronic viral hepatitis and 5 NASH).
Conclusions: This study demonstrates that no significant discrepancies exist in staging chronic liver diseases by using different staining methods. The frequently observed overestimation of liver fibrosis by the sirius red stain might be due to its ability to bind different types of collagen fibers, increasing accuracy and diagnostic performance in early stages of NASH.

Mercoledì 12 Ottobre 2022

Sala C 13.45 - 14.55

PATOLOGIA GINECOLOGICA 1

Moderatore: G. F. Zannoni

ID 746
IS MORPHOLOGY ALWAYS ENOUGH? HPV GENOTYPING, VIRAL LOAD AND INTEGRATION STATUS IN CERVICAL BIOPSIES

S. Varesano[1], M. Paudice[2], V. Parrella[2], A. Pulliero[3], A. Izzotti[3], M. Frascio[2], G. Icardi[1-3], V.G. Vellone[2]

[1] Hygiene Unit, IRCCS Ospedale Policlinico San Martino, Genoa, Italy; [2] Department of Surgical Sciences and Integrated Diagnostics (DISC), Genoa, Italy; [3] Department of Health Sciences (DISSAL), Genoa, Italy

Objectives: Human papillomavirus (HPV), the first cause of cervical cancer, is still a relevant health problem [1]. Pap test and HPV DNA test are the *first level* tests, but false negatives and falsepositives results are not negligible [2].
We propose a panel of molecular markers, including: genotyping, viral load determination and viral integration status assessment on FFPE cervical specimens.
Materials and methods: We analyzed 53 women with a previous abnormal result by Pap test and/or HPV DNA test. Each FFPE sample (biopsy, conization or hysterectomy) was IHC analyzed for p16 and ki67; while genotyping, coinfections and viral load through multiplex RT-PCR, simultaneously detecting 28 distinct HPVs (19 HR-HPV and 9 LR-HPV).
Results: We identified 23/53 CIN1 of which, 12/23 (52%) resulted negative for HPV; 10 CIN1 were co-infected with multiple genotypes and with high viral loads. In addition, we identified 22 CIN2-CIN3 lesions, 1 squamous carcinoma, 2 condylomas and 5 negative cases, all confirmed by molecular analysis.
Conclusions: Morphological analysis alone may overestimate the diagnosis of CIN1 [3], with repercussions in patient treatment. Molecular analyses may allow to detect co-infected and viral load-high low-grade lesions, improving diagnosis accuracy and allowing a personalized treatment protocol.

References

1. Sammarco M.L., Tamburro M., Pulliero A. et al. Human Papillomavirus Infections, Cervical Cancer and MicroRNAs: An Overview and Implications for Public Health. Microrna. 2020. https://doi.org/10.2174/2211536608666191026115045
2. Katki H.A., Schiffman M., Castle P.E. et al. Benchmarking CIN 3+ risk as the basis for incorporating HPV and Pap cotesting into cervical screening and management guidelines. J Low Genit Tract Dis. 2013.https://doi.org/10.1097/LGT.0b013e318285423c
3. Dalla Palma P., Rossi P., Collina G. et al. The Reproducibility of CIN Diagnoses Among Different Pathologists: Data From Histology Reviews From a Multicenter Randomized Study. Am J Clin Pathol. 2009. https://doi.org/10.1309/ajcpbrk7d1yiuwfp

ID 754
PULMONARY BLASTOMA ARISING WITHIN OVARIAN TERATOMA: THE FIRST CASE OF THIS ASSOCIATION AND FOLLOW-UP

[1]S. Sfregola, [1]F. Vegni, [1]Q. Zhang, [1]F. Policardo, [1]G. Scaglione, [1]A. Carlino, [1]F. Inzani, [1]A. Santoro, [1]A. Mulè, [1]G.F. Zannoni

[1]Unit of General Pathological Anatomy, Department for Women's Health Sciences, Children and Public Health, Fondazione Policlinico Universitario "Agostino Gemelli" IRCCS, Largo A. Gemelli 8, 00168, Rome, Italy.

Objectives: Malignant transformation of an ovarian teratoma is a rare event that can potentially occur in any of the component tissues, with squamous carcinoma being the most common reported form. Pulmonay blastoma (PB) is a rare and aggressive malignant neoplasm of the lung characterized by a mixture of immature epithelial and mesenchymal cells resembling fetal lung tissue. To the best of our knowledge, PB has never been reported to arise within ovarian teratomas; sporadic cases have been reported as metastases to the ovaries.
Materials and methods: a 47-year-old woman was referred to our institution for chronic pelvic pain and abdominal tension. Macroscopic examination revealed the presence of a large unilateralbulky solid-cystic right ovarian mass. Histologically, samples showed mature tissues along with solid areas consisting of immature neuroepithelial tissue and various mesenchymal cells. The cystic component was lined by secretory columnar cells with interposed embryonal type mesenchymal tissue resembling fetal-type lung.
Results: by IHC, the epithelial component showed positive staining for Ck7 and negative staining for PAX8, ER, PR, TTF-1, Vimentin, Ck20, S100 and

GFAP; the mesenchymal one showed positive partial staining for ASMA, Desmin and
Myoglobin. Both components exhibited focal aberrant nuclear/cytoplasmic β-catenin positivity. Direct sequencing for hot-spot mutational regions of *DICER1* gene showed no somatic mutations.
Conclusion: after excluding a metastatic origin from a primary lung neoplasm (negative thoracic PET/CT scan) or an endometrioid well differentiated adenocarcinoma with secretory features (negativity of PAX8, ER,PR), our final histopathological report was a pulmonary blastoma arising in ovarian immature teratoma, G1, Stage I. This neoplasm showed a favourable clinical outcome; after 11 years of follow-up and lack of adjuvant therapy, the patient is still in good general conditions, without evidence of recurrences or distant metastases.

ID 757
SYNCHRONOUS ENDOMETRIAL AND OVARIAN CANCERS: A COMPREHENSIVE ANALYSIS OF A MONO-INSTITUTIONAL SERIES

M. Moro[1], A. Guerriero[1], G. Munari[2], V. Angerilli[1], L. Alessandrini[1], O. Isidori[1], M. Balistreri[1], E. Maffii[1], M. Fassan[1,2], A.P. Dei Tos[1,2]

[1] Department of Pathology, Azienda Ospedale Università Padova, Padua, Italy; [2] Department of Medicine, University of Padua School of Medicine, Padua, Italy

Objectives: The diagnosis of synchronous carcinomas involving both the endometrium and ovaries is not a rare finding in gynecological pathology (up to 10% of cases) and represents a challenge for pathologists with implications on tumor staging and therapeutic decision-making.
Materials and methods: A mono-institutional series of 34 paired synchronous endometrial and ovarian carcinomas was reviewed by two expert pathologists on the basis of previously published histopathologic criteria. The series was investigated for MMR, p53 and *POLE* status and was subject to NGS targeting 67 cancer-related genes (Variantplex; ArcherDX).
Results: Out of 17 pairs, 15 had the same histotype (9 endometrioid, 4 serous high-grade, 2 clear cell); two pairs had discordant histotypes. By using histologic criteria, 5 (29.4%) cases were defined as independent synchronous tumors (3 pairs of endometriod carcinomas and 2 pairs with discordant histotype); 6 (35.3%) were defined as primary tumor with synchronous metastasis (1 pair of endometrioid carcinomas; 4 pairs of serous high-grade carcinomas and 1 pair of clear cell carcinoma); 6 (35.3%) were regarded as indeterminate (5 pairs of endometrioid carcinomas and 1 pair of clear cell carcinoma). The series was reclassified on the basis of TCGA classification and NGS targeted sequencing. A clonal relationship was determined in 7 (41.2%) cases, while 10 (58.8%) cases were molecularly heterogenous. Out of the 6 undetermined cases, 4 were reclassified as synchronous tumors and 2 as primary with metastasis; out the 5 synchronous tumors, one case was reclassified as primary with metastasis; out the 6 primary tumors with metastasis, 2 were reclassified as synchronous tumors.
Conclusions: Histologic criteria may not be sufficient to determine the existence of a clonal relationship between synchronous gynecologic cancers; especially for cases with endometrioid histology. Molecular integration is necessary to stage these lesions correctly, ensuring the best treatment and survival outcome for the patient.

ID 763
ADENOCARCINOMA OF THE UTERINE CERVIX: WHO 2020 HISTOPATHOLOGICAL CLASSIFICATION REPRODUCIBILITY AND VALIDATION BY HPV-DNA MOLECULAR ASSAY: A SINGLE-CENTER RETROSPECTIVE STUDY.

R.D'Amato Pascarella[1,3]; A.Visioli[2]; T.Dell'Anna[2]; L.Cimetti[1]; A. Pellegrino[2]; M.Piatti[1]; A.Vanzati[1]; E.Dainese[1]; C.Riva[1].

[1] Unit of Pathology, Department of Oncology, Manzoni Hospital, Lecco, Italy; [2] Department of Gynaecology, Manzoni Hospital, Lecco, Italy; [3] Department of Pathology, University Hospital of Insubria, Varese, Italy.

Objectives: A series of endocervical adenocarcinomas (EAs) was studied to assess WHO 2020 histopathological classification reproducibility and reliability of International Cervical Cancer Classification System (IECC) 2018 in distinguishing, by morphology alone, two main categories,HPV-associated (HPVA) and HPV-independent (NHPVA) adenocarcinoma. In order to validatefindings HPV-DNA and p16 were performed. Clinical features were evaluated to confirm different biological behaviour.
Materials and methods: 46 EAs observed at Manzoni Hospital Lecco in ten years (2010-20) were retrieved from Gynaecology and Pathology archives. Morphology was studied according to IECC criteria: grade, MI/Ki-67, pattern (tubular, papillary, villoglandular and clear cell), in situ or SIL component, LVSI and Silva Pattern. p16 was scored as: block-type/patchy/negative. Stage and follow-up data were recorded. HPV-DNA molecular assay was performed by RT PCR using EasyPGX qPCR instrument 96 with the EasyPGX ready HPV kit for HR genotypes.
Results: 30 out of 46 cases were classified HPVA, 6 cases in situ HPVA and 9 cases NHPVA (HPVA 80% vs NHPVA 20%); 1 case was reviewed as cervical involvement of endometrial carcinoma. HPV-DNA confirmed the diagnoses. p16 showed block-type stain in all HPVA, but also in 5/9 NHPVA.HPVA showed mostly "usual type"(90 %) Silva pattern A/B in 71%; in all NHPVA high grade and pattern C were observed. LVSI prevailed in NHPVA (57% vs 27%). NHPVA

cases showed older age (53 vs 41.5 ys), more advanced stages (II/III vs I/II) and worse prognosis (recurrences 11.2% vs 5.1%; deaths 33.3% vs 8.3%).
Conclusions: The IECC system reliability is confirmed and morphology alone is effective to distinguish HPVAand NHPVA. Useful features in predicting HPVA are high Ki-67, villoglandular component and in situ/SIL component; on the contrary, patchy p16, high grade and LVSI, along with solid, papillary and clear cell growth suggest NHPVA. p16 expression *per se* is not specific for HPV involvement.NHPVA and Silva pattern C are predictors of worse outcome.

ID 766
T-LYMPHOBLASTIC LEUKEMIA/LYMPHOMA PRESENTING AS OVARIAN MASS

P.Antonini[1,] M.G. Mastrosimini[1-2,] M. Ceccaroni[3,] C. Zorzi[3,] G.Zamboni[1-2,] A. Pesci[2.]

[1] Department of Pathology University of Verona, Verona; [2] Unit of Anatomic Pathology, IRCCS Ospedale Sacro Cuore-Don Calabria, Negrar di Valpolicella, Verona, Italy; [3] Gynecology IRCCS Ospedale Sacro Cuore-Don Calabria, Negrar di Valpolicella, Verona, Italy

Objectives: Lymphomas primarily involving the female genital tract are very rare[1] and may be either localized or part of a systemic disease[2]. Due to their rarity, a carcinoma is always considered clinically. We describe a case of T-lymphoblastic leukemia/lymphoma presenting as an ovarian mass in a 61-year-old woman and discuss the main differential diagnoses.
Materials and Methods: Patient underwent bilateral salpingo-oophorectomy. Both ovaries were fleshy, whitish, and increased in size (12 and 4.5 cm respectively). Tubes were apparently normal. On microscopic examination, the neoplastic cells were small to medium-sized with convoluted nuclei, small nucleoli, and scant cytoplasm. Apoptotic bodies and mitotic activities were easily identified. The neoplastic cells were arranged in a diffuse pattern with occasionally a sex-cord-like trabeculae and were embedded in a sclerotic stroma. Tubes were diffusely involved. The main differential diagnoses were: undifferentiated carcinoma with sex-cord pattern, sex-cord/stromal tumor/Sertoli/adult granulosa cells tumor, neuroendocrine carcinoma, small cell carcinoma-hypercalcemic type, undifferentiated carcinoma, primary or metastatic, high grade lymphoma.
Results: The neoplastic cells were positive for T-cell markers (CD7, CD3, CD5), for TdT, CD10 and CD99, with a Ki67 index > 80%. The final diagnosis was T-lymphoblastic leukemia/lymphoma.
Conclusions: we described a rare case of a systemic T-lymphoblastic leukemia/lymphoma presenting as a pelvic mass and discuss the main differential diagnoses.
1. Vang R, Medeiros LJ, Fuller GN, et al. Non-Hodgkin's lymphoma involving the gynecologic tract: a review of 88 cases. Adv Anat Pathol 2001;8(4):200-17. https://doi.org/10.1097/00125480-200107000-00002
2. Kosari F, Daneshbod Y, Parwaresch R et al. Lymphomas of the female genital tract: a study of 186 cases and review of the literature. Am J Surg Pathol 2005;29(11):1512-1520.https://doi.org/10.1097/01.pas.0000178089.77018.a9

ID 795
INDETERMINATE GRADE SEROUS OVARIAN CARCINOMA: A CASE SERIES OF A RARE OVARIAN TUMOUR

R. Ciudino[1], A.G. Corradini[1], F. Rosini[1], T. Maloberti[1], D. de Biase[1], G. Tallini[1], D. Santini[1], A. De Leo[1]

[1] Pathology Unit, University of Bologna Medical Center, Bologna, Italy

Objectives: Zarei et al. described a heterogeneous group of ovarian cancers characterised by mixed morphology and intermediate pathological features of high-grade and low-grade serous carcinomas-named as Indeterminate-Grade Serous CArcinomas (IGSCAs). The present study aims to describe the morphological and immunohistochemical (IHC) characteristics of a consecutive series ofIGSCAs and to investigate the molecular profile including the Homologous Recombination Deficiency (HRD) status.
Materials and Methods: We analyzed 7 cases of IGSCAs among over 170 consecutive serous ovarian cancers diagnosed at our Institution between 2019 and 2021. IHC analysis and molecular characterization by custom-designed Next-Generation sequencing panel were performed. HRD status was assessed by using myChoice® HRD Plus assay and was considered positive if deleterious or suspected deleterious mutations in *BRCA1* or *BRCA2* were detected and/or Genomic Instability Score (GIS) was positive (GIS ≥ 42).
Results: In our series, we identified 148 (87%) High-Grade Serous Carcinomas (HGSC), 17 (9%) Low Grade Serous Carcinomas (LGSC), 7 (4%) IGSCAs. The median age at diagnosis was 61years (range 42 - 74). IHC analysis showed p53 mutation-type in 5 cases and wild-type p53 immunostaining pattern in 1 case. In 1 case the expression of p53 was heterogeneous (wild-type and mutation-type). Clinically significant mutations in *BRCA1* or *BRCA2* were detected in 2 caseswhile *TP53* was mutated in all cases. Other mutations in *PIK3CA* and *NRAS* were identified in 2 cases. The GIS score was positive for 5 tumours, while HRD status was positive in 6 patients; the only negative case exhibited *NRAS* and *TP53* concomitant mutations.
Conclusions: Our study confirmed IGSCA as a rare subtype of serous ovarian tumourscharacterized by histopathological and molecular heterogeneity. The integrated morphological and molecular approach

could contribute to better define these rare elusive neoplasms providing anaccurate classification and a possible targeted therapy.

ID 837
NHERF1 PROTEIN LEVELS IN ANOGENITAL LESIONS INDUCED BY MUCOSAL HUMAN PAPILLOMAVIRUSES

C. Saponaro[1], T. Gheit[2], L. Galati[2], M. Milella[1], M. Zambetti[1], F.A Zito[1], R.A Cardone[3], SJ Reshkin[3] and M. Tommasino[1]

[1] IRCCS Istituto Tumori "Giovanni Paolo II" of Bari, Italy; [2] International Agency for Research on Cancer, World Health Organization, Lyon, France; [3] Department of Biosciences, Biotechnology and Biopharmaceutics, University of Bari, Italy

Objectives: The E6 oncoprotein from mucosal high-risk (HR) human papillomaviruses (HPV) exerts its functions by interacting with several cellular proteins, including the PSD-90/Dlg/ZO-1 homology (PDZ) family proteins[1]. Some studies indicated that the ability to inactivate the PDZ protein, the Na+/H+ exchanger regulatory factor-1 (NHERF-1), is not shared among all HR HPV types[2], while others reported that NHERF-1 is similarly targeted by all mucosal HPV types[3].
Methods: HPV genotyping was performed by Luminex beads-based assay in different type of genital lesions, namely (i) benign anogenital warts (n = 8) (ii) premalignant lesions (L-SIL and H-SIL) (n = 43), (iii) invasive cervical squamous cell carcinomas (SCC) (n = 17). NHERF-1 protein levels was determined by immunohistochemistry.
Results: Decrease of NHERF-1 protein level was not observed in genital warts positive for LR HPV types in comparison to healthy epithelium. NHERF-1 protein levels were decreased mainly in HPV16-positive pre-malignant and malignant lesions, while the other HR HPV types appeared to have a marginal effect. In HPV16-positive SIL NHERF-1 protein levels correlate with the severity of cervical lesion, being lower in low grade than high-grade pre-malignant lesions.
Conclusions: These findings report that mucosal HR and LR HPV types differently impact on NHERF-1 protein level. Thus, NHERF-1 expression levels in pre-malignant or malignant cervical lesion could be used as additional surrogate marker offering the possibility to improve the specificity of diagnosis.

References

1.Thomas M, Myers MP, Massimi P, et al. PLoS Pathog. 2016;12(8):e1005766. http://dx.doi.org/10.1371/journal.ppat.1005766

2.Accardi R, Rubino R, Scalise M, et al. J Virol. 2011;85(16):8208-16. http://dx.doi.org/10.1128/JVI.00114-11

3.Drews CM, Case S, Vande Pol SB. PLoS Pathog. 2019;15(4):e1007575. http://dx.doi.org/10.1371/journal.ppat.1007575

Mercoledì 12 Ottobre 2022

Sala D 13.45 - 14.55

PATOLOGIA MOLECOLARE 1

Moderatore: F. Pagni

ID752
UVEAL MELANOMA: IMMUNOHISTOCHEMICAL AND MOLECULAR CHARACTERIZATION OF A MONO-INSTITUTIONAL SERIES

G. Zarrilli[1], G. Munari[2], M. Perilli[1], V. Angerilli[1], M. Balistreri[1], M. Sbaraglia[1], E. Rebellato[2], M. Sabbadin[2], M. Fassan[1], A. P. Dei Tos[1]

[1]Dipartimento di Medicina (DIMED), Unità di Anatomia Patologica, Università degli Studi di Padova, 35121 Padova, Italia; [2]Veneto Institute of Oncology IOV - IRCCS, Padova, Italia.

Uveal melanoma (UM) is a malignant melanocytic neoplasm arising in the choroid, ciliary body or iris, and it is the most common primitive ocular neoplasm in adults. From a molecular standpoint, about 90% of UM are characterized by two, mutually exclusive, main driver mutations in the*GNA11* or *GNAQ* genes. However, a comprehensive molecular characterization of this disease is still lacking.
A mono-Institutional series of 84 cases of UM (2010-2021) was considered and categorized according to their anatomical location and to their histotype, namely, spindle-cell (46 cases, ≈55%), epithelioid (13 cases, ≈15%) or mixed (25 cases, ≈30%). Cases were further characterized by IHC for p53, BAP1, CD3, CD8, PD-L1, MMR proteins and NTRK and by targeted multigene NGS with the analysis of 67 cancer-related genes (Variantplex; ArcherDX).
No NTRK positivity or MMR deficiency were observed. Clonal-pattern or complete loss of p53 was significantly more frequent in spindle-cell UMs (≈30%) than in epithelioid or mixed UMs (≈8%). An opposite pattern was observed for BAP1, with a higher frequency of loss of expression in epithelioid UMs (≈66%) when compared with mixed or spindle cell cases (≈39%).
In the series, ≈85% of our cases harbor mutually exclusive mutations in either *GNA11* or *GNAQ* genes. The third most frequently altered gene resulted *NOTCH1* (≈23%). Other interesting results are linked to potentially targetable mutations and/or pathways. In particular, six cases (10%) showed *ERBB2* missense mutations. Furthermore, *PTEN* and *PIK3CA*

showed alterations in respectively 11 and 8 cases (≃18% and ≃13%) in a not mutually exclusive way. Of note, six cases (10%) showed ten or more altered genes; this "hyper-mutational" state didn't show any link with an increased inflammatory response (assessed through CD3 and CD8 immunostains), as seen in other neoplasms.
This study shows new data about UM from an immunohistochemical and a molecular standpoint; we achieved promising results concerning prognostic and predictive markers which were seldom, if ever, tested before.

ID 760
CHPv2 PANEL IN CLINICAL MOLECULAR PATHOLOGY: THE CLASSIFICATION OF VARIANTS AND ACTIONABILITY LEVELS IN COLORECTAL CANCER

E. Melucci[1], E. Gallo[1], C. Ercolani[1], F. Rollo[1], A. Di Benedetto[1], A. Palange[1], F. Bartoccini[1], B. Casini[1], S. Buglioni[1], E. Pescarmona[1]

[1] Pathology Unit, IRCCS Regina Elena National Cancer Institute, Rome, Italy

Objectives: Screening for Colorectal cancer (CRC) somatic mutations is key to not only understanding its molecular pathogenesis but also to providing patients with innovative genotype-matched therapies. In contrast to conventional one-gene-at a-time diagnostic techniques, NGS technologies can be used for the simultaneous and rapid identification of gene mutations.
Materials and methods: The CHPv2 multigene panel (ThermoFisher) is a targeted, multi-biomarker assay that detects the mutational profile of 50 genes. From 240 FFPE samples of CRC, DNA was extracted, amplified, and templated into amplicon-based libraries. Paired-end massively parallel sequencing of 150-bp fragments was performed with Ion GeneStudio S5 Prime instrument on a ThermoFisher platform. In the workflow analysis we used: 1_Torrent Suite, which plans, monitosr, and tracks the runs all within a web interface while reviewing the quality and accuracy of your sequencing run. 2_Ion Reporter, which calls and annotates SNPs/InDels. 3_Oncomine Reporter, which with Ion Torrent, obtains clear information that links biomarkers to relevant evidence.
Results: Data obtained with the CHPv2 panel was then used to search for genetic markers capable of identifying new levels of actionability. In fact, we classified the variants identified using oncoKB (Precision Oncology Knowledge Base). In particular, for actionable level 3B, we found that the genes are: KRAS, PIK3CA, ATM, PDGFRA, NRAS, AKT1, EGFR, IDH1, IDH2, ERBB2 (92 Level 3B mutations). For actionability level 4, the genes with this category of mutations are PTEN, EGFR, SMARCB1, BRAF and CDKN2A (14 Level 4 mutations). We focused our attention on a subgroup of 100 mCRC, wich have the mutated KRAS gene. From this subgroup, we excluded the 10 cases with the KRAS G12C mutation. This subgroup of patients, in normal clinical practice, are not eligible for molecular-targeted anti-EGFR therapies. Therefore, we found 30 mutations that have actionability level 3B and/or level 4 and involve the following genes: PIK3CA, ATM, AKT1, EGFR, IDH2, ERBB2, PTEN, and SMARCB1.
Conclusions: Our results suggest that a large panel of genes can detect mutations with 3B and 4 actionability levels, in accordance with OncoKB. By expanding the mutational target, it will be possible to guarantee patients possible future therapeutic approaches.

ID 762
"STRANGER THINGS": THE POTENTIAL PROVIDED BY THE WIDE RANGE OF MOLECULAR TESTS

C. Ercolani[1], E. Gallo[1], F. Rollo[1], A. Di Benedetto[1], A. Palange[1], F. Bartoccini[1], E. Pescarmona[1], B. Casini[1], S. Buglioni[1], E. Melucci[1]

[1] Pathology Unit, IRCCS Regina Elena National Cancer Institute, Rome, Italy

Objectives: Precision medicine needs an accurate bio-molecular assessment to ensure a precise classification of tumours. However, some rare cases, with apparently incongruous results, can only be clarified by further reflex tests.
Materials and methods: we selected 4 cases: an endometrial carcinoma (EC), a melanomametastasis (mM), a colon-rectal adenocarcinoma (CRC) and a non-small cell lung cancer (NSCLC). All cases presented incongruous results comparing 2 or more of the following methods: immunohistochemistry (IHC) for Mismatch Repair (MMR) proteins, PCR-based fragment sizing assay Promega specific for mononucleotide microsatellite loci: BAT25,BAT26,NR21,NR24,MONO27, real-time PCR (qPCR) Idylla™MSI Test for 7mononucleotide monomorphic microsatellite loci into the genes ACVR 2A,BTBD7,DIDO1,MRE11,RYR3,SEC31A,SULF2, Idylla™BRAF and EGFR Tests (Biocartis), Next-Generation Sequencing (NGS)(Focus and CHPV2 panels,Thermo).
Results: Case1: EC with a dMMR (deficient) proteins evaluated by IHC, showed a discordant MSS (Stable) microsatellite profile for all loci by Promega panel and a MSI-H (High Instability) profile in 4/7 loci by Idylla™MSI Test consistent with IHC Results: This is probably due to the different loci instability between CRC and EC that occur in some cases.
Case2: mM with *BRAF^{wt}* gene by qPCR and *two-BRAF^{mut}* in *cis* V600E+R603G by NGS. The failure to

detect the *BRAF*^{mut} V600E by qPCR could be due to the primer used, which probably binds the nucleotide sequence altered by the second *BRAF*^{mut}.
Case3: CRC with a pMMR (proficient) proteins evaluated by IHC that was contrasted with a MSI-H phenotype evaluated with Promega panel. This MSI-H tumour would have been missed by IHC alone. This is due to the presence of MMR proteins that are structurally/enzymatically defective but still have an intact epitope.
Case4: A patient with NSCLC *EGFR*^{del} exon19+T790M^{mut} by qPCR showed resistance to Osimertinib. The next NGS result was in agreement with the patient clinical outcome showing in addition to *EGFR*^{del}, two *EGFR*^{mut} in *cis* T790M+C797S.
Conclusions: In our experience, when apparently discordant results occur, it is desirable to haveavailable as many reflex tests as possible in the Pathology Units in order to ensure the most correct bio-molecular diagnosis.

ID 775
TECHNICAL EVALUATION OF TUMOR MUTATIONAL BURDEN MEASUREMENT ON CYTOLOGICAL SAMPLES

F. Pepe[1], P. Pisapia[1], V. Gristina[2], A. Iaccarino[1], G. Gragnano[1], G. Russo[1], C. De Luca[1], M. Nacchio[1], G. Troncone[1], U. Malapelle[1]

[1] Department of Public Health, University Federico II of Naples, Naples, Italy; [2] Section of Medical Oncology, Department of Surgical, Oncological and Oral Sciences, University of Palermo, Palermo, Italy.

Objectives: Currently, evaluation of the expression level of programmed death-ligand 1 (PD-L1) has proven highly successful as a positive predictive biomarker for Immune Checkpoint Inhibitors (ICIs). (1) In addition to PD-L1, other predictive biomarkers are under investigation, including high tumor mutational burden (TMB-H). However, measuring TMB-H is still considered an opening challenge due to the scant amount of tissue available from NSCLC patients. As regards, our aim was to evaluate the technical feasibility of analyzing TMB on Cell Block specimens (CBs).
Materials and methods: A total of n = 8 pairs of histological and CB samples from NSCLC patients were analyzed by using Oncomine Tumor Mutational Load Assay (TML) on Ion Torrent S5 GS next-generation sequencing (NGS) platform.
Results: Overall, 6/8 CBs (75.0%) were successfully analyzed. Similar results between histological matched CBs specimens were obtained in terms of median total reads (7207048.80 vs 7558817.80), median mapped reads (7075753.83 vs 7513822.00), median read lengths (115.50 vs. 113.00), median percentage of reads on-target (97.49% vs. 98.45%), median average reads per amplicon (454.67 vs 476.14), and median uniformity of amplicon coverage (83.52% vs 84.13%). (2)
Conclusions: In this pilot study, we demonstrated the technical feasibility of assessing TMB on CBs from diagnostic routine practice.

References

1. Reck M, Rodríguez-Abreu D, Robinson AG, et al. Pembrolizumab vs. chemotherapy for PD-L1-positive non-small-cell lung cancer. N Engl J Med. 2016; 375:1823-1833. https://doi.org/10.1056/NEJMoa1606774
2. Pepe F, Pisapia P, Gristina V, et al. Tumor mutational burden on cytological samples: A pilot study. Cancer Cytopathol. 2021; 129:460-467. https://doi.org/10.1002/cncy.22400.

ID 776
A VALIDATION OF A NOVEL CUSTOM DNA-BASED NGS PANEL FOR THYROID INDETERMINATE NODULES

M. Nacchio, E.Vigliar, U. Malapelle, P.Pisapia, F. Pepe, C. De Luca, A. Iaccarino, L. Palumbo, G.Russo, G. Troncone, C. Bellevicine

Department of Public Health, University Federico II of Naples, Naples, Italy

Objectives: the undetermined diagnostic categories still represent an opening challenge in thyroid nodules. In this scenario, next-generation sequencing (NGS) platforms play a pivotal role for the molecular testing of thyroid fine-needle aspiration (FNA) specimens but prohibitive cost and lack of reimbursement impacts on large diffusion in diagnostic practice. (1) As regards, we developed and validated a novel custom NGS pane, (*Nexthyro)* on a retrospective series of diagnostic routine cytological specimens for diagnostic purposes.
Materials and methods: A series of cell-line derived reference standard and n = 72 FNA archival samples previously tested with the RtqPCR approach was evaluated.
Results: *Nexthyro* panel highlighted 100% specificity and limit of detection (LoD) of 2%. Moreover, in 5 out of 72 cases (7%), it detected clinically relevant mutations in *BRAF* and *RAS* genes not previously identified with RtqPCR approach. (2)
Conclusions: The study revealed that *Nexthyro* is a reliable NGS panel for routine thyroid FNA allowing patients with undetermined thyroid nodules affordable access to NGS solution.

References

1. Roy- Chowdhuri S, Goswami RS, Chen H, et al. Factors affecting the success of next- generation sequencing in cytology specimens. Cancer Cytopathol 2015;123:659–68. https://doi.org/10.1002/cncy.21597
2. Sgariglia R, Nacchio M, Migliatico I, et al. Moving towards a local testing solution for undetermined thyroid fine-needle aspi-

rates: validation of a novel custom DNA-based NGS panel. J Clin Pathol. 2021:jclinpath-2021-207429. https://doi.org/10.1136/jclinpath-2021-207429.

ID 778
UTILITY OF OCA-PLUS NGS PANEL FOR THE IDENTIFICATION OF NOVEL DRUG-ACTIONABLE MARKERS INPATIENTS WITH LITTLE THERAPEUTIC CHANCE

B. Casini[1] and E. Gallo[1], R. Covello[1], F. Rollo[1], A. Di Benedetto[1], C. Ercolani[1], A. Palange[1,] E.Melucci[1], S. Buglioni[1], V. Ferraresi [2]

[1] Pathology Unit, [2] Sarcomas and Rare Tumors Unit

Objectives: An intriguing purpose of extended genomic analysis is to identify new therapeutic targets in patients that have exhausted therapeutic options or with histotypes known to be resistant to medically available labeled treatments. The purpose of this study is to perform a comprehensive genomic profiling with a panel of 500 NGS cancer-associated genes so as to detect, in addition to the mutation landscape, also the multiple gene biomarkers like Microsatellite Instability (MSI), Tumor Mutational Burden TMB and a genomic scare to evaluate HRD.
Materials and methods: we selected 11 samples of different neoplastic histotypes: 2 kidney carcinoma (KC) with primary tumor and metastasis, 1 NSCLC with primary and metastasis,1 melanoma, and 1 chondroblastic osteosarcoma (COS), 1 undifferentiated sarcoma, 1 intimal sarcoma of the heart and 1 endometrial cancer (EC). From the sample FFPE DNA and RNA wereextracted and was performed library preparation for NGS panel Oncomine™ Comprehensive Assay Plus (OcaPlus) Thermofisher, that is suitable for our purpose.
The sequencing data obtained were analyzed by using IR (Ion Reporter 5.18) and by analysis with specific workflows. We also performed the comparison of the MSI data obtained from the 76 microsatellite loci of the OcaPlus panel and the 5 loci of the Promega KIT.
Results: the molecular targets of therapeutic significance identified were: a pathogenic variant in PMS2 detected in one case of primary KC that can be considered for drug in trial specific for the DNA repair pathway. A pathogenic variant in KRAS was detected in one case of NSCLC both in primary and metastasis that predict response to target therapy and for drug clinical trial.
The cases of COS and melanoma showed high LOH 45.2% and 81.7% respectively suggesting the possibility of an off label PARP inhibitor therapy. The intimal sarcoma shows PIK3CA E542K that can be considered for drug clinical trial that target the PI3K/AKT/MTOR pathway and MDM4 amplification for MDMX inhibitor. Microsatellite status of EC obtained with OcaPlus and Promega KIT showed the same MSI-H result and a completely concordance we detected in the other samples that were all MSS.
Conclusions: our study although with preliminary data shows how the OcaPlus panel may allow us to identify possible therapeutic targets even if still included in clinical trials. Further studies with a larger number may allow to introduce the system in routine clinical practice.

Mercoledì 12 Ottobre 2022

Sala E 13.45 - 14.55

PATOLOGIA FETOPLACENTARE

Moderatore: L. Resta

ID 874
VIRTOPSY IN FETAL PATHOLOGY

V.Parrella[1], M.Paudice[1], M.Cosso[2], P.De Biasio[3], E.Fulcheri[4], VG.Vellone[5]

[1] Department of Integrated Surgical and Diagnostic Sciences (DISC), University of Genoa, Italy; [2] Department of Radiology, IRCCS Ospedale Policlinico San Martino, Genova, Italy; [3] Prenatal Diagnosis and Perinatal Medicine Unit, IRCCS Ospedale Policlinico San Martino, Genova, Italy;[4] Fetal, Perinatal & Gynecologic Pathology Center. IRCCS Istituto Giannina Gaslini, Genoa, Italy; [5] Anatomic Pathology University Unit, IRCCS Ospedale Policlinico San Martino, Genoa, Italy.

Objectives: Autopsy practice is declining worldwide; however, fetal autopsies remain a relevant activity because of their role in the registry of malformations, the identification of syndromic diseases and the planning of subsequent pregnancies.
Virtual Autopsy (Virtopsy) is a revolutionary technique in forensic medicine and anatomic pathology: it combines the conventional autopsy with the most modern imaging techniques and it can be useful in establishing skeletal age, gender identification, morphostructural features, presence of foreign bodies in the body, presence of trauma, and signs of previous surgery/orthopedic procedures.
Material and methods: A retrospective critical review for the period 2012-2021 of all fetal autopsies submitted to virtopsyat Policlinico San Martino Hospital. The conventional autopsy on the fetus was performed by macro and microscopic protocol, while virtopsy was conducted by multilayer CT scan.
Results: Eight cases were considered (6 cases of bone malformative pathology, 2 cases of other malformations) with a mean gestational age of 22 weeks. The following diseases were diagnosed: Jeune Syndrome, Sacral Agenesis, Cornelia De Lange or Amsterdam Dwarfism, Seckel Syndrome or Bird's

Head Dwarfism, Thanatophoric Dysplasia, Conjoined Twins, Omphalocele and Hydrocephalus
Conclusion: Virtopsy is an important supplement to fetal autopsy and has allowed better documentation of body alterations while conventional autopsy remains unavoidable for the detection of somatic and splanchnic alterations. Both methods may be complementary to direct genetic investigations of syndromic diseases and to verify diagnostic hypotheses formulated during prenatal imaging.

ID 876
LEFT VENTRICULAR NON-COMPACTION CARDIOMYOPATHY: A RARE CAUSE OF SUDDEN FETAL DEATH

G. D'Abbronzo[1], A. Ronchi[1], E. La Mantia[1], M. La Verde[2], A. Feola[3], N. De Stefano[4], R. Franco[1]

[1] Pathology Unit, Department of Mental and Physical Health and Preventive Medicine, University of Campania "Vanvitelli", Naples, Italy; [2] Gynecology Unit, Department of Woman, Child and General and Specialized Surgery, University of Campania "Vanvitelli", Naples, Italy; [3] Department of Experimental Medicine, University of Study of Campania "Vanvitelli", Naples, Italy; [4] Pathology Unit, "S.Giuseppe Moscati" Hospital, Avellino, Italy

Objectives: Left ventricular non-compaction cardiomyopathy (LVNC) is a rare form of congenital heart disease characterized by the arrest of cardiac morphogenesis in the last phase that causes the lack of compaction of cardiac muscle tissue[1]. Although LVNC can occur asymptomatic, rarely LVNC can cause sudden cardiac death during fetal or neonatal life[2]. We present a rare case of sudden cardiac death of a fetus at 38 weeks due to LVNC.
Materials and methods: A fetus died at 38 weeks. Pregnancy had passed without complications; however, no fetal heartbeat was found at the last prepartum checkup. Autopsy was performed.
Results: Anthropometric parameters were normal. Histologic examination of the heart showed two muscle layers with different organization. The external layer presented a normal tissue organization. The inner layer was formed by a prominent anastomosed muscular trabecular network, withnumerous irregular recesses, lined with endocardium, in communication with the ventricular lumen. This aspect affected more than 50% of the total thickness of both ventricles. A diagnosis of LVNC was rendered.
Conclusions: LNVC is a rare case of fetal sudden cardiac death that can only be diagnosed with a careful histo-morphological examination of the heart.

References

1) Jeffrey A Towbin, Angela Lorts, John Lynn Jefferies, Left ventricular non-compaction cardiomyopathy in Lancet, n. 386, 2015, pp. 813-25, Https://doi.org/10.1016/S0140-6736(14)61282-4.

2) Brescia ST, Rossano JW, Pignatelli R et al. Mortality and sudden death in pediatric left ventricular noncompaction in a tertiary referral center. Circulation. 2013 Jun 4;127(22):2202-8.HTTPS://DOI.ORG/10.1161/CIRCULATIONAHA.113.002511

Giovedì 13 Ottobre 2022

Sala Mantegna 08.30 - 09.20

UROPATOLOGIA 1

Moderatore: G. Martignoni

ID 761
IMPACT OF ROL SUBSTAGING ON PT1 HIGH-GRADE BLADDER CARCINOMA PROGRESSION: A CONFIRMATORY ANALYSIS

M. Valeri[1,3], R. Contieri[2,3], V. Fasulo[2,3], C. De Carlo[1,3], A. Bressan[1,3], L. Dore[1,3], M. Iuzzolino[1,3], R. Hurle[2,3], L.M. Terracciano[1,3], P. Colombo[1,3].

[1] Department of Pathology, IRCCS Humanitas Clinical and Research Hospital, Rozzano, Italy; [2] Department of Urology, IRCCS Humanitas Clinical and Research Hospital, Rozzano, Italy; [3] Humanitas University, Department of Biomedical Science, Pieve Emanuele, Italy

Objectives: The management of patients with pT1 high-grade (HG) urothelial carcinoma (UC) is still a challenging issue in urological practice, and depth and amount of lamina propria tumor invasion is a key prognostic variable. Compared to other substaging methods, the Rete Oncologica Lombarda (ROL) system for substagingpT1HGUC - simply based on a 1-mm threshold- showed high predictive value for progression after transurethral resections (TURBT) in retrospective studies. We aimed to validate ROL system on a prospective large mono-institutional series.
Materials and methods: Between 2013 and 2020, we collected clinico-pathological data of all patients who underwent TURBT with a diagnosis of pT1HGUC. Using a cut-off of 1 mm (or diameter of an objective 20x high-power field) we stratified tumors in ROL1 and ROL2, corresponding to one invasive focus or multiple foci extending together for < 1 mm and for > 1 mm, respectively.
Results
A total of 229 confirmed pT1HGUC were analyzed. Mean age was 73yrs, with male predominance (74.7%); 70 tumors showed multifocality (30.57%), 33 divergent differentiation (14.4%). Associated CIS and vascular invasion occurred in 14% and 9% of cases. ROL was feasible in all but one case (99.6%): 94 cases were ROL1 (41%) and 134 ROL2 (59%). At a median follow up of 23 months (IQR 12.33-38.5), 59 patients had recurrence (25.76%) and 37 progression (16%). ROL predicted progression in univariate

(OR = 3.58, 95% CI 1.50-8.56; p = 0.004) and multivariate (OR = 2.95, 95% CI 1.11-7.87; p = 0.03) Cox regression analysis. At Kaplan-Meier estimates, ROL showed correlation with progression (p < 0.01), but not with recurrence (p > 0.05).
Conclusions: Our results confirmed the strong predictive role of ROL system for progression in pT1HGUC on a large prospective series. We foster the application of ROL system for substaging T1HGUC, a simple and feasible method alternative to pT1a/b that might identify high-risk patients and drive urological decision-making.

ID 774
NEU-LIKE PHENOTYPE AND MOLECULAR PLASTICITY IN MUSCLE-INVASIVE BLADDER CANCER: A MONO-INSTITUTIONAL STUDY

A. Bressan[1,2], C. De Carlo[1,2], L. Dore[1,2], M. Iuzzolino[1,2], M. Valeri[1,2], S. Zanella[1,2], GM. Elefante[1,2], M. Cieri[1,2], L. M. Terracciano[1,2], P. Colombo[1,2]

[1] Department of Pathology, IRCCS Humanitas Clinical and Research Hospital, Rozzano, Italy; [2] Humanitas University, Department of Biomedical Science, Pieve Emanuele, Italy

Molecular subtyping studies have identified a subset of muscle-invasive urothelial carcinoma (MI UC) consistent with neuroendocrine (NE) differentiation without NE histology (Neu-like), representing a potentially high risk subgroup which may require a different therapy. We used an immuno-phenotypical score (Piescore) to discriminate Luminal from Basal from Neu-like carcinoma. Aim of this study was to identify Neu-like tumors in a cohort of pT2 UC patients correlating the results with outcome.
TUR specimens with pT2HG UC from 251 pts have been submitted for immunohistochemical analysis, using relevant markers for Basal type (CD44, CK5/6) and Luminal type (CK20 and pPARg). In MI component, Piescore divided Basal and Luminal types when markers were consistent with a specific phenotype; Neu-like if all markers were negative. Non muscle-invasive component (NMI) was also evaluated.
Overall, we identified no markers expression of 49/251 pts (19,5%), all consistent with NEU-like phenotype, with 7 cases showing neuroendocrine histology, and 42 pts with urothelial differentiation. Among these, 7/42 pts were negative both in NMI and MI compartment of the tumor, while the majority of cases (35/42)underwent a phenotypical switch to null phenotype from NMI to MI component: 26 pts from Luminal phenotype, 4 from Basal, and 5 from a Mixed type. With a median follow up of 216 months, Neu-like pts were affected by a worse prognosis compared with the other phenotypes, although not statistically significant (p = 0.09). Instead, a significant worse OS (p = 0.027) was observed for Luminal pts switching to Neu-like, compared with Luminal pts switching to other phenotypes. The phenotypical switch toward Neu-like did not affect LVI, pT, DFS, and OS compared with non-switched cases.
The Neu-like urothelial carcinoma could be identified by Piescore analysis. These tumors suggested a worse prognosis when MI tumor component underwent a phenotypical switch from NMI Luminal counterpart. Analysis for TILs evaluation are ongoing to characterize microenvironment in this subset of cases.

ID 815
INTERTUBULAR SEMINOMA AND RETROPERITONEAL LIPOMA: UNUSUAL AND RARE ASSOCIATION

V. Belsito[1,2], P. Colombo[1,2], S. Pancetti[1,2], S.Zanella[1,2], A. Bonometti[2]

[1] Humanitas University, Rozzano, Italy; [2] Pathology Unit, Humanitas Research Hospital, Rozzano, Italy

Objectives: to investigate the currently unreported and intriguing association between two rare and potentially misdiagnosticable neoplasms.
Materials and Methods: a 50-year-old male, that underwent a right nephrectomy at the age of 2, presented complaining an abdominal pain. CT scan revealed two closely related right retroperitoneal masses, with different radiological density, suspicious for a dedifferentiated liposarcoma (DL). Due to un unconclusivebiopsy, a debulking surgery was performed for diagnostic and therapeutic porpoises.
Results: Grossly, the major mass was yellowish, soft and with homogeneous cut surface, while the smaller was recognized as a small retained testis with a tan-grayish cut surface and without macroscopic alteration. Histologically, the larger lesion was made by isometric mature HMGA2+, MDM2- adipocytes, and failed to show amplification of either MDM2 or CDK4 genes. The testis was examined in toto and showed marked tubular atrophy, diffuse germ cell neoplasia in situ (GCNIS) and an exclusively intertubular neoplastic proliferation of epithelioid cells with bland nuclei and clear cytoplasm. Both inter- and intratubular proliferations resulted OCT3/4+, and c-Kit+. Overall, we rendered a diagnosis of retroperitoneal lipoma (RL) associated with intertubular seminoma (IS).
Conclusions: In the retroperitoneal the most frequent mesenchymal neoplasms are DLS and angiomyolipoma (AML). RL is extremely rare, known in only 30 since 1947. Even though it may radiologically mimic DL, RL may be recognized by its mature adipocytic morphology and the lack of melanocytic markers of AML and of molecular aberration of DL. Moreover it display an indolent behavior. In uropathology, identification of intertubular growth in seminomas is common, but mostly associated tomass-forming lesions. Pure IS is reported in only 24 cases, it frequently associates to cryptorchidism, and should be differenti-

ated from hematological and melanocytic neoplasms through immunohistochemistry. Association of RL and IS is unreported and presented with a histological challenge, giving their prognostically significant differential diagnoses.

ID 843
PRIMARY EWING SARCOMA OF THE URETER: AN EXCEPTIONAL FINDING

L. Dore[1,2], M. Valeri[1,2], M. Iuzzolino[1,2], A. Bressan[1,2], V. Belsito[1,2], M. Cieri[1], G.M. Elefante[1], C. Lancellotti[1,2], L.M. Terracciano[1,2], P. Colombo[1,2].

[1] Department of Pathology, IRCCS Humanitas Clinical and Research Hospital, Rozzano, Italy; [2] Humanitas University, Department of Biomedical Science, Pieve Emanuele, Italy

Objectives: Ewing sarcoma (ES) is the second most common malignant bone tumor in children, with a highly aggressive behavior and molecularly characterized by FET-ETS translocation (85% with EWSR1). It is a small blue cell tumor that mainly occurs in the soft tissue; urogenital tract involvement is a rare event so ureteral localization is exceptional. Here we report the first Italian case of primary ureteral ES as unique manifestation of disease.
Case presentation: A 24 y/o patient with left lower back pain performed abdominal US and uro-CT that showed a narrowed left ureteral lumen and ipsilateral hydronephrosis. At intraoperative frozen section, morphology initially suggested an urothelial carcinoma. A left distal ureteral segmental resection was performed. Grossly, an obstructing grayish mass of 2 cm was found in the ureteral wall. Histological examination demonstrated solid growth of round to spindle small cell proliferation, with foci of necrosis. Immunophenotype showed CD99+, FLI1+, NKX2.2+, Synaptophysin+, GATA3-, Myogenin-, ERG-, WT1-, CKpan-. FISH revealed *EWSR1* translocation and primary ureteral ES was diagnosed. The patient underwent systemic therapy and radicalization surgery. After 12 months of follow-up the patient was alive with no sign of recurrence or metastatic disease. A literature review revealed only 4 further cases of primary ureteral ES: 2 were males and median age was 39,5 years. Hematuria and flank pain were the most frequent symptoms. Only 1 patient had local recurrence and distant metastases. All underwent surgery and 3 received systemic therapy. Tumors were all CD99+ and showed typical morphologic features and molecular analysis. *EWSR1-FLI1* fusion was detected in 2 patients. One patient died from disseminated disease.
Conclusions: This is the fifth reported cases of primary ureteral ES and the first in Italy. ES of the urogenital tract is a rare condition with nonspecific clinical presentation and a challenging diagnosis. We encourage awareness for these exceptional events in the differential diagnosis of ureteral lesions in young patients.

ID 853
TSC/MTOR MUTATED EOSINOPHILIC VACUOLATED RENAL CELL CARCINOMA. A PATHOLOGICAL AND MOLECULARSTUDY

S. Marletta[1,2], A. Caliò[1], G. Settanni[3], E. Munari[4], M. Brunelli[1], S. Fratoni[5], F. Pierconti[6], M.R. Raspollini[7], A. Marchetti[8], G. Martignoni[1,2]

[1] Department of Diagnostic and Public Health, Section of Pathology, University of Verona; [2] Department of Pathology, Pederzoli Hospital, Peschiera del Garda; [3] Department of Pathology, IRCCS Sacro Cuore Don Calabria Hospital, Negrar; [4] Department of Molecular and Translational Medicine, University of Brescia; [5] Division of Anatomic Pathology, S. Eugenio Hospital, Rome; [6] Division of Anatomic Pathology and Histology, Catholic University, Rome; [7] Histopathology and Molecular Diagnostics, AOU Careggi, Florence; [8] Division of Anatomic Pathology and Histology, University of Chieti

Objectives: A subset of high grade eosinophilic renal tumors harboring pathogenic mutations in tuberous sclerosis complex (*TSC*) or mammalian target of rapamycin (*MTOR*) genes has been recently described. Limited data are available so far and an indolent behaviour has been claimed for these emerging tumors.
Materials and methods: Herein four additional cases have been retrieved and extensively studied as for their morphological, immunohistochemical and molecular features.
Results: All tumors were made up of cells arranged in solid to nested architecture with eosinophilic cytoplasm, round nuclei, and prominent nucleoli. Intracytoplasmic vacuoles and wide cells with cytoplasmic shrinkage were observed, recalling the “spider cells” of cardiac rhabdomyomas of tuberous sclerosis patients. Three cases revealed histological documented metastases respectively involving a regional lymph node (case 1), the skull (case 2), and the liver (case 4). All tumors expressed PAX8, cytokeratin 8-18, and cathepsin K but not vimentin. Pathogenetic mutations in the *MTOR* gene were found by NGS in primary and metastatic tumors in all but one case, which showed alterations in the *TSC2* gene.
Conclusions: By reporting the clinical, morphological and molecular features of four additional cases, three of which with histologically confirmed metastases, we have widened the current knowledges on high grade eosinophilic renal tumors. Unique morphology, consistent immunoprofile, and specific molecular findings claim the belonging to a new entity for these tumors. Considering the possible aggressive behaviour, we propose the name TSC2/MTOR mutated eosinophilic vacuolated renal cell carcinoma.

Giovedì 13 Ottobre 2022

Sala A 08.30-09.40

PATOLOGIA MOLECOLARE 2

Moderatore: A. Marchetti

ID 781
TECHNICAL EVALUATION OF MSI ANALYSIS IN ENDOMETRIAL CANCER: A COMPARISON STUDY

C. De Luca[1], Laura Libera[2-3], Nora Sahnane[2-3], G. Gragnano[1], F. Conticelli[1], Paola Parente[4,] G.Troncone[1], Daniela Furlan[2-3,] U. Malapelle[1]

[1] Department of Public Health, University Federico II of Naples, Naples, Italy; [2] Unit of Pathology, Department of Medicine and Surgery, University of Insubria and ASST Sette-Laghi, Varese, Italy; [3] Research Center for the Study of Hereditary and Familial Tumors, Department of Medicine and Surgery, University of Insubria, Varese, Italy; [4] Unit of Pathology, Fondazione IRCCS Casa Sollievo Della Sofferenza, San Giovanni Rotondo, 71013 Foggia, Italy

Objectives: Mismatch repair system deficiency (dMMR), which causes microsatellite instability (MSI), characterizes about 30% of endometrial cancers (ECs). (1) The identification of dMMR and severe microsatellite instability (MSI-H) in endometrial cancers (ECs) plays a pivotal role in the screening, diagnosis and therapeutic stratification of tumour patients.
Materials and methods: We compared the diagnostic performance of four MSI molecular tests based on fragment length assay in capillary electrophoresis (OncoMate™ MSI assay, Promega) and in microcapillary electrophoresis (TapeStation 4200, Agilent); with high-resolution melting (HRM)analysis approaches (Idylla™ MSI Test, Biocartis; EasyPGX® ready MSI, DiatechPharmacogenetics) on a series of 56 ECs.
Results: The diagnostic power and the sensitivity of fluorescence capillary electrophoresis (AUC 0.98 and sensitivity 96.8%) were higher respect to other methodologies. Otherwise, HRMapproaches and microcapillary electrophoresis platform failed to detect MSI-ECs showing minimal microsatellite shifts. (2)
Conclusions: In conclusion, whereas in colorectal site several technologies are eligible for MSI test, in ECs MSI test should be based on fluorescent capillary electrophoresis as it identifies a higher proportion of cases that could be misdiagnosed with other strategies.

References

1. Gilson P, Levy J, Rouyer M, et al. Evaluation of 3 molecular-based assays for microsatellite instability detection in formalin-fixed tissues of patients with endometrial and colorectal cancers. Sci Rep 2020;10(1):16386. https://doi.org/10.1038/s41598-020-73421-5.
2. Libera L, Sahnane N, Carnevali IW, et al. Microsatellite analysis of sporadic and hereditary gynaecological cancer in routine diagnostics. J Clin Pathol 2017;70(9):792-97. https://doi.org/10.1136/jclinpath-2017-204348

ID 819
LANDSCAPE OF CELLULAR INTERACTIONS IN HEPATOCELLULAR CARCINOMA

S Piscuoglio[1,2], M Coto-Llerena[1,2], A Benjak[1], C. Ercan[1,2], CKY Ng[4,5], LM Terraciano[3]

[1] Institute of Medical Genetics and Pathology, University Hospital Basel, 4031 Basel, Switzerland; [2] visceral Surgery and Precision Medicine Research Laboratory, Department of Biomedicine, University of Basel, 4031 Basel, Switzerland; [3] Unit of Anatomic Pathology, Humanitas University, Pieve Emanuele, Milan, Italy; [4] Department for BioMedical Research, University of Bern, 3008 Bern, Switzerland; [5] SIB Swiss Institute of Bioinformatics, Lausanne, Switzerland.

Objective. Hepatocellular carcinoma (HCC) typically develops on a background of cirrhosis resulting from the chronic inflammation. Immune checkpoint inhibitors, which act on the molecular pathways and cellular interactions of the cells in the tumour microenvironment (TME), have shown efficacy in HCC. However, little is known about the landscape of cellular interaction in HCC. Here, we aim to define the landscape and architecture of cellular populations in HCC and to identify TME reprogramming by hepatocytes.
Material and methods: We perform single-cell RNA-sequencing of 15 HCCs and 2 normal livers. Using bioinformatics tools and incorporating external datasets of normal cells, we define the multi-layered cellular ontology and their molecular signatures, the influence of hepatocytes on the TME and the influence of endothelial cells on the immune cell populations using ligand-receptor analyses.
Results: We identified 4 cell clusters consisting predominantly of hepatocytes, 2 clusters of macrophages and monocytes, 2 clusters of endothelial cells, 6 clusters of T/NK-cells, 2 cluster of B-cells, 1 cluster of fibroblasts, 1 cluster of dendritic cells and 1 cluster of mast cells. Using the NATMI tool, we identify 15092 interacting ligand-receptor pairs between diverse cell types. Fibroblasts and endothelial cells were the signaling cells involving the largest number of ligand-receptor pairs in both paracrine and autocrine signaling. By contrast, few ligand-receptor pairs were identified in T/NK cells. In terms of overall expression, hepatocytes primarily act on macrophages, endothelial cells and fibroblasts but also on all other cell types in the TME.
Conclusions: Our preliminary results provide novel insights into the complexity of the HCC TME and the interaction between cell types.

ID 863
DETECTION OF RET FUSIONS IN NSCLC PATIENTS BY INTEGRATED NGS ANALYSES OF TISSUE AND LIQUID BIOPSIES

S. Buglioni[1], M. Allegretti[2], V. De Pascale[2], P. Visca[1], E. Gallo[1], C. Ercolani[1], P. Giacomini[3], G. Ciliberto[4], F. Cappuzzo[5], G. Minuti[3].

[1,2,3,4,5] IRCCS Regina Elena National Cancer Institute, Rome, Italy; [1] Pathology Unit, [2] Oncogenomic and Epigenetic Unit, [3] Phase 1-Clinical Trial Center, [4] Molecular Tumor Board, [5] Medical Oncology 2

Objectives: To define an integrated NGS workflow for the identification and longitudinal monitoring of RET fusions in tissue and blood samples from NSCLC patients.
Materials and methods: Patients (n = 3) with diagnosis of RET-positive, metastatic NSCLC were sequentially recruited in our Institute between January to June 2022. Serial blood drawings were prospectically collected at recruitment and at fixed timepoints. All samples were subjected to NGS analysis by the Ion Genexus system and the Oncomine Precision Assay (OPA) gene panel. Correlation between molecular data and clinical imaging was done according to RECIST criteria v1.1.
Results: By comparing tissue and blood profile at baseline, a complete overlap (100%) in RET fusions calling was observed in all patients. In one case, the two RET fusions, previously observed in the primary tumor, were detected at the same ratios also in the corresponding blood sample. Moreover, at the first evaluation after therapy administration (4 weeks), liquid biopsy analysis was able to document a significant reduction of RET fusions as well as of some DNA mutations into the bloodstream. Tumor mutations were used as surrogates of tumor content and clinical response, e.g. to avoid false negative results on fusion calling. These results appeared more evident at 8 weeks after the initiation of targeted therapy where a complete abrogation of RET fusions and tumormutations were noted. This highlights once again the strict complementarity of these techniques. Importantly, clinical evaluation of disease evolution at the last timepoint correlates with molecular data in 3/3 patients.
Conclusions: Molecular analysis of tissue and blood samples by OPA NGS gene panel is reliable for detecting RET-positive NSCLC. Moreover, longitudinal monitoring of RET fusions by liquid biopsy may offer additional possibilities to monitor disease evolution and anticipate clinical progression of patients.

ID 899
REAL-WORLD DATA ON UNCOMMOM *EGFR* MUTATIONS AND *ERBB2* EXON 20 INSERTIONS IN NSCLC

A. Bragoni[1,2], A. Chesta[2], A. Sapino[1,2], E. Berrino[1,2], C. Marchiò[1,2]

[1] Department of Medical Sciences, University of Turin, Turin, Italy; [2] Pathology Unit, FPO-IRCCS Candiolo Cancer Institute, Candiolo, Italy

Objectives: The treatment choice of non-small-cell lung cancer (NSCLC) patients relies on the investigation of mutations in Epidermal Growth Factor Receptor (*EGFR*). Several phase III studies have demonstrated the benefit of tyrosine kinase inhibitors (TKIs) targeting EGFR in *EGFR* mutated tumors, compared with standard chemotherapy. Exon 19 deletions and exon 21 L858R point mutations represent the majority of *EGFR* mutations (90%) and positively correlate with TKIs sensitivity. The remaining 10% includes a heterogeneous subgroup of cases with different prognostic values and variable TKI sensitivity, yet to be fully elucidated. In addition, *ERBB2* exon 20 insertions can be identified in 4% of NSCLC, in a mutually exclusive manner with other driver alterations, and like *EGFR* exon 20 insertion are associated with resistance to TKIs.
Materials and methods: A retrospective series of advanced NSCLC patients harbouring an uncommon *EGFR* mutation (12 cases) or an *ERBB2* exon 20 insertion (4 cases) and diagnosed between 2011 and 2022 was collected. Formalin fixed paraffin embedded (FFPE) tissues were retrieved and DNA was extracted and sequenced using the OCAv3 panel (Thermo Fisher Scientific), covering 161 cancer genes.
Results: Among *EGFR* mutated cases, 5 harboured exon 18 point mutations (3 G719A, 1 E709V and 1 E709K), 4 showed exon 20 insertions, and 3 had exon 21 point mutations (2 L861Q and 1 L858M). Across the whole cohort *TP53* (6,37%) and *NOTCH2* (4, 25%) were the most frequently mutated genes. All *ERBB2* mutated cases harboured a *TP53* mutation and 2 out of 4showed a concomitant low copy number *ERBB2* gain. Two *ERBB2* mutated cases harboured a concomitant *ERBB4* mutation and an *ATM* mutation respectively. Two *EGFR* mutated cases have a concomitant *MAPK* / *ERK* pathway genes mutation (*KRAS* and *BRAF* respectively).
Conclusions: Uncommon-*EGFR*-mutated and *ERBB2* mutated NSCLCs harbour an articulated mutational profile. The concurrent presence of mutations in *MAPK* / *ERK* pathway genes or in other ErbB family genes may justify the differential sensibility to TKIs.

ID 930
ALTERATION OF THE RATIO BETWEEN GALECTIN-3 AND CD68 IN ALVEOLAR MACROPHAGES IN LETHAL COVID-19 LUNG DISEASE

G. Pannone[1], M.C. Pedicillo[1], G. Villani[1], M.A. Ramunno[1], C. Pizzulli [1], I.S. De Stefano[1], R. Franco[2], F. Zitomarino[2], A. Ronchi[2]

[1] University of Foggia, Italy; [2]University of Napoli Campania, Italy

As of March 1, 2022, the outbreak of SARS-CoV-2

disease has spread six continents, and almost six millions of people had died. Despite selective pressure of variants considered less lethal, a still high number of patients continue to die in the world, making the histological study of lethal variants of the disease particularly relevant. About 155 thousands of these deaths occurred in Italy. This study was focused on lethal lung disease 25 cases, with informed consent, occurred during the first two waves of COVID-19, before delta variant spreading.
Sars-CoV-2 has been detected in lung tissues of lethal Covid-19 disease by both PCR-based and In Situ Hybridization methods. Immunoistochemical analysis was performed on formalin-fixed paraffin embedded specimens derived from autoptical procedures. Immunostaining has been performed using specific monoclonal antibodies against CD61, TLR4, TLR-2, Galectin-3, CD-68, IL-6R, revealed by standard methods. This study has 4 major theorethical objectives, 1) To correlate detailed description of histopathological findings in lethal Covid-19 lung disease with 2) up-regulation of M1 medullary derived monocyte-macrophage inflammatory cells with defective M2 Macrophage activation (Galectin-3) of lung resident macrophages, related to lung resolution of inflammation, and 3) to demonstrate activation of hyperinflammation syndrome via innate immune persistent up-regulation, which in turn is related to lung megacaryocytes/platelet activation (CD61) with interstitial microtrombosis.
This study showed TLR-4/TLR-2 mediated hyperinflammation syndrome, with persistence of virus in the lungs (Spike ISH+ in machrophages; RT-PCR+ for E, N, S strands), unresolved inflammation with persistent innate immune associated to defective macrophage shift (Galectin-3/CD68 unbalanced ratio), and increased number of hyperactivated lung megacaryocytes/ platelets (CD61+).
Lethal disease may counteracted by early therapy aimed to repress innate immune hyperinflammation, restore pro-resolving macrophage action, switching production from proinflammatory to pro-resolving mediators in order to avoid microthrombosis.

ID 945
FEASIBILITY OF COMPREHENSIVE GENOMIC PROFILING IN REAL-LIFE CLINICAL PRACTICE

L. Nibid[1], G. Sabarese[1], G. Merlini, G. Perrone[1]

[1] Pathology Unit, UCBM Hospital, Rome, Italy

Objectives: Here it was investigated the feasibility of Comprehensive Genomic Profiling (CGP) in real-life clinical practice in a prospective court of patients enrolled in STARTRK clinical trial.
Material and methods: 185 patients affected by solid tumors were evaluated by FoundationOne test, analyzing 324 genes through the DNA-based Next Generation Sequencing (NGS) technique. The histological and molecular diagnosis was previously performed at the Pathology Unit of UCBM Hospital. We classified CGP reports into three groups: completely satisfying (complete genomic data with MSI and TMB), partially satisfying (complete genomic data without result on MSI and TMB), and inconclusive (no genomic data obtained). For each case, the inclusion criteria for the sample were considered: age of the sample, the tumor area, and the percentage of neoplastic cells. CGP results were also compared with in-house molecular testing.
Results: The CGP approach led to inconclusive reports in 18% of tissue samples and partially satisfying in 7%. As regards NSCLC, inconclusive reports were obtained in 24% and partially satisfying in 10%. Satisfying reports were frequently obtained in tissue samples in line with inclusion criteria (≥20% of neoplastic cells and with a tumor area ≥25mm^2). To note, these standards do not guarantee a good result: 19/21 inconclusive reports were referred to samples in line with inclusion criteria. The age of the sample seems to be a central pre-analytical variable since 89% of satisfying reports were referred to tissue samples with an age lower than 6 months. The in-house molecular analysis detected the same targetable mutation of CGP; moreover, in the NSCLC setting, CGP identified 1/3 of ALK re-arrangements detected in-house.
Conclusions: The in-house evaluation of molecular status should be guaranteed to all patients for whom it is available a target therapy. CGP in real-life is useful to provide additional information (ie. TMB, loss of heterozygosity, etc.) to consider alternative therapies in selected patients. Tissue samples in line with inclusion criteria and with an age lower than six months should be preferred for the Comprehensive Genomic Profiling approach, even if complete results are not always guaranteed.

Giovedì 13 Ottobre 2022

Sala B 08.30 - 09.40

MISCELLANEA 2

Moderatori: C. Giordano, M. Paulli

ID 768
PRELIMINARY STUDY ON THE IMMUOHISTOCHEMICAL EXPRESSION OF GALECTIN-3 IN HYPERTROPHIC HEARTS

C. Salzillo[1], G. Serio[1], S. Maniglio[1], A. d'Amati[1], T. Lettini[1], A. Marzullo[1]

[1] Unit of Anatomic Pathology, Dept. of Emergency and Organ Transplantations, University of Bari, Bari, Italy

Galectin-3 (Gal-3) is a member of the galectins family of carbohydrate-binding proteins [1] expressed in the cytoplasm of different cell types, mainly by activated macrophages, and regulates basic cellular functions including growth, proliferation, differentiation, and inflammation. Gal-3 overexpression is also associated with fibroblastic proliferation and the production of collagen, resulting in increased cardiac fibrosis and remodelling [2].
Aim of the study was to investigate on the expression of Gal-3 in hypertrophic hearts. We examined 16 surgical specimens taken from interventricular septum of 8 patients with Tetralogy of Fallot (9-15 months), 4 with aortic valve stenosis (48-75 years old) and 4 myocardial biopsies of patients submitted to heart transplantation (51-77 years old).
All the samples were routinely processed, stained with Ematoxylin-Eosin, Trichromic stain and elastic fiber stain and selected by having the morphological features of myocardial hypertrophy: myocytolysis, nuclear pleomorphism, interstitial fibrosis.
At immunohistochemistry, myocardial fibres showed cytoplasmic expression of Gal-3 in the 4 patients with aortic valve stenosis (diffuse in 3 and mild in 1) and in the 3/4 transplanted hearts (mild). The 8 patients affected from Tetralogy of Fallot resulted negative.
The results agreed with the hypothesis that Gal-3 may play a role in cardiac hypertrophy; its expression in myocardial fibers is not related with the morphological aspects as suggested by the absence in paediatric cases. The presence in myocardial biopsies of transplanted hearts would suggest a possible role in predicting clinical outcome of such patients.

References

1. Bi J, Garg V, Yates AR. Galectin-3 and sST2 as Prognosticators for Heart Failure Requiring Extracorporeal Life Support: Jack n' Jill. Biomolecules. 2021 Jan 27;11:166. https://doi.org/10.3390/biom11020166
2. Frunza O, Russo I, Saxena A, et al. Myocardial Galectin-3 Expression Is Associated with Remodeling of the Pressure-Overloaded Heart and May Delay the Hypertrophic Response without Affecting Survival, Dysfunction, and Cardiac Fibrosis. Am J Pathol. 2016;186:1114-27.https://doi.org/10.1016/j.ajpath.2015.12.017

ID 954
COVID-19 VACCINE AND DEATH

E. Maresi[1], G. De Lisi[1], E. Orlando[1], S. Zerbo[1], A. Argo[1]

[1] Department of Promotion of Health and Mother and Child Care, Internal Medicine and Medical Specialties, University of Palermo, Italy

Objectives: The rare vascular adverse reactions temporally related to anti SARS CoV-2 vaccine administration have induced diffidence in the population. In particular, researchers worldwide are focusing on the so-called "thrombosis and thrombocytopenia after vaccination".
This study aims to establish a practical workflow to define the relationship between adverse events following immunization (AEFI) and COVID-19 vaccination. Post-mortem investigation plays a pivotal role to support this causality relationship when death occurs.
Materials: From January 2020 to July 2022 at the Institutes of Forensic Medicine of Catania and Palermo, were carried out 11 autopsies (n. 2 in Catania; n. 9 in Palermo) of people died after anti SARS CoV-2 vaccination (Astra Zeneca: 7 cases, 64%; Pfizer: 2 cases, 18%; Moderna: 2 cases, 18%). Seven cases were male; four cases were female. Age ranged from 37 to 75 years (mean age 57 years; median age 65 years). In 10 cases (91%) death occurred after > 1 week from administration, in 1 case only after 12 hours. In 9 (81%) cases death occurred after first administration of the vaccine while in 2 (19%) cases after the 2nd vaccination.
Results: In 6 (54%) cases the death occurred secondary to a thrombosis (venous thrombosis, 3 cases; artery thrombosis, 3 cases). Venous thrombosis were located in intracranial and mesenteric vessels while artery thrombosis were located at the coronary level. In the remaining cases the cause of death was: hyper-eosinophilic syndrome, 1 case; macrophage activation syndrome, 1 case; active lymphocytic myocarditis, 1 case; perforated bacterial enteritis, 1 case; dilated ischemic cardiomyopathy, 1 case.
Conclusions: Our cases highlight the high frequency (54%) of vascular thrombosis in the determinism of death after SARS CoV-2 vaccine. The coronary thrombosis, in 2 cases, was observed in the context of vulnerable atherosclerotic plaques while in 1 case the culprit coronary arteries were free from pathological substrates. Venous thrombosis was observed only in patients vaccinated with Astra Zeneca vaccine, whereas arterial thrombosis was associated with all anti-SARS CoV-2 vaccines.
In no case there was vascular thrombosis associated with the presence of antibodies against platelet factor 4.

ID 764
INDETERMINATE CELL HISTIOCYTOSIS IS CLINICALLY AND PROGNOSTICALLY DIFFERENT BETWEEN PEDIATRIC AND ADULT PATIENTS

S. Zanella[1,2], V. Belsito[1,2], S. Pancetti[1,2], A. Bonometti[2]

[1] Humanitas University, Rozzano, Italy; [2] Surgical Pathology Unit, Humanitas Research Hospital, Rozzano, Italy

Objectives: indeterminate cell histiocytosis (iCH) is a rare myeloid neoplasm of adulthood, with some cases described during childhood, presenting with single or multiple erythematous skin papules and nodules. It is histologically similar to Langerhans cell histiocytosis (LCH), both by morphology and positivity for

S100p and CD1a. iCH however lack the expression of CD207/langerin. Its clinical course is variable and other associated hematological neoplasms (AHN) are sometimes present. Giving the lack of literature on this disease, we decided toinvestigate if there are any clinical or pathological features of iCH with prognostic relevance and ifthey differ between age groups.
Materials and methods: we systematically reviewed the international medical literature, gathering 93 papers and including 106 patients with histologically confirmed diagnosis of iCH in the study.We therefore collected from each patient a set of clinical and histopathological data and divided them according to age at diagnosis and AHN. Finally, we performed univariate statistical analysis to compare qualitative variables between groups.
Results: all patients displayed CD1a+ mononuclear cell infiltrates, lacking CD207 expression and/or Birbeck granules at electron microscopy. The median age at diagnosis was of 50 years, M:F ratio was 1.4:1 and around 25% of them were children, with a median age of 5 years. Most patients displayed a skin-limited disease, but a tenth had a multisystem involvement. Interestingly, AHN was observed in a quarter of patients, and mostly in adults with a multisystem disease ($p = 0.007$). Patients' mortality was of 26% and correlated with age, multisystem involvement, and presence of AHN ($p < 0.02$).
Conclusions: our work suggests that adult iCH patients have a higher risk of presenting systemic involvement, of develop AHN, and of lower survival. On the other hand, children invariably presents with single-system disease that may self-heal. For this reasons it is critical for pathologiststo proper recognize iCH and to differentiate it from LCH, that on the contrary has more often a worst prognosis in children, but a better one in adults.

ID 827
PHENOTYPIC CHARACTERIZATION OF KIKUCHI-FUJIMOTO DISEASE

Federico Scarmozzino[1], Marco Pizzi[1,2], Rachele Biancotti[1], Maurilio Ponzoni[3], Elena Sabattini[4], Angelo Paolo Dei Tos[1,2]

[1] Department of Pathology, Azienda Ospedale-Università Padova, Italy; [2] Department of Medicine, University of Padua School of Medicine, Italy; [3] Ateneo Vita-Salute San Raffaele University; Pathology Unit, San Raffaele Scientific Institute, Milan, Italy; [4] Haematopathology Unit, IRCCS Policlinico Sant'Orsola, Bologna, Italy

Objectives: Kikuchi-Fujimoto disease (KFD) is a self-limiting disorder of cytotoxic T cells, sometimes mimicking T-cell lymphomas. Histological criteria of KFD are established, but few studies have addressed the phenotypic aberrations of this disease. To address this issue, we evaluated a multi-institutional series of KFD focusing on potential diagnostic pitfalls.
Materials and Methods: In a discovery set of 16 KFD (Department of Pathology, Azienda Ospedale-Università Padova), the following parameters were considered: patients' demographics, KFD histological phase, extent of necrosis, loss of T cell antigen(s), Ki67 index and MUM1 expression in cytotoxic T cells (through CD8/MUM1 co-localization), since MUM1 can be expressed in T cell lymphomas. The obtained results were matched with a validation set of 21 cases (Pathology Units of San Raffaele Instituteand Policlinico Sant'Orsola). Wilcoxon-Mann-Whitney test and Pearson analyses were applied for statistical considerations.
Results: The discovery set showed a prevalence of female patients (M:F ratio = 0.7) with median age at diagnosis of 22.3 years (range: 10-57). Proliferating and necrotic phase KFD was documented in 8/16 (50.0%) cases, each. Partial/complete loss of pan-T cell antigens was reported in 7/16 (43.8%) cases. CD5 was the most frequently defective marker (6/16, 37.5%). In all cases, cytotoxic T cells were immunoreactive for MUM1 (mean positivity: 37.6% cells; range: 1-80%) and the latter was more expressed in proliferating than necrotic phase KFD (55.7% *vs* 13.7% cells; $p < 0.01$). MUM1positivity was also associated with the Ki67 index ($r = 0.82$; $p < 0.01$) and inversely correlated withthe extent of necrosis ($r = -0.71$; $p < 0.01$). These results were confirmed in the validation set.
Conclusions: Loss of T cell antigen(s) and frequent expression of MUM1 by cytotoxic T cells are additional findings in KFD. Besides their diagnostic relevance, these features deserve investigation under a pathogenetic perspective.

ID 871
EXPRESSION AND PROGNOSTIC VALUE OF FKBP51 IN HODGKIN LYMPHOMA

S. Varricchio[1], D. Russo[1], R. Accarino[1], P. Moretta[1], L. Stasio[1], E. Vigliar[2], M. Picardi[3], S. Romano[4] MF. Romano[4], M. Mascolo[1]

[1] Department of Advanced Biomedical Sciences, Pathology Section, University of Naples "Federico II", Naples, Italy; [2] Department of Public Health, University of Naples Federico II, Naples, Italy; [3] Department of Clinical Medicine and Surgery, Hematology Section, University of Naples "Federico II", Naples, Italy; [4] Department of Molecular Medicine and Medical Biotechnology, University of Naples Federico II, Naples, Italy

Objectives: Although Hodgkin lymphoma (HL) has an excellent prognosis with very high cure rates of >90%, approximately one-third of patients will not respond completely to frontline treatment or relapse1. Therefore, new predictive biomarkers are needed to stratify therapy according to the risk profile. Several recent studies evidenced FK506-binding protein 51 (FKBP51) as a reliable prognostic factor in several

human solid malignancies2. FKBP51 plays a relevant role in activating NF-kB signal transduction pathway2, constitutively deregulated in HL3. We aimed to investigate the possible value of FKBP51 expression as a new outcome marker in patients with HL.
Materials and methods: FFPE tissue sections of 114 HL cases were selected and evaluated for FKBP51 expression on tumour cells and microenvironmental T-lymphocytes. Results were compared with clinicopathological data and the outcome of patients.
Results: FKBP51 was expressed in HL (69.9%) on immunohistochemistry, with mostly weak intensity. All HL cases showed a nuclear FKBP51 in the background lymphocytes, particularly in the CD4+ cells adjacent to Hodgkin/Reed-Sternberg cells. Multivariate analysis revealed a significant association between unfavourable outcome and overexpression of FKBP51 in microenvironmental T-lymphocytes (p=0.004).
Conclusions: We suggest a potential role for FKBP51 as a new reliable prognostic biomarker for HL.

References

1. Swerdlow SH, Campo E, Harris NL, et al. World Health Organization Classification of Tumours of Haematopoietic and Lymphoid Tissues. 4th ed. IARC Press; Lyon, France: 2017.
2. Hähle A, Merz S, Meyners C, et al. The Many Faces of FKBP51. Biomolecules. 2019;9(1):35. Published 2019 Jan 21. doi:10.3390/biom9010035.
3. Weniger MA, Küppers R. NF-κB deregulation in Hodgkin lymphoma. Semin Cancer Biol. 2016 Aug; 39:32-9. doi: 10.1016/j.semcancer.2016.05.001.

ID 891
AN AGGRESSIVE CASE OF MDS/MPN-U WITH CLINICAL, HISTOPATHOLOGICAL, AND MOLECULAR FEATURES INTERMEDIATE BETWEEN CNL AND CMML

A. Bonometti[1], S. Zanella[1,2], S. Pancetti[1,2], V. Belsito[1,2], D. Rahal[1]

[1] Pathology Unit, Humanitas Research Hospital, Rozzano, Italy; [2] Humanitas University, Rozzano, Italy

Objectives: Chronic myeloid neoplasms with monocytosis include MDS/MPN and MPN with monocytosis. Their differential diagnosis may be challenging and bear therapeutic and prognostic consequences. We hereby present an example of the biological fluidity of myeloid neoplasms and of a possible diagnostic and therapeutic dilemma.
Materials and methods: A 68 years-old male patient, presented to our attention with massive splenomegaly, normocytic anemia, and thrombocytopenia, with marked neutrophilia and monocytosis, progressively worsening in the last six months. In the suspicious of a myeloid neoplasm, a bone marrow biopsy was performed.
Results: Histologically, the trephine biopsy was hypercellular with an increase of the myeloid:erythroid ratio with preserved granulocyte maturation and frequent monolobated megakaryocytes. CD14+ monocytes were increased, while CD34+ blast accounted for 3% of BM cellularity. Gömöri silver stain highlight a grade 1 fibrosis. Karyotype was normal, while MPS analysis revealed the presence of pathogenic mutations in CSF3R, DNMT3A, JAK2, NRAS, SETBP1, U2AF1, and potentially pathogenic mutation in ETV6, PRPN11, and ZRSR2.
As a whole, we rendered a diagnosis of MDS/MPN-U with features intermediate between chronic neutrophilic leukemia and chronic myelomonocytic leukemia. The patient was candidated for HSCT transplantation. However, due to the worsening of its cytopenias was treated with hypomethylating agents off-label and died 2 months after the diagnosis.
Conclusion: While the WHO-defined diagnostic criteria for myeloid neoplasm at diagnosis are well defined, it is known that disease evolution may present with hybrid features. Our patient presented at diagnosis with bicytopenia, stable neutrophilia, and monocytosis, and bear the diagnostic molecular alteration of CNL in association with other mutations also observed in CMML. According to WHO 2017 criteria, he had a single exclusion criterion for both CNL and CMML and was therefore considered "unclassifiable". Furthermore, despite the only minimal dysplastic changes and the lack of ASXL1 mutations, the patients pursued a very aggressive course.

ID 944
BCR EXPRESSION IN BURKITT LYMPHOMA: NEW INSIGHT IN MUTATIONAL LANDSCAPE

M. Vannucchi[2], M.C. Siciliano[1], S. Tornambè[2], M. Granai[3], F. PArcuri[2], E. Sorrentino[2], V. Mancini[2], N. De leone[2], S. Lazzi[2], R. Siebert[4], L. Leoncini[5], C Bellan* [5]

[1] Medical Biotechnologies, [2] Medical Biotechnology, University of siena, [3] Institut für Pathologie und Neuropathologie,University of Tubingen, [4] Institut für Humangenetik, Universitätsklinikum Ulm, 5MedicalBiotechnology, University of Siena

Objective:BCR activation is an important step in lymphomagenesis. In sBL the progressive acquisition of mutations in the TCF3/ID3 genes results in intrinsic tonic activation of BCR signaling. Therefore, neoplastic cells may grow in an EBV (and/orother antigen) independent way. Usually in BLsurface IgM represents functional BCR. However high number of IgA transcript have recently reported.Furthermore, IgM/D/G/A-negative cases have also been reported. Interestingly, heterozygous *NRAS* and *KRAS* gain of function mutations have been identified in one sample of such cases, suggesting that BL cells acquiring *RAS* mutations may lose the selective pressure to

express a functional BCR. The aim of the present study is to determines the expression of BCR by IHC in a large series of BL and to investigate the possible constitutive activation of the MAPK pathway through RAS mutations.
Materials and Methods: We performed an IHC analysis to detect IgA/IgM/IgD/IgG expression in 72 FFPE BL samples. We performed qPCR for RAS hotspot mutations and an NGS analysis (Illumina-platform) with a capture-base custom panel covering 83 genes known to be involved in lymphomagenesis.
Results*:* 56/72 cases expressed surface IgM but we also found 9/72 expressing IgA. Interestingly, 7/72 lacked BCR expression, being negative for all surface immunoglobulins. BL expressing surface IgA were localized in the oral and gastrointestinal mucosa, in line with previous studies.Wefounded *KRAS* somatic mutations in the classic hotspots of the gene by qPCR in 2/5 cases lacking expression of BCR. No *NRAS* mutations were identified in our cohort. Interestingly, the cases with *KRAS* gain of function were those with a high mutational burden, carrying more than 5 mutation. EBV infection as detected in 1/2 *KRAS* mutated samples.
Conclusion: We confirmed the presence of RAS mutations in IgM/D/G/A-negative BL supporting the previous hypothesis that the constitutive RAS/MAPK activation can bypass the requirement for a functional BCR/PI3Kδ axis.

Giovedì 13 Ottobre 2022

Sala C 08.30 - 09.20

PATOLOGIA PLEUROPOLMONARE 1

Moderatore: R. Franco

ID 759
EVALUATION OF IMMUNE MICROENVIRONMENT IN NON-SMALL CELL LUNG CARCINOMA AFTER IMMUNOTHERAPY

E. Rullo, M. Leopizzi, G. Pata, G. d'Amati, A. Pernazza

Department of Radiological, Oncological and Pathological Sciences, Sapienza University of Rome, Rome, Italy

Objectives: Immune checkpoint inhibitors have been used in a limited number of cases in the neoadjuvant setting for therapy of non-small lung cell carcinoma (NSCLC). Data regarding histological features of the tumor microenvironment in such cases are limited. The aims of this study are: i) to analyze the histological features of NSCLC after immunotherapy; ii) to characterize by immunohistochemistry the lymphocytes subsets within the tumor microenvironment; iii) to correlate the histological and immunohistochemical findings with the tumor regression grade (TRG).
Materials and methods: We evaluated lung specimens from patients with NSCLC undergoingsurgery after neoadjuvant therapy with immune checkpoint inhibitors in our Institution from January 2020 to December 2021. For each case we recorded the histological features and performed the following immunohistochemical stains: CD20, CD3, CD4, CD8, CD56, CD68, CD138, IgM, CD137, FOXP3, Perforin, Granzyme e PD-L1. We then digitalized the slides and counted thenumber of cells positive for each marker in multiple areas of equal size, representative of inflammatory "hotspots" (total area: 5mm^2). The results of the counts were compared to TRGthrough Pearson correlation.
Results: Six cases satisfied the inclusion criteria of the study. Prevalence of tertiary lymphoid structures, low cytotoxic lymphocytes count, low expression of PD-L1 and high expression of CD137 and Granzyme from T cells within the tumor microenvironment were associated with higherTRG. The expression of other markers was not significantly different in relation to outcome.
Conclusions: Our results confirm the role of the immune microenvironment in the response of NSCLC to immunotherapy and highlight the importance of CD137 expression as a marker of immune activation, as proven by the effective tumor killing activity in cases with the highestexpression of CD137 from T cells. The inverse relationship between the extent of TRG and T cytotoxic lymphocyte counts is an unexpected finding of our study, which deserves further investigation.

ID 773
TUMOUR INFILTRATING LYMPHOCYTES (TILS) AND PD-L1 EXPRESSION IN MALIGNANT PLEURAL MESOTHELIOMA.

G. Bellezza[1], M. Mandarano[1], C. Floridi[1], F. Di Luca[1], R. Del Sordo[1], R. Colella[1], S. Ceccarelli[2], G. Metro[3], F. Puma[2], A. Sidoni[1].

[1] Section of Anatomic Pathology and Histology, Department of Medicine and Surgery, University of Perugia, Italy; [2] Section of Thoracic Surgery, Department of Medicine and Surgery, University of Perugia, Italy; [3] Medical Oncology, Perugia Hospital, Italy.

Objectives: Malignant pleural mesothelioma (MPM) is an aggressive form of tumour. In MPM immunotherapy has some antitumour activity, but predicting response remains challenging. Large studies of PD-L1 expression in MPM using approved anti-PD-L1 assay are lacking[1]. Here we investigate the correlation between tumour infiltrating lymphocytes (TILs) and PD-L1 expression with clinical and pathological features.

Materials and methods: 90 samples of MPM were reviewed. Some clinical and histological characteristics were considered; grading was assigned for epithelioid MPM using the new scoring system (WHO 2021). For TILs, we evaluated intensity of infiltrating lymphocytes as absent, low, moderate and high. For PD-L1 evaluation, Kit DAKO 22C3 pharmaDX was used. Scoring was dichotomized into positive (≥1%) or negative (< 1%).
Results: This study comprised 72 male (80%) and 18 female (20%) with a median age of 69 years (range 42-84); 79 MPM (88%) were epithelioid, 10 (11%) sarcomatoid and 1 biphasic (1%). As regards histological grade, 67 (85%) epithelioid MPM were classified low grade and 12 (15%) high grade. 46 samples (52%) showed a low or moderate component of TIL, particularly in epithelioidMPM (45%). No significant correlation was observed between grade and intensity of TILs. PD-L1 expression was more expressed in sarcomatoid than in epithelioid MPM (90%vs26%) (p = .000046) and in high grade respect to low grade epithelioid MPM (83%vs15%)(p = .00001).
Conclusions: Our study suggests that PD-L1 expression is related to an aggressive MPM behaviourand the role of PD-L1 as a possible predictive marker needs to be further investigated.

References

1) Sobhani N, Roviello G, Pivetta T, et al. Tumour infiltrating lymphocytes and PD-L1 expression as potential predictors of outcome in patients with malignant pleural mesothelioma. Mol Biol Rep. 2019;46:2713-2720. https://doi.org/10.1007/s11033-019-04715-9.

ID 794
MESOTHELIOMA AND BAP1 TUMOUR PREDISPOSITION SYNDROME: FROM DIAGNOSIS TO PREVENTION

A. Marzullo[1], A. d'Amati[1], F. Fortarezza[2], S. Maniglio[1], D. Cavone[3], G. Marulli[4], A. De Palma[4], C. Caporusso[1], F. Pezzuto[2], G. Serio[1]

[1] Pathology Unit, Department of Emergency and Organ Transplantation, University of Bari, Bari, Italy; [2] Pathology Unit, University Hospital of Padova, University of Padova, Padova, Italy; [3] Interdisciplinary Department of Medicine, Occupational Medicine, University of Bari, Bari, Italy; [4] Thoracic Surgery Unit, Department of Emergency and Organ Transplantation, University of Bari, Bari, Italy

BRCA-1 associated protein 1 (BAP1) tumour predisposition syndrome is a hereditary condition determined by germline mutation of the tumour suppressor gene BAP1. This disorder is associated with the development of a variety of both benign and malignant tumours, mainly involving skin, kidney, liver, eye, CNS and mesothelium. We report on the case of a family unaware of carrying BAP1 germline mutation, in which the condition has been firstly suspected in the father, died of pleural mesothelioma. The suspicion of BAP1 tumour predisposition syndrome has been raised because in the past he had already received diagnosis of uveal melanoma and clear cell renal cellcarcinoma. Furthermore, just months before father's mesothelioma diagnosis, one of his sonsdeveloped colorectal cancer. In order to assess a possible BAP1 tumour predisposition syndrome, germline mutations of BAP1 were searched in peripheral blood samples from the man's relatives after his death. In all relatives, a germline mutation of the BAP1 gene was detected. We report on a familial cluster of BAP1 tumour predisposition syndrome, identified through a comprehensive molecular approach extended to all relatives. Since tumours arising in this context may have specific histological features and peculiar clinical behaviour, pathologists should be aware of this condition and its identification through appropriate genetic counselling could be vital for adequate primary, secondary and tertiary prevention measures for offspring.

ID 801
TRANSITIONAL MESOTHELIOMA PATTERN AND IMPACT ON SURVIVAL: A SINGLE-CENTRE COHORT STUDY

S. Maniglio[1], A. Marzullo[1], A. d'Amati[1], C. Salzillo[1], L. Scuccimarri[1], G. Marulli[2], A. De Palma[2], T. Lettini[1], L. Vimercati[3], G. Serio[1]

[1] Dept of Emergency and Organ Transplantation, Pathology Unit; [2] Thoracic Unit, Bari University, Italy; [3] Dept of Interdisciplinary Medicine, Occupational Health Unit, Bari University, Italy

Histologic subtype of mesothelioma is a major prognostic factor which guides the clinician in the choice of therapy. According to current guidelines, patients with epithelioid mesothelioma are considered possible candidates for surgery, and those with sarcomatoid mesothelioma (SM) are not.For the biphasic mesothelioma (BM), characterized by epithelioid and sarcomatoid pattern together, the percentage of sarcomatoid component limit the therapy. In the WHO 2021 the rare transitionalmorphologic pattern (TM) is not clearly recognized as an independent mesothelioma type; it is described as an aggressive sarcomatoid subtype. When observed in BM, patients seem to be refractory to therapy. The diagnostic criteria to identify this unusual and ambiguous pattern have not been established. We report on the prognostic significance of TM pattern detected in 132 (87 BM and 45 SM) cases collected from the Apulia Mesothelioma Register. Patients received only palliative or adjuvant chemotherapy/radiotherapy. We included all the cases with at least 5% of transitional component. Reticulin stain was used to recognize TM. A Cox regression model was used to identify predictors of survival;

Kaplan-Meier curves to summarize overall survival. On univariate analysis, stage (p = 0.02), age ≤70 (p = 0.021), Ki67 ≤25% (p = 0.004) and epithelioid prevalent pattern in BM (p = 0.0001) were significantly associated with better survival. Nosignificant survival differences were associated to transitional component (p = 0.55) in both BM and SM. The multivariate analysis confirmed that epithelioid features (p = 0.001), Ki67 (p = .05) and chemotherapy (p = 0.0001) were independent prognostic factors. The k intraobserver reproducibility test to recognize TM pattern was 0.23-0.37 without and 0.62-0.77 with reticulin stain. Thispreliminary study suggested that in BM the TM pattern should not limit the choice of therapy and does not affect the prognosis. Multicentric studies are needed to better clarify the prognosticsignificance of the transitional patter of mesothelioma.

ID 809
VASCULAR ENDOTHELIAL GROWTH FACTOR PATHWAY: A TOOL FOR CLUSTERING LUNG NEUROENDOCRINE TUMORS

F. Pezzuto[1], F. Fortarezza[1], F. Lunardi[1], G. Comacchio[1], L. Vedovelli[1], G. Serio[2], G. Pelosi[3], F. Rea[1],F. Calabrese[1]

[1] Department of Cardiac, Thoracic, Vascular Sciences and Public Health, University of Padova, Italy; [2] Department of Emergency and Organ Transplantation, University of Bari, Italy; [3] Department of Oncology and Hemato-Oncology, University of Milan, Italy

Objectives: The current WHO classification of lung neuroendocrine tumors (NETs) has remained largely unchanged with some limitations in reproducibility and clinical impact, particularly for carcinoids. They are considered low-grade NETs, although metastases and recurrences may occur. Neoangiogenesis is essential for tumor growth and is regulated by many angiogenic factors, including vascular endothelial growth factor (VEGF) and its receptors VEGFR1,2,3. The aim of the study was to evaluate the expression of VEGF pathway and its prognostic implications in lung NETs.
Materials and methods: A multicenter retrospective study was carried out on 109 consecutive lung NETs (67 typical carcinoids-TCs, 22 atypical carcinoids-ACs, 20 high grade-HG NETs). The expressions of VEGF and VEGFR1,2,3 were evaluated by immunohistochemistry, real-time polymerase chain reaction (PCR) and correlated with morphological (histotype, stage, Ki67) and clinical (age, sex, smoke, follow-up) features. A random forest-based machine-learning algorithm (Boruta) was implemented to identify which variables were associated with NET classification.
Results: ACs showed statistically significant higher expression of VEGF, VEGFR1,2,3 than TCs.In comparison with HG-NETs, ACs had significantly higher VEGFR1. VEGFR1 showed a bimodal distribution. Clustering for its expression, the lowest values were grouped under the HG-NETs. The Boruta algorithm demonstrated that Ki67, tumor size, stage, VEGFR2,3 were the most important variables for discriminating NET grade. The expression of VEGFR3 was significantly associated with lymph-node metastases in TCs and ACs. Real-time PCR analysis confirmed VEGFR1,2,3expression at the mRNA level.
Conclusion: The evaluation of the VEGF pathway in lung NETs may be useful for histological classification, patients' stratification, and potentially for personalized therapies. The activation of VEGFR3 plays a crucial role in lymphangiogenesis and its expression may be involved in lymphatic dissemination.

Giovedì 13 Ottobre 2022

Sala D 08.30 - 09.30

MISCELLANEA 3

Moderatori: E. Maiorano, M. Volante

ID 880
ALTERED EXPRESSION OF CSPG4 SUPPORTS MALIGNANCY OF PANCREATIC NEUROENDOCRINE TUMORS

G. Malpeli[1], R. Perris[2], S. Bersani[3], C. Luchini[3]

[1] Department of Surgery, Dentistry, Paediatrics and Gynaecology, University of Verona, Verona, Italy; [2] Department of Biosciences, COMT-Centre for Molecular and Translational Oncology, University of Parma, Parma, Italy; [3] Department of Diagnostics and Public Health, ARC-NET Research Centre, University of Verona, Verona, Italy

Objectives: Chondroitin sulfate 4 proteoglycan (CSPG4) is a highly complex membrane proteoglycan found in immature progenitor cells and also in pericytes.Cultured pancreatic islet-derived stem/precursor cells are known to express CSPG4 in addition to typical mesenchymal markers. CSPG4 has previously been found overexpressed in benign exocrine tumors of the pancreas, but no data is available for pancreatic neuroendocrine tumors (PanNET).
Materials and methods: Eighty-eight PanNET including 70 nonfuctional (NF) and 18 functional (F) were immunostained using a non-commercial anti-CSPG4 mAb by immunohistochemistry (IHC). IHC scores were correlated with the patients' clinical variables. To define the functional context of CSPG4 in PanNET, we performed signaling pathways analysis based on genes whose expression is correlated with that of CSPG4 in 32 PanNET of the ICGC (International Cancer Genome Consortium).
Results: CSPG4 weakly stained the cytoplasm and

plasma membrane of the islets of Langerhans of the normal pancreas. In PanNET, the CSPG4 score was 0 in 26 cases (25 of 26 were NF, P = 0.02) and 1-12 in 62 cases. Overall, CSPG4 expression was detected more frequently in F than in NF PanNET (mean score 6.5 vs 3.0, P = 0.00001). Percentage of Ki67-positive cells was higher in negative than positive cases (mean 29.5% vs 9.4%, P = 0.00005). Tumor size correlated positively with CSPG4 score in NF PanNET (P = 0.00001). Patient survival decreased based on the increase in CSPG4 score in the NF subtype (P = 0.03). Transcription related to the CSPG4mRNA level indicated main involvement of the epithelial-mesenchymal transition, focal adhesion, remodeling of the extracellular matrix, b1 and b3 integrin signaling pathways.
Conclusions: Overexpression of CSPG4 characterized most of PanNET and correlated with poorer prognosis in the NF subtype. Abnormal expression of CSPG4 may reflect benign expansion of islet cells in the F subtype and progression to malignancy in the NF subtype.

ID 941
PATHOLOGICAL AND MOLECULAR FEATURES OF ADRENOCORTICAL CARCINOMAS WITH MISMATCH REPAIR DEFICIENCY

G. Trucco[1], V. Zambelli2, I. Rapa[3], L. Daniele[4], M. Papotti[1], A. Berruti[5], M. Terzolo[6], M. Volante[2]

[1] Department of Medical Sciences, University of Turin, Turin, Italy; [2] Department of Oncology, University of Turin, Orbassano, Italy; [3] Pathology Unit, San Luigi Gonzaga Hospital, Orbassano, Italy; [4] Pathology Unit, Mauriziano Hospital, Torino, Italy; [5] Department of Medical and Surgical Specialties, Radiological Sciences and Public Health, University of Brescia, Brescia, Italy; [6] Department of Clinical and Biological Sciences, University of Turin, Orbassano, Italy;

Objectives: Mismatch repair (MMR) gene defects have been previously described in adrenocortical carcinoma (ACC), however its pathological features are unknown.
Our aim was to investigate prevalence, pathological and molecular features of MMR-deficient ACCs.
Materials and methods: Immunohistochemical expression of *MLH1*, *MSH2*, *MSH6* and *PMS2* was evaluated on one-hundred-twenty cases of ACC. Cases with altered MMR protein expression were tested for microsatellite instability (MSI) using a colon and endometrial cancer-approved kit (EasyPGX ready MSI KIT, Diatech Pharmacogenetics) and profiled using next-generation sequencing (NGS; Oncomine Comprehensive V3 panel, Thermofisher).
Results: Altered MMR protein expression was shown in 11/120 (9.2%) cases. *MSH6* loss, alone (5/11, 45%) or coupled with *MSH2* loss (3/11, 27), was predominant.
Loss of MMR associated with a higher prevalence of non-oncocytic histotype, higher T stage, as well as higher Weiss and Helsinki scores and higher Ki67 index.
Although no evidence of MSI was demonstrated in any of the cases, NGS found MMR gene mutations to be concordant with the protein expression in 7/11 cases (63%), suggesting an ACC-specific profile of instability. One case displayed *MSH2* and *MLH1* mutations with *MSH6* protein loss, whereas in the remaining three cases, no mutations were found, suggesting alternative deregulation mechanism(s).
Relevantly, all MMR-deficient cases harbored mutations in chromatin remodeling genes (≥1 mutation on *ARID1A*, *ATM*, *SMARCA4* or *ATRX*) and enriched for *TP53* gene mutations (9/11, 82%).
Conclusions: In our cohort, MMR alterations, mostly *MSH6* mutations, were found in 9.2% of ACC, and were coupled with mutations on chromatin remodeling genes and a high prevalence of *TP53* mutations. They associated with unfavorable pathological features.

ID 793
THE ROLE OF CD73 IN PREDICTING THE RESPONSE TO IMMUNOTHERAPY IN HEAD AND NECK CANCER PATIENTS

B Cerbelli[1], R Carletti[2], C Di Gioia[3], A Corsi[4], A Botticelli[3], A Cirillo[3], A Pisano[3], M Leopizzi[1], A Pernazza[1], G d'Amati[3]

[1] Department of Medical-Surgical Sciences and Biotechnologies; [2] Department of Translational and Precision Medicine; [3] Department of Radiological, Oncological and Pathological Sciences; [4] Department of Molecular Medicine, Sapienza University of Rome

Objectives: immunotherapy has a crucial role in the treatment of recurrent or metastatichead and neck squamous cell carcinoma (R/M HNSCC). However, only a small percentage of patients achieve long-term benefit in terms of overall response and survival. It was previously shown that HNSCC has an immunosuppressive microenvironment due to highlevels of both regulatory T cells and immunosuppressive molecules, such as lymphocyteactivation gene 3 and CD73.
The aim of our study was to investigate if the expression of CD73 by neoplastic cells couldaffect the efficacy of anti-PD-1 immunotherapy.
Materials and methods: we reviewed data from 35 patients with R/M HNSCC receiving first line immunotherapy with or without chemotherapy based on a combined positive score (CPS) > = 1 from March 2021 to November 2021. CD73 expression by cancer cells was evaluated on pre-treatment biopsies. Positivity for CD73 was defined as the presence of cytoplasmic and/or membrane staining, either strong or weak on neoplastic cells. The percentage of stained neoplastic cells (0–100%) was recorded. We ana-

lyzed the association between CD73 expression and early progression (EP), defined as progressionoccurring within 3 months.
Results: twenty-four patients were male (69%) and median age was 67 years.
In the majority of patients (91%) the primary tumor site was in the oral cavity or larynx.
All patients received pembrolizumab which was associated to chemotherapy in 22 (63%) of patients.
In 31 out of 34 patients CD73 was positive (91%) ranging from 1% to 80% (median value: 36%)
We observed a statistically significant association between CD73 expression over the median value and EP disease (p = 0.03).
Conclusions: our findings suggest that higher expression levels of CD73 could predict resistance to immunotherapy in patients with CPS positive R/M HNSCC. The addition of this biomarker to the routine evaluation of CPS could help to select the patients primary resistant to anti-PD-1 immunotherapy.

ID 855
POTENTIAL BIOMARKERS AS PREDICTORS OF IMMUNE RESPONSE IN HNSCC PATIENTS TREATED WITH IMMUNOTHERAPY (HEAD AND NECK PATHOLOGY)

F.L. Gambaro[1], R.Carletti[1], A.Botticelli[1], A.Cirillo[1], B.Cerbelli[2], G.d'Amati[1], A.Corsi[3], C.Della Rocca[2], C.di Gioia[1].

[1] Dpt of Radiological, Oncological and Anatomopathological Sciences; [2] Dpt of Medical-Surgical Sciences and Biotechnology; [3] Dpt of Molecular Medicine. Sapienza University of Rome

Objectives: The advent of immunotherapy has changed the landscape of oncologic treatment for PD-L1 positive recurrent/metastatic head and neck squamous cell carcinoma (R/M-HNSCC). However, although it has been shown that HNSCC microenvironment is distinctly immunosuppressive due to the high concentration of regulatory T cells (Treg cells) and lymphocyte activation gene 3 (LAG-3), only a small proportion of patients achieve a benefitin terms of overall response and overall survival from current immunotherapeutic protocols.Aim of our study is to evaluate if the tumour tissue expression of peculiar biomarkers can predict the therapeutic efficacy of anti-PD-1 drugs.
Materials and Method: Data from 35 CPS positive R/M-HNSCC patients receiving first lineimmunotherapy or immunotherapy with chemotherapy, between March-November 2021 were prospectively reviewed. Stromal and intra-tumoral Treg cells, LAG3 expression and CPS expression were evaluated on pre-treatment biopsies by immunohistochemistry to define an Immune Suppressive Profile (ISP) as LAG3+ and Treg+ cells. We evaluated the associations between early progression (EP), progression-free survival (PFS), overall survival (OS), objective response rate (ORR) and the expression of LAG3 and with the ISP.
Results: Twenty-four patients (69%) were male. Median age of the patients was 67 years. Primary tumor site was the oral cavity or larynx in 91%.Twenty-three (66%) patients presented both expression of LAG3 and presence of Treg cells.We showed both a significant association between patients with LAG3 positive HNSCC and EP disease (p = 0.005) and between ISP and EP (p = 0.023). Significative associations between PFS, OS, ORR with LAG3 or Treg status weren't observed.
Conclusions: Our findings showed that in CPS positive R/M HNSCC patients the contemporary expression of Treg cells and LAG3+cells could better select the patients primary resistant to anti-PD-1. The results of our study identify a potential immunological patients' profile, contributing to draw a novel scenario in precision immune-oncology**.**

ID 867
RARE SOFT TISSUE TUMORS OF THE ORAL CAVITY

C.C. Lin[1], L. Salvatorelli[1], L. Puzzo[1], G. Broggi[1], G.M. Vecchio[1], R. Garro[1], G. Magro[1]

Department of Medical and Surgical Sciences and Advanced Technologies, G.F. Ingrassia, University of Catania, Catania, Italy

Objectives: Among soft tissue tumors, those arising primarily in the oral cavity are relatively uncommon. They encompass numerous entities with different biological behavior, ranging from benign to highly aggressive lesions. Due to the rarity of these lesions in this anatomic site, serious differential diagnostic problems may be encountered by pathologists. We herein present a series of 5 rare cases of soft tissue tumors of the oral cavity, emphasizing the main diagnostic clues.
Materials and methods: We retrieved from the files of the pathology archives of Anatomic Pathology Section of University of Catania 23 cases of primary soft tissue tumors. Among these tumors, 5 rare cases, considered "*diagnostically challenging cases*" were selected. The age of patients ranged from 15 to 57 years. For each single case, clinico-radiologic features and paraffin blocks were available for immunohistochemical analyses.
Results: The following cases were studied: i) a 47-year-old man with spindle cell lipoma exhibiting unusual morphology of the tongue; ii) a 54-year-old woman with solitary fibrous tumor of the oral cavity, showing predominantly leiomyomatous-like features; iii) a 57-year-old woman with low-grade fibromyxoid sarcoma of the parapharyngeal space; iv) a 15-year-old girl with small round cell myofibroblatoma of the oral cavity, v) a 54-year-old man with *NTRK3*-rear-

ranged spindle cell tumor with coexpression of CD34 and S100 protein.
Conclusions: A wide variety of diagnostically challenging soft tissue tumors may be encountered in the oral cavity. The unusual/unexpected site may contribute to the diagnostic difficulties. In our series a small cell myofibroblastoma was misdiagnosed as embryonal rhabdomyosarcoma and, conversely, a low-grade fibromyxoid sarcoma was interpreted as a benign tumor; in addition, a case of solitary fibrous tumor was originally misdiagnosed as leiomyoma. Notably, we herein report the first case of *NTRK3*-rearranged spindle cell tumor arising in the oral cavity. The application of strict morphological criteria, along with the interpretation of the immunohistochemical and molecular analyses, is crucial for a correct diagnosis.

ID 875
PREDICTIVE AND PROGNOSTIC ROLE OF IMMUNOHISTOCHEMICAL ANALYSES OF CD44, PDL1 AND ATG7 IN 40 SQUAMOUS CELL CARCINOMAS

G. Tinnirello[1], L. Salvatorelli[1], M. Mazzucchelli[1], L. Puzzo[1], R. Garro[1]

Department of Medical and Surgical Sciences and Advanced Technologies, G.F. Ingrassia, University of Catania, Catania, Italy.

Objectives: The aim of our study is to highlight new prognostic and predictive factors, such as CD44, PDL1, ATG7, in surgical samples from patients with head and neck Squamous Cell Carcinoma (SCC) processed with Tissues Micro Array (TMA) technique.
Materials and methods: We analyzed the immunohistochemical expression of CD44, PDL1 and ATG7 in 40 cases of squamous cell carcinoma (SCC), including 28 cases occurring in the glottis, 7 in the epiglottis, 3 in the oropharynx and 2 in the hypoglottis.
Results: Immunohistochemical analyses showed 25 out of 40 cases positive to CD44; 9 out of 40cases positive to PDL1 positive and 8 out of cases positive to ATG7. The relationshipbetween clinical data (site, grade, stage, radio/chemotherapy and follow up) and immunohistochemical markers was evaluated.
Conclusions: In our cases PDL1 was expressed in low stage and in high stage carcinomas, but in the lattercases, patients with lymph node metastases at the time of diagnosis developed also distantmetastases during the follow up, despite radio/chemotherapy. We suppose that these high stage tumors could have developed chemoresistance to conventional therapies and couldobtain better results with immunochemotherapy to restore immunosurveillance. Target therapy based on monoclonal antibodies against PDL1 may 4rfwbe useful in high stage head and neck squamous cell carcinoma.
Finally, autophagy plays a critical role in the dormant phenotype of cancer stem cells, contributing to the survival, resistance to treatment and resilience of dormant cells. The inhibition of autophagy by drugs or genetic methods could restore cancer stem cells to proliferative phase, making them more responsive to common therapeutic strategies. Moreover, CD44 and ATG7 overexpression may also be useful in the evaluation of tumorbehavior, both in early and advanced SCCs. Further studies need to be performed to confirmour preliminary results.

Giovedì 13 Ottobre 2022

Sala E 08.30 - 09.30

PATOLOGIA MAMMARIA 2

Moderatore: F. Pietribiasi

ID 886
INVASIVE BREAST CARCINOMA IN THE ELDERLY

E. Kuhn[1,2], L. Runza[2], S. Ferrero[1,2], O. Garrone[2], G. Tomasello[2], L. Despini[2], D. Gambini[2]

[1] Department of Biomedical, Surgical, and Dental Sciences, Università degli Studi di Milano; [2] Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milan, Italy.

Objectives: The lengthening of the lifespan average led to an increase of breast cancer diagnosed in very old age. In these patients, the risk/benefit ratio of any intervention depends on the patient's life expectancy, the potential benefit obtained by the neoplastic disease, and comorbidities. Some studies have reported evidence of a reduction in breast cancer-related deaths in patients over eighty undergone surgery.
Our study aims to investigate the clinico-pathological features of invasive breast cancer in ultra-elderly patients.
Materials and methods: Patients diagnosed with invasive breast cancer in ultra-elderly(≥ 80 years) treated in our institution between 2014 and 2017 were included. Clinico-pathological data(age, size, side, treatment, histotype, stage, grading, biological markers, and follow-up data) were prospectively collected and analyzed.
Results: A total of 91 patients with a median age of 85 years (range:80-101) were treated for breast cancer. Among tumors, 47(53%) were left and 42(47%) right. Overall, 62(68%) underwent surgery(14 mastectomies, 46 quadrantectomies, 1 annessiectomy and 1 nodulectomy), while the other 29(32%) had only a biopsy. In resected tumors the median size was 1.7 cm(range: 0.14-105). Out of 90 cases, 54(60%) were classified no special type, 14(16%) lobular, 9(10%) mucinous, 3(3%) mixed and the remaining

were other special types, including 4 apocrine, 4 papillary and 2 metaplastic. Of 90 cases, 9(9%) were grade 1, 49(54%) grade 2, and 32(35%) grade 3. Molecular intrinsic suptype was luminal A in 38(42%) cases, luminal B in 35(38%), luminal B-HER2 positive in 7(8%), triple-negative in 7(8%), HER2+ in 2(2%), and undetermined in 2(2%). Median follow-up was 52 months(range: 2-88) months, and 25(29%) of 86 patients died. We observed a longer follow-up time between patients undergone surgery(median 61 versus26 months).
Conclusions: Our findings indicate that invasive breast carcinomas in the elderly are enriched with hormone-sensitive tumors(88%) and luminal subtypes(80%). The impact of surgery on these patients and the reason for shorter follow-up time in patients not surgically treated have to be further investigated.

ID 889
RARE CASE OF NODAL LOCALIZATION OF DUCTAL ADENOMA OF BREAST IN 86-YEAR-OLD WOMAN
A BREAST PATHOLOGY CASE REPORT WITH REVIEW OF LITERATURE

V. Pellegrini[1], R. Cappellesso MD[1], M. Lo Mele[1], A.P. Dei Tos[1]

[1] Surgical Pathology Unit, Department of Medicine (DIMED), Hospital-University of Padua, Italy

Objectives: The purpose is drawing attention on the rare phenomenon of intranodal papillary lesions creating a rarity in scientific literature to start a debate and keeping researching on it.
Material and Methods: Specimens were sampled and examined in the surgical pathology department and embedded in paraffin sections, cut at 4 µm from blocks and stained with H&E.
Results: An 86-year-old woman visited our hospital with a nodule located in outer quadrants of right breast with consensual lymphadenopathy, suspect of metastasis. She had a clinical history of multiple, homolateral intraductal papillomas and no familiar history for breast cancer. Gross appearance showed a nodule of 1,8 x 1,6 x 1,3 cm with solid consistence. Lymph node was 2,6 cm in its major axis. Microscopic appearance showed a solitary, solid adenomatous tumor consisting of epithelial and myoepithelial tubules without significant atypia surrounded by a thickened capsule both in the lymph node and in the mammary tissue.
Conclusion: Theory of pseudo-metastasis related to benign neoplastic changes is proliferation in ectopic breast tissue. Among these rare cases, the most common phenomenon is intranodal intraductal papilloma[1]. Although pathogenesis of ductal adenoma is still unclear, authors suggest they might arise from intraductal papilloma thus making the second theory the most supported by facts. The rarity in this case is the fact that no ectopic tissue was found and only the localization of the ductal adenoma was available in the lymphnode sent.

References

1. Boulos FI, Granja NM, Simpson JF et Al. Intranodal papillary epithelial proliferations: a local process with a spectrum of morphologies and frequent association with papillomas in the breast. Am J Surg Pathol. 2014 Mar;38(3):383-8. https://doi.org/10.1097/PAS.0000000000000115.

ID 905
DISCORDANCE OF HER-2 AND KI-67 BETWEEN PRIMARY BREAST CANCER AND AXILLARY LYMPH NODE METASTASIS

A. Giorlandino[1], C. Trovato[1], I. Tornabene[1], G. Leone[1], F. Motta[1], L. Gullotti[1], L. Falsaperna[1], G. Falzone[1], G. Spoto[1], A. Rizzo[1]

[1] Unit of Anatomic Pathology, Humanitas Istituto Clinico Catanese, Misterbianco (CT), Italy

Objectives: The aim of this study is to assess the preoperative incidence of discordant immunoistochemical biomarkers status in primary tumor (PM) and axillary lymph node (ALN) metastasis and to evaluate the role of ALN biopsies for the reassessment ofpreoperative neoadiuvant chemotherapy strategies of patients with breast cancer and synchronous ALN metastasis.
Material and methods. 140 cases of both breast cancer and lymph node core biopsies(14 G) over 12 months were investigated in our Department. Pathology slides were rewieved for evaluation of the immunohistochemical stain of breast tumors prognostic and predictive factors. The purpose is to establish the diagnostic discordance between primary breast cancer and synchronous ALN metastasis in order to establish the most appropriate neoadiuvant therapy to the patient.
Results: Our study showed a divergent expression of HER2 and Ki67 in 8/140 cases (5,7%). Among them, 2 cases have low expression or negative (score 0-1+ ASCO-CAP)in the PM but overexpression (score 3+ ASCO-CAP) in ALN metastasis and 6 cases have score 0/1+ ASCO-CAP in the PM and score 2+ ASCO-CAP in ALN metastasis with no amplification results on FISH evaluation. 2 cases showed strongly discordance in Ki67 nuclear expression between primary tumors and ALN metastasis.
Discussion. Some retrospective studies showed significant discordance in human epidermal growth factor receptor-2 and Ki-67 expression between primary and metastatic tumors on surgical specimen. Synchronous assessment of the breast primary tumors and matched ALN metastasis could therefore provide further insight into the required therapy type

and modify clinical outcomes. The discrepancies observed in our study may be clinically relevant in the neoadjuvant setting. All this allows a better characterization of intratumoral heterogeneity and may be important in case of complete pathological response to therapy. In addition, the evaluation of immunoistochemical biomarkers status in ALN metastasis could offer an opportunity to capture cellular subpopulations with more aggressive behaviour than those of the primary tumors.

ID 920
TRIPLE NEGATIVE BREAST CANCER BIOMOLECULAR PROFILE: IDENTIFICATION OF PROGNOSTIC AND PREDICTIVE FACTORS FOR RESPONSE TO TREATMENT

V. Zuccalà[1], N. Staropoli[2], C. Labanca [2], V. Farenza[2], L. Maltese[1], M. Renne[4], A.M. Lavecchia[1], P. Tassone[3], L. Tucci[1], P.Tagliaferri[2]

[1] Pathological Anatomy Division A.O.P.C. Catanzaro [2] Medical and [3] Translational Medical Oncology Unit, Department of Experimental and Clinical Medicine, Magna Græcia University, Catanzaro; [4] Surgery Unit Department of Experimental and Clinical Medicine, Magna Græcia University Catanzaro

Objectives: Triple negative breast cancer (TNBC) is a specific subtype of breast cancer that does not express estrogen receptor (ER), progesterone receptor (PR), or human epidermal growth factor receptor 2 (HER-2). Our research project is focused on identification of biomarkers able to better define TNBC characteristics aimed to develop new TNBC treatment strategies.
Materials and Methods: A retrospective evaluation on triple negative cases, has been developed in the last three years. Together with a clinical oncologist we selected 16 TNBC patients. For 11 of these 16 physician knew germinal mutational BRCA1/2 status. 4 patients were BRCA1/2 germline mutated. We collected a tissue sample for 5 of these 11 patients to detect NGS major cellularity and somatic mutation BRCA1- 2 state (paraffin material). Selection was blinded (just physician was aware of germinal mutational BRCA1/2 status of patients). Somatic mutational status of BRCA 1 e 2 by next-generation-sequencing (NGS) method was evaluated on section cut at 10 microns on non-polarized slide where the most cellular areas were identified and delimited. Subsequently the somatic mutational status of BRCA 1 and 2 was compared with the germinal mutational status.
Results: At the end of this first sequencing analysis, we found that 5 patients had both somatic and germline BRCA1/2 mutations. Interestingly was about two young and treatment responders patient that evaluated for BRCA 1-2 was wild type in germline but analysed in somatic they resulted BRCA mutated. Another important data was represented by determination of many pathogenetic mutations without significant allelic frequency in patients treated with one or more chemotherapy regimen for advanced breast cancer.
Conclusions: As just reported in the literature, our data help us to suppose that chemotherapy is involved in BRCA acquired mutations. This supposition born from a proof of concept evaluation; it's necessary an adequate analysis to confirm our hypothesis.

ID 929
TRIPLE NEGATIVE BREAST CANCER. A TISSUE MICROARRAY STUDY OF CXCR4 AND CORRELATION WITH MICROENVIRONMENT, P65, SRC, ERK, AKT

C. Pizzulli[1], R. Barile[1], M.C. Pedicillo[1], I.S. De Stefano[1], F. Collina[2], M.R. Brogna[2], D. Iervolino[2], V. Di Lauro[2], M. Di Bonito[2], G. Pannone[1]

[1] University of Foggia; Italy; Foggia; [2] IRCCS Fondazione G Pascale, Napoli

Triple-negative breast cancer (TNBC) is a highly metastatic cancer with poor prognosis. High expression of CXCR4 chemokine receptors and its CXCL12 ligand has beendetected in a wide range of cancers. CXCR4 is required for migration of breast cancer cells to other sites such as lung, bone, and lymph nodes, which express high levels of CXCL12 chemokine. Aim of this work is to evaluate association among CXCR4 in patients with TNBC, PFS and OS, and to found a relationship with different signaling pathways such as p65, Src, AKT, ERK, and a possible association with tumor microenvironment in order to identify targets to increase effectiveness of therapy. Analysis have made on TMA of TNBC using the CellProfiler image analysis, and SPSS statistical software. It has been identified a statistically significant association withparameters T, N and M ($p < 0.05$), and this would explain the significant difference in terms of PFS and OS in patients with high expression of CXCR4. Furthermore, astatistically significant association between CXCR4 and nuclear positivity of p65 and ERK with AKT has been identified ($p < 0.05$). Up-regulation of CXCR4 directly correlated with Src overexpression, an upstream activator of both pathways PI3K/Akt and Ras/Raf/ER, whereas, CXCR4 low levels showed inverse relationship with TIL in tissue microenvironment. We also identified a significant association among CXCR4 expression, M2 macrophages and fibroblasts ($p < 0.05$). Furthermore, our research identified an association between CXCR4 expression and taxanes, most likely because taxanes tend to overexpress p65. Since CXCR4 leads to a hypoxic microenvironment, we hypothesized that it could be associated with less efficacy of radiotherapy. This point has been confirmed in current literature. Potential drugs have been identified to be used in combi-

nation with standard therapy either directly blocking CXCR4 or blocking downstream pathways, enhancing the action of chemo-therapeutics, immunotherapy and radiotherapy since from literature analysis they have been revealed cytostatic in TNBC.

ID 946
MORPHOLOGIC AND IMMUNOHISTOCHEMICAL FEATURES OF TRIPLE NEGATIVE BREAST CANCERS

R. Lobrano[1], P. Paliogiannis[2,3], M. A. Piras[3], V. Marras[3], A. Deiana[3] M. A. Fedeli[3], A. Fara[3], A. Cossu[2,3]

[1] Department of Pathology, University Hospital (A.O.U.) of Cagliari, Via Ospedale 46,09123, Cagliari, Italy; [2] Department of Medicine, Surgery and Pharmacy, University of Sassari, Viale San Pietro 43, 07100, Sassari, Italy; [3] Anatomic Pathology and Histology, University Hospital (A.O.U.) of Sassari, Via Matteotti 60, 07100, Sassari, Italy

Objectives: In the present study the immunohistochemical features of triple negative breast cancers (TNBC), in relation with their histopathological characteristics were investigated.
Materials and methods: Consecutive invasive breast cancer cases pathologically diagnosed at the Anatomic Pathology and Histology Unit of the University of Sassari from 2005 through 2013 were enrolled in the study; in situ tumors have been excluded. All the included cases were re-classified in accordance with the Saint Gallen criteria. TNBCs were defined as tumors with synchronous lack of immunohistochemistry expression of ER, PR and HER2. Immunohistochemistry was performed with Ventana BenchMark Ultra system with the following antibodies: E-cadherin, Ki67, Cytokeratin 17, Cytokeratin 14, Cytokeratin 5/6, MUC1, Androgen receptors, IgF1R, p53, Claudin, VEGF, and PD-L1.
Results: Globally, 2572 cases with a mean age of 59 years were evaluated, and among them 199 (8%) were identified as TNBC, but four were excluded because they did not fulfil the inclusion criteria. Therefore, 195 TNBC patients have been analysed. At the time of diagnosis, most of them had a ductal NOS breast carcinoma (DBC, 109, 55.8%), 112 (57.4%) of them had a T2 or greater disease stage, but 108 (52.8%) had no axillary lymph node involvement. The grade of the disease was G3 in 146 (74.8%) cases. Based on the expression of the biomarkers tested, seven TNBC subtypes have been identified: cytokeratin negative DBC, basal-like DBC, apocrine cancers, medullary cancers, pleomorphic lobular cancers, anaplastic cancers, and metaplastic tumors.
Conclusions: TNBC show multiple morphological and immunohistochemical features, which characterize specific clusters with different biological and clinical behaviour. Knowledge of these features are essential for correct diagnosis and treatment of patients with TNBC.

Giovedì 13 Ottobre 2022

Sala F 08.15 - 09.05

CITOLOGIA

Moderatori: G. Fadda, K. Ludwig

ID 750
VICTORIA'S CELLS
THE VISUAL LANGUAGE OF CELLS

S. Cartesio, V. Lombardo, G. Fadda, E.D. Rossi

Unicatt, Asp ME, Unicatt

Objectives: Victoria's cells project was created with the humanitarian mission of facilitating the training of health personnel, cytoscreeners, in disadvantaged countries where prevention is absent and there is an extremely high rate of sexually transmitted diseases (85%).
This innovative method allows to associate cellular images and/or patterns characterized by a wide range of shapes and color shades, evoking animals, common objects, colorful aquariums with features easily memorized by analogy, and interpreted as benign and/or malignant under the microscope.
Materials and methods: Cervico-vaginal cytology with the Papanicolau staining processed with conventional and liquid based cytology (LBC).
Results: The images are visual of impact that communicate and educate about the importance of studying cells and their diagnostic role. Infectious diseases are easily recognize as well as granulocytes, haemocytes and even cytolysis. The budding of mycetes resembles a starfish, or squamous metaplasia recalls the carapace from the sea turtle. We can also identify fish tanks populated by pufferfish made up of endometrial cells looking curiously at an anglerfish with its long dorsal fin of keratinized parabasal cells, but also a sperm whale of granulocytes, and a garfish of mucous striae. A huge manta ray of endometrial cells and a small tracina of only two parabasal paracheratotic cells or a jellyfish of granulocytes and two goldfish of keratinized cells. Thus, 3-D sly cat of endometrial cells, or a tender little elephant of squamous cells or even a dog and a koal of keratinized cells. Additionally, a crab-like of metaplastic cells, scorpion of cluster of basal/parabasal cells with granulocytes. Other preparations resemble a gymnast, a geisha of partly keratinized basal squamous cells or a plunging diver of granulocytes.
On LBC, a hummingbird of endometrial cells soars in flight, while a heron of immature metaplasia, with also endocervical cells mimicking water lily and peony with a dancer's foot of a blood clot mixed with keratinized squamous cells. SIL pattern looks like monsters, eyes, or a foul tongue, whilst AGC resembles an eagle, and feathers.

Conclusions: The recognition of visual images can make the study of cytology simpler and enjoyable leading to the final purpose of prevention and cure.

ID 751
WHAT'S ABOUT THE ROLE OF PD-L1 IN ONCOCYTIC THYROID LESIONS ON CYTOLOGICAL SAMPLES

P. Tralongo, M. Dell'Aquila, F. Policardo, A. Granitto, M. Curatolo, V. Fiorentino, T. Musarra, F. Pierconti, L.M. Larocca, E.D. Rossi

Division of Anatomic Pathology and Histology-Fondazione Policlinico Universitario"A. Gemelli"-IRCCS, Rome Italy

Objectives: The role of PD-L1 expression as a relevant predictive biomarker in anti-PD-L1 cancer immunotherapy has been shown in several papers. However, a very scant literature hasdealt with PD-L1 in thyroid fine needle aspiration cytology (FNAC), demonstrating a possible diagnostic and prognostic correlation with papillary thyroid carcinoma (PTC). Our group studied the role of PD-L1 in thyroid follicular lesions with conclusive Results: However, its role in oncocytic thyroid lesions remains controversial, showing a weak expression in some of them. Based on this evidence, we accordingly evaluated the performance of PD-L1 immunostaining in liquid-based cytology (LBC) from oncocytic lesions.
Material and methods: From January 2019 to March 2021, 63 thyroid lesions diagnosed by FNAC from lesions with a predominant oncocytic component were enrolled for evaluation by PD-L1 immunostaining on both LBC and corresponding histology samples.
Results: We also included 51 benign (B, negative controls). The oncocytic series was composed of 4 Atypia of undetermined significance/Follicular lesions of undetermined significance (AUS/FLUS) with oncocytic prevalence, 57 follicular lesions (FN/SFN) with oncocytic pattern, and 2 suspicious for malignancy (SFM) cases. Forty-three cases (2 AUS/FLUS, 39 FN/SFN and 2 SFM) had histological follow-up including: 2 AUS/FLUS cases as oncocytic adenomas (OAs); 39 FN/SFN included 27 OAs, 4 FA, and 8 oncocytic follicular carcinoma (OFC). The 2 SFM cases were diagnosed on histopathology as OAs. Increased plasma membrane and cytoplasmic PD-L1 expression was found in 47 cases of the LBC cases (41.2%). Among the histological series, 67.3% of OAs and 75 % of OFC had PD-L1 expression, whilst negative PD-L1 was found in hyperplastic oncocytic cells in HT. A positivity in more than 30% of the neoplastic cells was found in 72.9% of the cases including six OFC.
Conclusions: These data suggest that PD-L1 expression is expressed in oncocytic thyroid lesions. While weak PD-L1 expression failed to discriminate benign from malignant lesions, OFC demonstrated more intense cytoplasmic and membranous expression.

ID 765
P53 EXPRESSION IN THYROID CYTOLOGICAL SAMPLES. A NEW MARKER FOR THE EARLY STAGE OF THYROID TUMOR?

F. Policardo, P. Tralongo, M. Curatolo, Q. Zhang, F. Vegni, A. Feraco, L. Cardisciani, S.Cartesio, L.J. Marin Torres, E.D. Rossi

Division of Anatomic Pathology and Histology Fondazione Policlinico Universitario"A.Gemelli"-IRCCS, Rome Italy

Objectives: TP53 mutation seems to play a role in the malignant transformation of thyroid cells as well as thyroid tumor progression(TTP). TP53 mutation has been assessed in poorly(PDTC) and undifferentiated thyroid carcinoma(UTC), whilst only in few well differentiated thyroid carcinoma (WDTC) and absent in benign conditions. This data suggested that mutational inactivation of TP53 is a late stage of TTP. P53 detection is likely to be a significant and independent prognostic factor in WDTC. We investigate the role of p53 on thyroid cytology.
Material and methods: One hundred prospective cyto-histological samples diagnosed from categories III(atypia of undeterminate significance-AUS) to VI(malignant-M) were studied with the p53 antibody. Its intensity was graded from 0(negative) to 3+(strong). We correlate p53 with HBME-1.
Results: The cytological diagnoses were 44AUS, 24FN, 23SFM, 1M. Among them 17 had histological follow-up(1AUS, 6FN, 9SFM, 1M). We reported 68 cases with p53 negative and 32 positive cases. Among the latter group, only 9 cases documented a weak and focal cytoplasmic positivity(+1), whilst 21 cases had a moderate(2+) to strong(3+) cytoplasmic/nuclear expression. Considering the cyto-histological series, 12 cases were negative for p53 including 8 classic PTC. The remaining 5 positive cases had a 3+ p53 nuclear expression corresponding to more aggressive types of PTC(tall-TCV, columnar, solid types). 93% AUS and 83.3% FN were p53 negative and none of them had malignant histology. Among the indeterminate lesions, 8 HBME-1 positive cases(3AUS and 5FN) had a negative p53 and a benign histology. Among the SFM, 50% were p53 and HBME-1 positive with a malignant histology. All M cases were p53, with a larger size(> 1.5cm), TCV subtype multifocality and extrathyroidal infiltration, 50% nodal metastases.
Conclusions: P53 might be useful in discerning different groups of thyroid follicular lesions. P53 is likely to be a diagnostic marker useful in characterizing WDTC as PTC. The use of a panel made up of p53 and HBME-1 suggests a possible correlation between their expression and more aggressive PTC subtypes.

ID 767
IMPLEMENTATION OF THE MILAN SYSTEM IN A CYTOPATHOLOGIST-PERFORMED US-GUIDED FNA PRACTICE

A. M. Carillo[1], P. Pisapia[1], E. Vigliar[1], G. Troncone[1], C. Bellevicine[1]

[1]Department of Public Health, University of Naples Federico II, Naples, Italy

Background: The Milan System for Reporting Salivary Gland Cytopathology (MSRSGC) aims to standardize reporting terminology of salivary glands FNAs. In our practice, salivary gland US-FNAs are performed by the interventional cytopathologist. The aim of this study is to evaluate the added value of the MSRSGC implementation in our pathologist-performed US-FNA practice at University of Naples "Federico II" and determine both the risk of neoplasm (RON) and malignancy (ROM) of each category.
Methods: Our database was searched for salivary gland FNA performed from January 2018 to July 2021 and classified by the MSRSGC. All FNAs were performed with 23-gauge needles. From each sample at least one smear was air dried and stained with Diff-Quik to allow the rapid on-site evaluation (ROSE). In selected patients, one or more additional pass were performed to obtain either Papanicolaou stained smears or cell-block for ancillary stainings.
Results: A total of 538 salivary gland FNA specimens were retrieved with a male predominance (M:F = 1,1:1) and a mean age of 59 years. The localization of the lesions was reported in the majority of the cases (487/538) and the majority of FNAs, (N = 409) were performed in the parotid glands. Based on MSRSGC, 93 cases were classified as I, 87 as II, 72 as III, 210 as IVa, 35 as IVb, 15 as V and 26 as VI. In199 out of538 FNAs the histological follow-up was available: o the RON was 36,4% for I category, 18,2% for II, 69,6% for III, 100% for IVa, 95,2% for IVb, 100% for V and 100% for VI, while ROM was 3% for I, 0 for II, 21,7% for III, 0 for IVa, 19% for IVb, 71,4% for V and 91,7% for VI category.
Conclusions: The MSRSGC coupled with US-FNA performed by interventional cytopathologist show a high diagnostic yield and is useful to standardize the diagnostic reporting. Most of FNAs in our series were classified as IVa which featured a RON of 100% with no malignant neoplasm retrieved. This result may allow a tailored surgical management for patients with FNA classified as IVA, avoiding extensive resections.

ID 870
CYTOMATRIX VS CELL-BLOCK EVALUATION ON FNC IN NODAL METASTASIS BY MELANOMA

M. Montella[1], A. Ronchi[1], E. Clery[1], G. D'Abbronzo[1], S. Lucà[1], E. Carraturo[1], R. Franco[1], I. Cozzolino[1].

[1] Unit of Anatomic Pathology, Department of Mental and Physical Health and Preventive Medicine, University of Campania "Luigi Vanvitelli", Naples, Italy

Objectives: Fine-needle cytology (FNC) is often used for the pre-operative diagnosis of melanoma metastases. The diagnosis cannot be established relying just on morphology evaluation, and immunocytochemistry is often mandatory (1). In our experience, the cell-block material is an excellent technique to perform qualitatively satisfactory immunocytochemical analysis. The Cyto Matrix® (CM) is a novel synthetic support that is giving excellent results in the preparation of cyto-inclusions. Our aim is to compare the Shandon™ Cytoblock™Cell Block Preparation System (CB) and the CM techniques in the preparation of a cyto-inclusions, comparing morphological and immunocytochemical yield.
Materials and methods: FNC was performed on 22 lymph nodes of patients with cutaneous melanoma diagnosis, for each of whom both a CB and a CM were set up. Both preparations were compared for adequacy, morphologic evaluation and immunocytochemistry adequacy for S100, MELAN-A, HMB-45, SOX10 and PRAME.
Results: The results obtained by the two methods were quite comparable. The adequacy of the CM compared to the CB was slightly better (100% vs 82%), while the morphological detail was better interpreted on the CB (45% vs 40%). The evaluation of S100 was adequate in 77% of the CBs and in 91% of the CMs, SOX10 was adequate in 82% of the CBs and in 100% of the CMs; MELAN-A was adequate in 64% of CBs and in 100% of CMs; HMB-45 was adequate in 73% of CBs and 90% of CMs; PRAME was adequate in 73% of CBs and in 100% of CMs.
Conclusions: In our experience, CM is a valid technique on which to perform immunocytochemical investigations for the evaluation of diagnostic markers.

References

Ronchi A, Zito Marino F, Toni G, Pagliuca F, Russo D, Signoriello G, Moscarella E, Brancaccio G, Argenziano G, Franco R, Cozzolino I. Diagnostic performance of melanocytic markers for immunocytochemical evaluation of lymph-node melanoma metastases on cytological samples. J Clin Pathol. 2022 Jan;75(1):45-49. https://doi.org/10.1136/jclinpath-2020-206962.

Venerdì 14 Ottobre 2022

Sala Mantegna 08.00-09.10

PATOLOGIA APPARATO DIGERENTE 2

Moderatori: R. Fiocca, M. Guido

ID 834
IMMUNOHISTOCHEMICAL ANALYSIS OF ARID1A EXPRESSION IN COLON CANCER

R. Gafà[1], V. Tagliatti[1], E. Degani[1,2], I. Maestri[1], G. Lanza[1]

[1] Anatomic Pathology Unit, University Hospital of Ferrara - Department of Translational Medicine, University of Ferrara, Ferrara, Italy. [2] University of Bologna, Bologna

Objectives: The AT-rich interaction domain 1A (ARID1A) gene, member of the SWItch/sucrose non fermentation (SWI/SNF) chromatin remodeling complex, is frequently mutated in several types of tumors. ARID1A variations have been demonstrated in a variable proportion of colorectal cancers (CRC) but the specific role and the prognostic value of ARID1A in CRC is still controversial.
Aim of the present study was to investigate ARID1A expression in a series of colon adenocarcinomas (CADs) to evaluate the relationship between ARID1A status and clinicopathologic and molecular parameters.
Materials and methods: ARID1A expression was investigated by immunohistochemistry in 108 CADs using FFPE tumor samples and a rabbit monoclonal antibody. DNA mismatch repair (MMR) status was determined by immunohistochemical analysis of MLH1, MSH2, MSH6 and PMS2 proteins expression and by microsatellite instability analysis.
Results: Most CADs (94, 87%) showed intense nuclear staining of nearly all neoplastic cells and were classified as ARID1A positive. Complete or partial ARID1A loss of expression was demonstrated in 14 tumors (13%). ARID1A loss was more frequent in older patients and in carcinomas arising in the proximal colon. A significant relationship was observed between ARID1A expression and MMR status. Loss of ARID1A expression was detected in 10 of 45 (22.2%) MMR-deficient (MMR-D) carcinomas, but in only 4 of 63 (6.3%) MMR-proficient tumors (p = 0.021). 10 of the 14 (71.4%) ARID1A negative cases were MMR-D adenocarcinomas.
Most of the MMR-D/ARID1A negative tumors were MLH1/PMS2 negative; however, loss of ARID1A expression was also observed in two MMR-D MSH2/MSH6 negative cases.
Interestingly, 9/10 MMR-D ARID1A negative tumors were poorly differentiated and 8/10 were in advanced TNM stage (stage III).
Conclusions: Loss of ARID1A expression is detectable in a significant fraction of CADs. Loss of ARID1A protein immunoreactivity occurs more often in MMR-D carcinomas and seem to be associated with aggressive clinical and pathologic features in this tumor molecular type.

ID 854
FERROPTOSIS IN EXTRAHEPATIC CHOLANGIOCARCINOMA

D. Sacchi [1], D. De Bartolo [1], S. Sarcognato [1], I. Cataldo[1], F. Baciorri[1], V. Maffeis[1], S. Brignola[1], M. Cacciatore[1], A. Furlanetto[1], M. Guido[1,2]

[1] Pathology Department ULSS2, Treviso Hospital, Treviso, Italy; [2] Department of Medicine-DIMED, University of Padova, Padova, Italy

Objectives: Extrahepatic cholangiocarcinoma (eCCA) is an aggressive tumor with a poor prognosis,due to the absence of specific systemic treatments. A pro-inflammatory and anti-apoptotic microenvironment has been described in some eCCAs. Ferroptosis is a novel type of highly regulated iron-dependent cell death with a well-known role in carcinogenesis. Triggering ferroptosis in cancer therapy is a promising option. Nothing is known about ferroptosis in eCCA. Therefore, the aim of this study was to investigate tissue expression of ferroptosis effectors in eCCA, and any correlation with prognostic and histological features.
Design. A series of 45 consecutive eCCA were enrolled and histologically reviewed; the main histological features were recorded. None of the patients received any neoadjuvant therapy before surgery. Immunostaining was performed on tissue microarrays, by using the following antibodies: TFR1 (Transferrin-Receptor 1), GPX4 (the main ferroptosis inhibitor), STAT3 (as a marker of an anti-apoptotic milieu). Intracytoplasmic iron deposits were investigated by Perls' stain.
Results: A complete negativity for TFR1 was observed in 98%; none of the patients showed intracytoplasmic iron deposits in neoplastic cells. In 80% of cases GPX4 expression was absent. In 18% of cases a low GPX4-expression (< 50% positive neoplastic cells) was found; however, there was no correlation with prognosis. A high STAT3 expression in neoplastic cells was observed in 51% of cases, and it was associated with a worse prognosis (reduced overall [p = 0.001] and disease-free survival [trend, p = 0.061], and unfavorable histological features (vascular invasion, p < 0.001;perineural invasion, p = 0.001).
Conclusions: Overexpression of STAT3 confirms an anti-apoptotic background in eCCA. However, ferroptosis does not seem activated in eCCA and it seems to be not inhibited by GPX4, which is absent in most cases. This highlights the possibility to use ferroptotic drug-inducers in eCCA, such as Xc^- inhibitors and iron-overload drugs.

ID 857
MORPHOLOGICAL AND MOLECULAR CHARACTERIZATION OF COLORECTAL SERRATED POLYPS WITH INTESTINAL DYSPLASIA

F. Cappello[1], V. Angerilli[1], L. Dal Santo[1], G. Munari[1], M. Sabbadin[1], M. Lo Mele[1], G.Pennelli[1], P. Parente[2], S. Lazzi[3], M. Fassan[1]

[1] Department of Medicine (DIMED), University of Padua, Padua, Italy; [2] Unit of Pathology, Fondazione IRCCS Ospedale Casa Sollievo della Sofferenza, San Giovanni Rotondo (FG), Italy; [3] Department of Medical Biotechnology, University of Siena, Siena, Italy

Objectives: In sessile serrated lesions (SSLs) and traditional serrated adenomas (TSAs) with intestinal dysplasia, the dysplastic component and the serrated component without dysplasia should be considered as part of the same lesion, classified as SSL with dysplasia or TSA with dysplasia, respectively. However, some of these lesions may actually represent collisions between a serrated polyp and a conventional adenoma. Further supporting the "collision theory", intestinal dysplasia may be found in association with hyperplastic polyps (HPs).
Materials and methods: We collected 17 cases of colorectal serrated lesions with intestinaldysplasia, classifying them as LSS with dysplasia (n = 10) or as mixed lesions comprising a HP component and a conventional adenomatous component (n = 7). We characterized the dysplastic and the non-dysplastic component of each lesion, after microdissection, through the targeted mutational analysis of 11 most commonly altered genes in colorectal cancer (*AKT1, APC, BRAF, CTNNB1, KIT, KRAS, NRAS, PDGFRA, PIK3CA, PTEN* and *TP53*). We also characterized MMR and p53 status by immunohistochemistry.
Results: 14/17 (82.4%) cases harbored a mutation in at least one of the two components. The most altered genes were *BRAF* in 10/17 (58.8%) cases, *APC* in 2/17 (11.8%) and *TP53* in 4/17 (23.5%). Among the SSL with dysplasia, the mutational profile was concordant between the two components in 7/10 (70%) cases, while among the mixed lesions, the mutational profile was concordant in 1/7 (14.3%). In all the cases MMR status was concordant between the two components of the serrated lesions.
Conclusions: Our findings suggest that intestinal dysplasia may develop in SSL as part of the serrated lesion, even if some SSL with dysplasia may actually be collision lesions. On the other hand, the polyps that are morphologically classifiable as mixed lesions composed of a HP and a conventional adenomatous component are more likely to be collision lesions.

ID 861
GENOMIC PROFILING OF COLORECTAL ADENOSQUAMOUS CARCINOMA IS SIMILAR TO THAT OF THE NOT-OTHERWISE SPECIFIED HISTOTYPE

G. Businello[1], P. Parente[2], C. Angerilli[1], L. Mastracci[3], A. Vanoli[4], M. Paudice[5], M. Castelvetere[2], P. Graziano[2], F. Grillo[3], M. Fassan[1,6]

[1] Department of Medicine (DIMED), Surgical Pathology & Cytopathology Unit, University of Padua, Padua, Italy; [2] Fondazione IRCCS Ospedale Casa Sollievo della Sofferenza, Unit of Pathology, San Giovanni Rotondo, Italy; [3] Ospedale Policlinico San Martino IRCCS, Genova, Italy; Department of Surgical Sciences and Integrated Diagnostics (DISC), Anatomic Pathology, University of Genova, Genova, Italy; [4] Department of Molecular Medicine, Anatomic Pathology Unit, University of Pavia and Fondazione IRCCS San Matteo Hospital, Pavia, Italy; [5] Department of Surgical Sciences and Integrated Diagnostics (DISC), Anatomic Pathology, University of Genova, Genova, Italy; [6] Istituto Oncologico Veneto - IOV-IRCCS, Padua, Italy

Objectives: Colorectal adenosquamous carcinoma (ASC) is exceedingly rare, comprising less than 0.1% of all colorectal malignancies, and is characterized by an aggressive disease course, with a higher metastatic rate and worse outcome than conventional colorectal carcinoma. A comprehensive molecular profile of this group of neoplasms is still lacking.
Materials and methods: Genomic characterization of 24 cases of colorectal ASCs (23 primary tumors and one liver metastasis) and 2 cases of colorectal high-grade tubulo-villous adenoma (HG TVA with squamoid morules was subject to NGS targeting 67 cancer-related genes (VariantPlex Solid Tumor; Archer).
Results: In 22 of 24 (91.67%) ASC samples and in one of the two (50%) HG TVAs with squamoid morules at least one single nucleotide variant (SNV) or copy number variation (CNV) was detected. The most frequently mutated genes were *TP53* (50%), *APC* (42.31%), *KRAS* (34.62%),*BRAF* (15.38%) and *GNAS* (11.54%). The following CNVs were observed: *PTEN* loss (3.8%), *PIK3CA* loss (3.8%), *ERBB2* loss (3.8%), *NOTCH1* gain (3.8%), *ERBB4* loss (3.8%).
Conclusions: This study sheds light on the molecular landscape of colorectal ASCs. According to our data, the genomic profile of colorectal ASC is similar to that of conventional colorectal carcinoma, with significant druggable genetic alterations. Further studies are required to understand the more aggressive clinical behavior of this neoplasm.

ID 904
ORDINARY COLORECTAL CARCINOMA EXPRESSING SYNAPTOPHYSIN (OCCES): PROGNOSTIC IMPACT OF A NEW ENTITY

G. Sabella[1], G. Centonze[1], F. Grillo[2]; V. Lagano[1], M. Guaglio[1], M. Fassan[3], S. La Rosa[4], F.Sessa[4]; C. Capella[4], M. Milione[1]

[1] Istituto Nazionale Tumori Milano; [2] University of Genova;[3] University of Padua; [4] Uni-Insubria

Objectives: Although colorectal carcinoma (CRC) is extremely common, mixed tumors composed of exocrine and neuroendocrine (NE) elements are rare (< 2% of all colorectal malignancies). Theseheterogeneous neoplasms include poorly known categories of amphicrine tumors, where non-NEand NE phenotype is synchronously expressed within the same cell. Although NE differentiation in CRCs has been extensively assessed, no clear definition nor established criteria are currently available for OCCES.
Material and Methods: We investigated Synaptophysin (SYN) expression in 663 ordinary CRCand correlated the results with clinicopathological and molecular features and compared the survival characteristics of SYN positivity > 30% group to those of 14 MANECs. These data were correlated with Overall Survival (OS) and Disease-free Survival (DFS).
Results: Diagnosis of OCCES was confirmed in 27 patients and correlated with right colon (p = 0.003), Grade 2 (p = 0.0007), marked intratumoral lymphocyte infiltrate (TIL) (p = 0.0006), and *BRAF* mutation (p = 0.04). At univariate analysis variables associated with poor OS were 10-year increase in age (p = 0.001), stage IIII-IV (p = 0.001), OCCES status (p = 0.001), infiltrative growth at the invasive edge (p = 0.04), and residual tumor R1-2 (p = 0.03), while marked peritumoral TIL was associated with longer OS (p = 0.04). At multivariable analysis only 10-year increase in age, stage III-IV, and OCCES status (p < 0.001) remained associated with poor OS and marked peritumoral TIL with longer OS (p = 0.02). Comparable results were obtained with the same variables for DFS. In terms of OS and DFS, Syn expression in > 30% of gland forming tumor cells proved to be an independent negative prognostic factor. Patients with MANECs, on the other hand, showed a significantly shorter DFS than all conventional adenocarcinomas with or without SYN expression in univariate analyses (p < 0,001; HR: 5.04)
Conclusion: Our study investigated the clinicopathological and molecular features of an unexplored entity of CRC with variable NE differentiation providing the close correlation between prognostic impact of CRC and morphological and immunohistochemical phenotype.

ID 921
KI-67 EXPRESSION, MAST CELLS POSITIVE TO TRYPTASE AND ANGIOGENESIS IN GASTRIC CANCER PATIENTS UNDERGOING RADICAL SURGERY
GASTROINTESTINAL PATHOLOGY

A.M. Lavecchia[1], V. Zuccalà[1] M. Ammendola[2], C. Gentile [1], A.V. Filardo[1], L. Maltese[1], G. Funaro[1], G. Currò [2], L. Tucci[1]

[1]Pathological Anatomy Division A.O.P.C. Catanzaro; [2]Department of Health Science, Digestive Surgery Unit, University "Magna Graecia", Catanzaro

Objectives. Angiogenesis is a hallmark of cancer and several solid tumors are dependent on neoangiogenesis for their growth and proliferation. The cells present in tumor microenvironment such as mast cells and microvascular density (MVD) could play a pro-angiogenetic role.
Methods. A series of 85 gastric cancer (GC) tissue samples from patients undergoing radical surgery was evaluated for Ki-67 proliferation rate, number of mast cells positive to tryptase (MCPT) and MVD in tumor tissue (TT) and in adjacent normal tissue (ANT) through immunohistochemistry and image analysis. Statistical analysis was performed through Pearson t-test.
Results. A significant correlation in tumor tissue (TT) between Ki-67 expression, MCPT and MVD was found (p ranged from 0.01 to 0.03). A statistically significant difference of Ki-67 expression, MCPT and MVD between TT and adjacent normal tissue (ANT) was found (p ranged from 0,01 to 0,03).
Conclusions. The assessment of tumor angiogenesis, a process related to cancer growth and metastasis, may be useful in prognostic prediction. Our data regarding tumor microenvironment suggest that the increase of Ki-67 proliferation rate correlates with a greater expression of MCPT and MVD. For these reasons, Ki-67 expression and MVD may indicate the survival prognosis of patients and MCPT could represent a biological marker of radical surgery and angiogenesis. Furthermore, MCPT could be considered as a target of novel anti-angiogenic therapies in GC patients.
Key words: Tumor Proliferation; Angiogenesis; Mast Cells; Tumor Microenvironment; Gastric Cancer; Surgical Oncology

ID 947
A COMBINED CLINICAL-PATHOLOGICAL SCORE TO PREDICT THE RISK OF LYMPH NODE METASTASES IN PT1 CRC

G.M. Benazzo[1], A. Gambella[1], E.C. Falco[1], L. Bertero[1], P. Cassoni[1]

[1] Pathology Unit, Department of Medical Sciences, University of Turin, Turin, Italy

Objectives: The management of patients with early colorectal cancer (pT1 CRC) is based on nodal metastases risk estimation, carried out considering specific histopathological parameters. The current surgical overtreatment of patients with no metastatic disease makes it necessary to redefine this risk, to improve clinical-therapeutic management. This study proposes a combined clinical-pathological score to improve the stratification of the risk of lymph node metastasis.

Materials and methods: The clinical-demographic and anatomical-pathological data of a retrospective consecutive series of 207 cases of pT1 CRC treated with colectomy and locoregional lymphadenectomy were retrieved at the Città della Salute e della Scienza Hospital of Turin between January 2010 and March 2019. Histopathological parameters were re-evaluated on the original histological slides by three pathologists experienced in the gastroenteric tract.
Results: Of the 207 cases studied, 18 (8.7%) have lymph node metastases. The clinical and pathological parameters significantly associated with this event were lymphovascular invasion (OR:23.8; CI: 5.12-110.9) and high-grade tumor budding (OR: 5.21; CI: 1.60-16.8), correlated with an increased risk of lymph node metastases, while age at diagnosis > 65 years (OR: 0.26; CI: 0.09-0.71) and a high degree of intratumor lymphocytes (OR: 0.19; CI: 0.06-0.59) showed a protective effect. Combining these characteristics, we built a combined five-tier risk score that, applied to our series, significantly identified cases with a higher-risk (score ≥ 2) of lymph node metastases (OR:7.7; CI: 2.4-24.4).
Conclusions: In conclusion, we have developed a combined score effective in improving the stratification of pT1 CRC based on the risk of lymph node metastases. We believe that its employment in a multidisciplinary pT1 unit could improve patients' clinical management and limit surgical overtreatment.

Venerdì 14 Ottobre 2022

Sala A 08.30 - 09.10

UROPATOLOGIA 2

Moderatore: A. Caliò

ID 878
PSEUDOMESOTHELIOMATOUS METASTASES OF FUMARATE HYDRATASE DEFICIENT RENAL CELL CARCINOMA: A DIAGNOSTIC PITFALL

E. Bariani[1], A. Caliò[1], S. Marletta[1,2], G. Serio[3], L. Stefanizzi[2], L. Marcolini[2], P. Antonini[1], M. Brunelli[1], G. Martignoni[1,2]

[1] Department of Diagnostic and Public Health, Section of Pathology, University of Verona, Italy; [2] Department of Pathology, Pederzoli Hospital, Peschiera del Garda, Italy; [3] Department of Emergency and Organ Transplantation, Pathology Unit, University of Bari, Italy

Objectives: Diagnosis of peritoneal tumors may be challenging. Unspecific histological features could cause great difficulties in distinguishing between primary reactive or neoplastic processes and metastatic localizations when only relying on morphological findings. This is of concern especially when metastatic diseases firstly show up with peritoneal involvement.
Materials and methods: Herein we report three cases of metastatic peritoneal nodules mimicking mesothelial proliferations.
Results: Two patients were males, one female. Tumors were made up of cells arranged in microcystic to papillary architecture with clear to eosinophilic cytoplasm, round nuclei and prominent nucleoli, initially suspicious for a reactive mesothelial process or peritoneal mesothelioma. The neoplastic cells immunoexpressed CK8-18 and CKAE1/AE3 but were negative for mesothelial markers (CK5/6, calretinin, and WT1). Nuclear labelling for PAX8 suggested the possibility of a metastasis from a renal neoplasm, a previous history for which was known in only one case. Immunohistochemical loss of fumarate hydratase (FH) leaded to the final diagnosis of localizations of FH deficient renal cell carcinoma.
Conclusions: Proper classification of peritoneal nodules can be extremely difficult, especially in the intraoperative setting, as primary lesions may morphologically closely resemble metastases. Among these latter ones, FH deficient renal cell carcinomas should be considered. In our series of FH deficient renal cell carcinomas, nine on twelve patients (75%) displayed aggressive clinical behaviorwith local recurrence and/or metastases, involving the peritoneum in a noteworthy proportion of the cases (3/12, 25%) where they can be misdiagnosed for a mesothelial process. Thus, pathologists should be aware of such a pitfall and include PAX8 and FH in immunohistochemical panels for cases with controversial findings.

ID 881
BASAL PHENOTYPE AS PECULIAR FEATURE OF YOUNG UROTHELIAL CARCINOMA OF THE BLADDER: A COHORT ANALYSIS

M. Iuzzolino[1,2], M. Valeri[1,2], C. De Carlo[1,2], A. Bressan[1,2], L. Dore[1,2], M. Cieri[1], G.M. Elefante[1], P. Colombo[1,2].

[1] Department of Pathology, IRCCS Humanitas Clinical and Research Hospital, Rozzano, Italy; [2] Humanitas University, Department of Biomedical Science, Pieve Emanuele, Italy

Objectives: Urothelial carcinoma (UC) in young adults (YA, 18-40 yrs) is an uncommon and challenging diseasefor the clinical impact of potential recurrence and progression. The recently discovered molecular phenotypes and the genomic profile are poorly understood in YA. Here we analysed a YAUC cohort to evaluate these molecular categories using an immunohistochemical score system and to search forgenomic mutations.
Materials and methods: All YAUCs diagnosed in our department between 1998 and 2021 were reviewed. We performedimmunohistochemical analysis

for Luminal and Basal markers (CD44, CK5/6, CK20, GATA3), Androgen receptor (AR), and RB1. In a subset of cases NGS analysis was also performed.
Results: Overall, 51 cases were collected. Mean age was 34yrs and most pts were male (42); 36 pts had low-grade (LG) UC, 5 were high-grade (HG), 10 pts had neoplasm of low malignant potential (PUNLMP)or urothelial papilloma (UP). All but 2 cases where pTa. Although all cases were positive for GATA3, Basal phenotype was the most common finding (44/51; 86,3%). Indeed, most cases were CD44+(43/51) and CK 5/6+ (46/51), with a predominant reactivity in basal layers, while 7 cases were CK20+, mainly in apical cells. HG tumors showed a Luminal phenotype in 3/5 cases. UP, PUNLMP and LG tumors showed a similar phenotype. AR and RB1 expression was detected in 26 and 48 cases, with a homogeneous distribution irrespective of tumor grading. NGS analysis performed in 10 pts showed *FGFR2* mutation in 4 cases, while mutations in other genes were rare (*HRAS, KRAS, PIK3CA, CDKN2A, p53*). No correlations were identified between phenotypes and recurrence during follow-up, possibly due to the limited number of events.
Conclusions: YAUC show almost exclusively a Basal molecular phenotype and wild-type RB1, although our data may be biased by the predominance of LG tumors. In contrast to previous published data, GATA3 did not act as a Luminal marker. These results should be validated on a larger cohort of YA with a deeper investigation of molecular events related to this challenging category.

ID 959
CLEAR CELL PAPILLARY RENAL CELL TUMOR HAS TO BE COMMONLY DIAGNOSED IN ROUTINE PRACTICES: IMPACT TO PATIENT COUNCELING

B. Barioglio[1], B. Dal Farra[1,5], P. Antonini[1], M. Brunelli[1], A. Caliò[1], A. Brighenti[1], A. Damuri[2], S. Francesconi[3], G. Martignoni[1,4]

[1] Department of Diagnostics and Public Health, University of Verona, Verona, Italy; [2] Anatomic Pathology Unit,"A. Perrino" Hospital - ASL, Brindisi, Italy; [3] Pathology Unit, Santa Maria di Terni,Terni, Italy; [4] Pathology Unit, Pederzoli Hospital, Peschiera del Garda,Verona, Italy; [5] Department of Diagnostics and Public Health, University of Udine, Udine, Italy

Objectives: Clear cell papillary renal cell carcinoma has been categorized by upcoming the new 2022 classification as a low grade/low malignant potential tumour. We investigated the incidence in routine diagnostics.
Materials and methods: We reviewed consecutive renal cell neoplasia diagnosed at University of Verona during last three years among enucleoresection. Immunophenotypical and molecular analysis has been provided using multi-target antibodies such as CAIX, GATA-3, CK34BE12, CK7, S100A1, parvalbumin, AMACR and PAX-8. 3p and 8q chromosomal abnormalities has been tested by FISH.
Results: 340 renal cell neoplasia has been recruited. 24 (7%) clear cell papillary renal cell carcinoma has been diagnosed and renamed clear cell renal cell tumors after morphology and immunophenotypical profiling, such as all tumours showed diffuse CK7 and 34BE12 positive expression and CAIX cup-like+, PAX-8+ with additional 14/24 GATA-3+ nuclear expression. Two tumors (1%) showed the characters of the clear cell renal cell carcinoma with abundant angio-fibroleiomiomatous stroma, by diffuse expression of both cytokeratins but CAIX complete membranous positive expression and none with GATA-3 expression. Two (1%) tumours showed TCEB-1/8q abnormality, then classified as TCEB-1 mutated renal cell carcinoma and finally excluded from the cohort. All tumorus set pT1 and revealed no recurrence or metastases after at least 3 year of follow-up. 3p locus showed no abnormalities. 8q loss is observed only in the TCEB-1 mutated renal cell carcinoma.
Conclusions: Around 10% of renal tumour enucleations reveals the new low grade/low malignant potential tumor entity clear cell papillary renal cell carcinoma (acutally renamed tumor rather than carcinoma) and other emerging entities such as TCEB-1 mutated renal cell carcinoma and renal cell carcinoma with abundant angiofibroleiomiomatous stroma. Use of CK34BE12, CK7 and GATA-3 are sensible guide for appropiate diagnostics in conjunction with morphology. Clear cell renal cell tumouraccount for 7% of renal enucleation. Counceling patients affected by aforementioned "benign" tumours is of high value for patient cure and information.

ID 962
TPS PD-L1 EXPRESSION IN PAPILLARY RCCS VERSUS OTHER HISTOTYPES OF RCCS WITH PAPILLARY FEATURES

E. Marastoni[1], F. Zampieri[1], B. Dal Farra[2], S. Gobbo[3], P. Mattiolo[1], A. Caliò[1], G. Martignoni[1,4] and M. Brunelli[1,5]

[1] Department of Diagnostics and Public Health, University of Verona; [2] Department of Laboratory Medicine, University of Udine; [3] Department of Morphology, Surgery and Experimental Medicine, University of Ferrara; [4] Pathology Unit, PederzoliHospital, Peschiera del Garda, Verona; [5] FISH Lab, Hospital Trust, Azienda OspedalieraUniversitaria Integrata (AOUI) di Verona

Objectives: Discordant data are reported on TPS (tumor proportion score) PD-L1 expression in papillary RCCs sec. WHO 2016. Major discordances are due to erroneous inclusion of other histotypes with papillary features. We aimed to report PD-L1 expression in papillary RCCs versus other histotypes with papillary features. Clinical implications may impact to patient prognostication and predictiveness.

Materials and Methods: A series of consecutive papillary RCCs and other histotypes of RCCs with papillary features have been tested by both sp263 and E1L3N clones. Percentages of expression have been scored.
Results: Only 7% (3/42) of papillary RCCs did show PD-L1 expression. Both clones are positive in the same tumours. Only one out of three positive tumours showed > 50% of positive neoplastic cells. 17/20 (85%) collecting duct carcinoma with areas showing papillary features did show positive PD-L1 expression, such as most fumarate hydratase-deficient RCCs (3/5 positive tumours). 12 clear cell RCCs with areas picturing pseudo/papillae did not show any positive stains. Xp traslocation RCCs with diffuse pseudopapillae and papillary features showed and high percentages of positive cases (5/9, 56%).
Conclusion:
1) papillary RCCs does show few cases with PD-L1 expression; 2) clear cell RCCs with papillary or pseudopapillae do not usually show PD-L1 expression; 3) other histotypes with papillary features such as collecting ducts with papillae, fumarate hydratase-deficient RCCs and Xp11 traslocations RCCs with diffuse papillae show high percentages of PD-L1 positive cases; 4) PD-L1 appears to be expressed in histotyopes of RCCs with papillary features harboring worse prognostication per se versus papillary RCCs which does rarely show expression.

Venerdì 14 Ottobre 2022

Sala B 08.00 - 09.10

DERMATOPATOLOGIA

Moderatore: G. Ferrara

ID 803
PANCREATIC PANNICULITIS IN AN ASYMPTOMATIC CHRONIC PANCREATITIS

I.A. Neculaescu[1], R. Salmaso[1], A.P. Dei Tos[1], M. Alaibac[2]

[1] Department of Pathology, Padua University Hospital, Padua, Italy; [2] Department of Dermatology, Padua University Hospital, Padua, Italy

Objectives: Pancreatic panniculitis (PP) is a rare condition that occurs in 1 to 3% of patients with a pancreatic disorder. PP consists of subcutaneous fat necrosis due to the increase of lipase andamylase in the peripheral circulation. Males are affected more than females and the age is between 40 and 60 years. Clinically, the patients present painful erythematous nodules, usually on the extremities, that can ulcerate and exudate. Histologically, it shows fat tissue necrosis with anucleate adipocytes with cytoplasmic amorphous debris, called “ghost cells”, pathognomonic of this condition, associated to calcium deposits. The surrounding fat tissue is infiltrated by acute and chronic inflammatory cells. PP can be associated to pancreatic tumours or to acute and chronic pancreatitis, and most of the time it is diagnosed before the underlying disease. The differential diagnosis includes deep fungal infection, erythema nodosum, indurated erythema and alpha-1 antitrypsin deficiency.
Materials and methods: A 61-year-old woman presented with painful erythematous nodular lesions of the lower extremities. In her pathological history, there were noted kidney transplant in 1991, hypertension, anemia and celiac disease. She was receiving an immunosuppressive therapy and dialysis. There were made a skin biopsy of the lesion, a serum analysis and, subsequently, an abdominal CT and PET/RM scan.
Results: In the hypodermis, the skin biopsy revealed anucleate adipocytes with cytoplasm full ofamorphous debris and arc-shaped calcium deposits. This pattern directed towards the diagnosis of pancreatic panniculitis, although she didn’t have symptoms linked to a pancreatic disorder. Serum analysis revealed a lipase level of 1,384 U/L (normal 0-60 U/L) and amylase of 483 U/L (normal 13-53 U/L. The CT and the PET/RM scan was compatible with a chronic pancreatitis that was treated with prednisone.
Conclusions: Pancreatic panniculitis should be considered in the differential diagnosis of painful nodular lesions of skin, especially on the extremities. Serum amylase and lipase levels are often increased and imaging analysis can help to confirm a silent pancreatic disease.

ID 838
MOLECULAR CHARACTERIZATION OF SPITZOID MELANOMAS

R. Cappellesso[1], M. Vinci[2], R. Salmaso[1], M. Michelotto[1], P. Del Fiore[3], S. Mocellin[3,4], A.P. Dei Tos[2]

[1] Pathological Anatomy Unit, University Hospital of Padua, Padua, Italy;[2] Surgical Pathology and Cytopathology Unit, Department of Medicine (DIMED), University of Padua, Padua, Italy; [3] Soft-Tissue, Peritoneum and Melanoma Surgical Oncology Unit, Veneto Institute of Oncology IOV- IRCCS, Padua, Italy; [4] Department of Surgery, Oncology and Gastroenterology (DiSCOG), University of Padua, Padua, Italy

Objectives: Spitzoid melanomas are malignant melanocytic tumors sharing histologic features of Spitz nevi. Recently, the molecular background of Spitz neoplasms has been elucidated and it lacks the mutations of *BRAF* and *NRAS* usually found in conventional melanomas. The aim of this study was to assess whether the spitzoid melanomas with metas-

tases diagnosed at the Padova University Hospital belonged to the Spitz lineage.
Materials and methods: Overall, 108 spitzoid melanomas diagnosed between 1999 and 2021 with complete clinical and follow-up data were found. Mutational analyses of *BRAF* and *NRAS* were performed in metastasizing ones.
Results: 9/108 spitzoid melanomas had metastasis in the sentinel lymph node and 5 cases had distant metastasis (only one with positive sentinel lymph node). In the first group the mean age was 37.3 years (range 21-61 years), the lower extremities were affected in 6 cases, the mean Breslow thickness was 2.6 mm (range 0.7-4.4 mm), ulceration was present in only a case, there was only one death not related to the disease, and one tumor harbored a *BRAF* V600E mutation and another one a *NRAS* Q61K mutation. In the second group the mean age was 46.3 years (range 25-68 years), the extremities were the most affected sites, the mean Breslow thickness was 1.1 mm (range 0.5-2.1 mm), ulceration was always absent, there were three deaths of disease, and one tumor harbored a *BRAF* V600K mutation, one a *BRAF* V600E mutation and another one a *NRAS* Q61K mutation.
Conclusion: As previously reported, histology is an inconsistent predictor of Spitz lineage and tumor aggressiveness among spitzoid melanomas. Further analyses should assess the presence of Spitz specific molecular alterations (*I.e. HRAS* or *MAP2K1* mutation or *ALK, ROS1, NTRK, RET, MET, BRAF,* or *MAP3K8*) in the other metastasizing spitzoid melanomas.

ID 868
LINE-FIELD OPTICAL COHERENCE TOMOGRAPHY AND HISTOPATHOLOGICAL CORRELATION IN INFLAMMATORY DISORDERS

J. Farina[1], G. Broggi[1], R. Garro[1], C. C. Lin[1], M. Failla[1], R. Caltabiano[1]

[1] Department G. F. Ingrassia, Anatomic Pathology, University of Catania, Catania, Italy

Objectives: The aim of the study was to correlate line-field optical coherence tomography (LF-OCT)[1] imagingto histopathological sections features in some inflammatory disorders, such as psoriasis, eczema and lichen planus[2], to further demonstrate LF-OCT diagnostic accuracy.
Materials and methods: Our study included patients with histopathological confirmed diagnosis of psoriasis, atopic eczemaand lichen planus, previously studied with LC-OCT.
LC-OCT was performed with the commercially available LC-OCT device.
Results: A total of 15 adult patients with histopathological diagnosis of plaque psoriasis (N: 5), atopic eczema (N: 5), and lichen planus (N: 5) were included in our study. All 15 cases demonstrated how LC-OCT allowed the in-vivo recognition of the main microscopic features of the examined inflammatory skin diseases, with a strong correlation and overlapping with their respectivehistopathological features.
Conclusions: LC-OCT may represent a promising tool in inflammatory skin disorders in-vivo evaluation, with potential applications including more accurate diagnosis, biopsy guidance and follow-up, since it combines histology-like vertical mode to a confocal-like horizontal mode.

References

1 Dubois, A.; Levecq, O.; Azimani, H.; Siret, D.; Barut, A.; Suppa, M.; del Marmol, V.; Malvehy, J.; Cinotti, E.; Rubegni, P.; Perrot, J.-L. Line-Field Confocal Optical Coherence Tomography for High-Resolution Noninvasive Imaging of Skin Tumors. *J. Biomed. Opt.* **2018**, *23*(10), 1. https://doi.org/10.1117/1.JBO.23.10.106007.

2 Verzì, A. E.; Broggi, G.; Micali, G.; Sorci, F.; Caltabiano, R.; Lacarrubba, F. Line-Field Confocal Optical Coherence Tomography of Psoriasis, Eczema and Lichen Planus: A Case Series with Histopathological Correlation. *J. Eur. Acad. Dermatol. Venereol.* **2022**. https://doi.org/10.1111/JDV.18293.

ID 892
ARTIFICIAL INTELLIGENCE (AI) APPLICATED TO DIAGNOSIS OF MALIGNANT MELANOMA: A SINGLE INSTITUTIONAL EXPERIENCE

G. Cazzato[1], A. Massaro[2], A. Colagrande[1], E. Maiorano1, L. Resta[1], G. Ingravallo[1], A. Vacca[2]

[1] Section of Pathology, Department of Emergency and Organ Transplantation (DETO), University of Bari "Aldo Moro", Bari, 70124, Italy; [2] Centro Interdisciplinare Ricerca Telemedicina - CITEL, Università degli Studi di Bari "Aldo Moro"; 70124 Bari, Italy

Introduction: In the last two decades, an unprecedented development of information technologies associated with a considerable increase in memory units has allowed giant strides to be made in the futuristic field of artificial intelligence (AI). Although this is the prerogative of the informatics and technological branches, the use of software and new technologies has spread to many different fields of medicine, indeed to practically all branches, including pathological anatomy and, therefore, also the subbranch of dermatopathology [1,2]. For skin diseases that are more easily diagnosed like basal cell carcinoma (BCC), seborrheic keratosis (SK) and dermal nevus, excellent concordance results have been obtained for the first and embryonic convolutional neural networks (CNN). Instead, as was predictable, the difficulty in diagnosing ambiguous lesions, such as Spitz nevi and rare variants of malignant melanoma, together with the lack of interobserver agreement among dermatopathologists, has led to an objective difficulty in training artificial intelligence algorithms as well as those based on machine learning (ML) to a totally

reliable, reportable and repeatable level [3,4]. In this work we tried to train an AI algorithm using basic histopathological criteria that indicate and differentiate with a good probability a malignant melanoma from a severe dysplastic nevus (atypical nevus), and we provide information on the results obtained starting from routine histopathological images. (digitalpathology).

Material and methods: The artificial intelligence image processing algorithm used to classify and to enhance anomalies contained in the microscope image is the Fast Random Forest (FRF). The learning process of the algorithm is based on a preliminary classification of cluster of pixels of the same image [*] including possible Melanoma's areas: the preliminary identification of Melanoma morphological features, represents the labelling approach typical of machine learning supervised algorithms. The FRF testing provides as output the processed image with colored enhanced Melanoma pixel clusters (each class selected in the learning step is represented by a color), probabilistic maps (high probability highlighted by white to identify an anomaly in a specified image region), and algorithm performance indicators (precision, recall, and Receiver Operating Characteristic -ROC- curves [5]). The optimized hyperparameters and filter properties applied for the image FRF processing (features training) are [5]: Gaussian blur filter, Hessian matrix filter, membrane projections, membrane thickness equals to 1, membrane patch size equals to 19, minimum sigma equals to 1, maximum sigma equals to 16.

The algorithm was trained from these acquisitions.

Results: For five pixel clusters of the same dimensions (closest to the particular anomay), occurs a number of about 300 instances (computational cycles) to achieve the maximum precision (equals to 1), with a computational cost of about 2 minutes using a processor Intel(R) Core(TM) i5-7200U CPU, 2.71 GHz. The minimum recall performance parameter (near to 0) is achieved about 392 instances. The ROC curve (representing in the plane the true positive rate versus the false positive rate) is matching with the ideal curve of a perfect classifier (Figure 3). The performance indicators confirm the correct setting of the FRF hyperparameters.

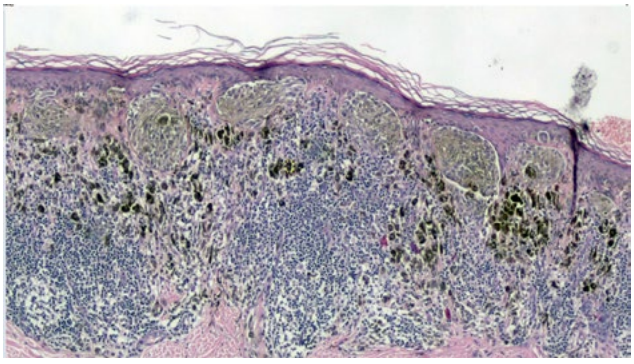


Figure 1. Example of human malignant melanoma in the horizontal and, secondarily, vertical growth phase with a brisk regression (inflammatory lymphocitic infiltrate, melanophages and neoangiogenesis).

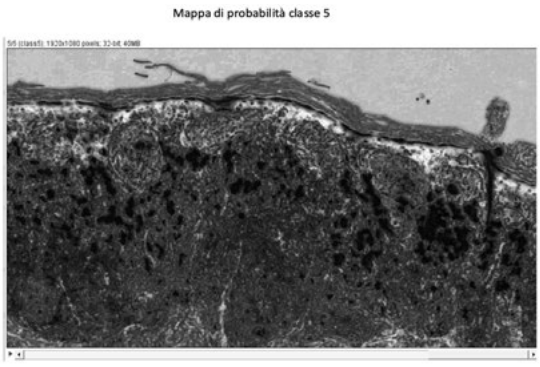


Figure 2. Probability image where the white color indicates a higher probability of recognition of the specific class.

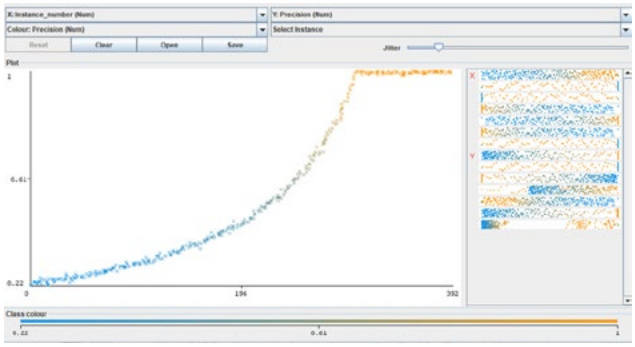


Figure 3. Performance of the classification.

The adopted image vision diagnostic protocol, is structured in the following steps:

- image acquisition by selecting the best zooming of the microscope;
- preliminary selection of image having a good resolution;
- preliminary identification of macro-areas of defect in each pre-selected image;
- identification of a class of a defect in the selected macro-area;
- training of the supervised machine learning FRF algorithm, by selecting the micro-defect in the macro-area;
- executing of the FRF algorithm until image vision performance indicator is good;
- analysis of the output images enhancing lesion defects.

Conclusions: The FRF images have been processed by following a specific image diagnostic protocol, oriented on reading and algorithm error minimization. An important tool for melanoma diagnosis is the

probability image estimated by the processed FRF output image. The probability image is useful to better discriminate information about ambiguous lesions. A single probability image is referred to a particular class of "defect", and enhances, by the white color, the defect distribution in the whole analyzed image. By knowing the dimension of the acquired microscope image, it is also possible to estimate the defect distribution percentage. All the adopted approaches are suitable to create a specific image vision platform for telemedicine digital pathology.

ID 896
METAPLASTIC OSSIFICATION OF THE SKIN: ACRAL ANGIO-OSTEOMA CUTIS

F. Arienzo[1], G. Soda[1]

[1] Department of Radiological, Oncological and Pathological Sciences,"Sapienza" University of Rome, Rome, Italy

Objectives: Cutaneous ossification may occur in association with a variety of cutaneous neoplasm or inflammatory conditions. Acral angio-osteoma cutis (AAOC) is a benign tumor characterized by vascular proliferation associated with newly formed bone deposition typically occurring on the acral skin (1). Differential diagnoses include Albright's hereditary osteodystrophy (AHO), subungual exostosis, pyogenic granuloma with metaplastic ossification, and/or osteoma cutis. We report here a case of AAOC involving the great toe of a 40 years-old man clinically diagnosed as pyogenic granuloma.
Materials and Methods: The patient presented with a single dome-shaped ulcerated lesion measuring 0.6×0.5x0.4 cm. No history of trauma was referred by the patient who was otherwise healthy. The lesion was totally excised. For its bony consistence, it was routinely processed for paraffin embedding after acid decalcification. Sections were stained with hematoxylin and eosin. Immunohistochemistry using antibodies for CD34, CD31, HHV8 and Ki67 was also performed.
Results: Histologically, the lesion was ulcerated and consisted of blood vessels without a lobular pattern lined by endothelial cells devoid of atypia and associated with interconnected tiny woven bone trabeculae. Osteoblasts were focally recognizable and devoid of cytologic atypia and mitotic activity. CD31 and CD34 immunostains highlighted the vascular component of the lesion. The overall Ki67 labeling index was lower than 5%. HHV8 immunostain was negative. The diagnosis of AAOC was rendered.
Conclusions: AAOC is rare. To date, few cases have been reported. Its pathogenesis is unknown. A history of trauma has been reported in few of the previously reported cases. Vascular endothelial growth factor (VEGF) and bone morphogenetic proteins (BMPs) have been thought to play a role in its development (2). Further studies are needed.

References

1. Googe PB, Page RN, King R, Griffin WC. Acral angioosteoma cutis. Am J Dermatopathol 2006; 28:228.
2. Won CY, Cho BK, Park HJ. Acral Angioosteoma Cutis on the Great Toe Mimicking Pyogenic Granuloma. Ann Dermatol 2015; 27:480-481. https://doi.org/10.5021/ad.2015.27.4.480. PMID: 26273178.

ID 913
TO INCLUDE OR NOT INCLUDE BULLA: THAT IS THE QUESTION. THE DILEMMA OF BRESLOW THICKNESS IN BULLOUS MELANOMA

L. Pepi, A. Filosa, M. Santarelli, F. Paparesta, C. Rubini, G. Goteri

Unit of Pathologic Anatomy and Histopathology, Polytechnic University of Marche Region, United Hospitals of Ancona, Italy

Objectives: Malignant melanoma presenting as a bullous lesion have rarely been reported. The rarity of the diagnosis is even greater if the patient does not have an underlying cutaneous bullous disease. The presence of a basilar, suprabasilar, or subepidermal bullous lesion characterizes the disease. There is a debate about the best name to describe melanomas histologically presenting with bullous lesions. Woltsche et al. proposed that tumor thickness should be measured excluding the bulla, as it represents a secondary phenomenon not directly related to the tumor burden.
Materials and methods: We report the case of a 45-year-old man with a pigmented lesion of the upper back that histologically resulted as superficial spreading melanoma, initially occupying the papillary dermis, with an intralesional blister.
Results: Melanoma thickness was measured by detracting the blister width according to Woltsche et al. The Breslow score was 0.3 mm with final staging pT1a. It is noteworthy that including bulla thickness, the Breslow score would have been greater than 0.8 mm with a pT1b final staging.
Conclusions: There are no guidelines as to how to measure tumor thickness in melanoma with bullous features. In our case, including or excluding the bulla resulted in a change in TNM classification thus showing that this is a real problem with influence on further management of the patients. The term bullous melanoma is suggested because acantholysis is a term defined as loss of cohesion between keratinocytes and cannot be applied to melanocytes. The diagnosis of bullous melanoma is based on criteria similar to those adopted for conventional variants of the disease, and the presence of bullous features do not pose particular diagnostic problems. The first question is the etiopathogenetic mechanism underlying blister formation. Many factors may contribute to the loss of cohesion, the most common of which

is friction. The location of the splitting and blister formation often reflects the involved mechanisms. In bullous melanoma, however, the crucial diagnostic aim is the definition of how to measure the Breslow index.

ID 919
PAGETOID RETICULOSIS OR UNILESIONAL MYCOSIS FUNGOIDES: THE DEBATE IS STILL OPEN

A. Filosa[1], M. Zanelli[2], A. Palicelli[2], S. Ascani[3], G. Goteri[1]

[1] S.O.D Anatomia Patologica Ospedali Riuniti Ancona; [2] S.O.D Anatomia Patologica Azienda USL-IRCCS Reggio Emilia; [3] S.O.D Anatomia Patologica Azienda Ospedaliero-Universitaria Ospedale Santa Maria Terni

Objective. Pagetoid Reticulosis (PR) present with a solitary, psoriasiform, erythematous patch at extremities, histologically showing epidermotropism with hyperplastic epidermis.
Material and methods: We recently observed a patient with a long standing solitary scaly lesion of the wrist. Physical examination did not show any other remarkable lesion.
Result: At histology we observed a dense intraepidermal lymphoid infiltrate with evident acanthosis and hyperkeratosis with papillomatosis. Intraepidermal lymphocytes were CD4+, with medium sized pleomorphic nuclei.
Conclusion: Besides localized PR, a solitary variant of mycosis fungoides (MF) has been described leading to a matter of classification. The 2018 WHO-EORTC classification recognizes only PR as a MF variant, with folliculotropic MF and Granulomatous slack skin. Examples with CD8+ and CD4–/CD8– phenotypes have been described.
A number of rarer CTCL commonly present with an isolated lesion, including CD4+ small/medium pleomorphic T-cell lymphoma and indolent CD8+ lymphoid proliferation of acral sites. Although these isolated lesion can run an indolent clinical course, theycontinue to pose diagnostic difficulty.

References

Kempf, Werner; Kazakov, Dmitry V; Kerl, Katrin. Cutaneous lymphomas: an update. Part 1: T- cell and natural killer/t-cell lymphomas and related conditions. American Journal of Dermatopathology, 2014; 36(2):105-123

Ally MS, Obson A: A review of the solitary cutaneous T-cell lymphomas. J Cutan Pathol2014;41:703–714.

VENERDÌ 14 OTTOBRE 2022

Sala C 08.00 - 09.00

NEUROPATOLOGIA

Moderatore: M. Gessi

ID 770
RESPONSE TO REGORAFENIB OF RECURRENT GLIOBLASTOMA. A CLINICAL AND NGS STUDY

M. Martini[1], Q. Giorgio D'Alessandris[2], C. Pizzimenti[1], V. Fiorentino[3], L. Lauretti[2], M. Balducci[4], R. Pallini[2], P. Tralongo[3], A. Ieni[1], G. Fadda[1]

[1] Dipartimento di Patologia umana dell'adulto e dell'età evolutiva "Gaetano Barresi", Università di Messina, Italia; [2] Dipartimento di Neuroscienze, Università Cattolica del Sacro Cuore, Roma, Italia; [3] Dipartimento di Scienze della vita e Sanità pubblica, Università Cattolica del Sacro Cuore, Roma, Italia; [4] Dipartimento di Scienze radiologiche ed ematologiche, Università Cattolica del Sacro Cuore, Roma, Italia

Objectives: Predictive factors for response to regorafenib in recurrent glioblastoma, IDH-wildtype, are scarcely recognized. The objective of this study was to identify molecular predictive factors for response to regorafenib using a clinically available platform.
Materials and methods: We analyzed a prospective cohort of 30 patients harboring recurrent glioblastoma, IDH-wildtype, and treated with regorafenib. Next-generation sequencing (NGS) analysis was performed on DNA extracted from paraffin-embedded tissues using a clinically available platform. Moreover, MGMT methylation and
EGFRvIII expression analyses were performed.
Results: Six-month progression-free survival (PFS) was 30% and median overall survival (OS) was 7.5 months. NGS analysis revealed a mutation of EGFR pathway in 18% of cases and a mutation in the mitogen-activated protein-kinase (MAPK) pathway in 18% of cases. In the remaining cases, no mutations were detected. MAPK pathway mutated patients had a poor response to regorafenib treatment, with a significantly shorter PFS and a nonsignificantly shorter OS compared to EGFR-mutated patients (for PFS, $p = 0.0061$; for OS, $p = 0.1076$). Multivariate analysis confirmed that MAPK pathway mutations independently predicted a shorter PFS after regorafenib treatment ($p = 0.0188$). The negative prognostic role of MAPK alterations was reinforced when we combined EGFR-mutated with EGFRvIII-positive cases.
Conclusions: Recurrent glioblastoma tumors with an alteration in MAPK pathway could belong to the mesenchymal subtype and respond poorly to regorafenib treatment, while EGFR-altered cases have a better response to regorafenib. We thus provide a molecular selection criteria easy to implement in the clinical practice.

ID 789
IMMUNOHISTOCHEMICAL EVIDENCE OF P62 IN HUMAN PRIMARY AND RECURRENT IDH1/2 WILD-TYPE GLIOBLASTOMAS

C. Pizzimenti[1], A. Ieni[2], G. Broggi[3], M.A. Franchina[2], M. Martini[2], R. Caltabiano[3], V. Fiorentino[4], G. Tuccari[2]

[1] Department of Biomedical, Dental, Morphological and Functional Imaging Sciences, University of Messina, Messina, Italy; [2] Department of Human Pathology in Adult and Developmental Age "Gaetano Barresi", Section of Pathology, University of Messina, Messina, Italy; [3] Department of Medical, Surgical Sciences and Advanced Technologies "G.F. Ingrassia", Anatomic Pathology, University of Catania, Catania, Italy; [4] Division of Anatomic Pathology and Histology, Fondazione Policlinico Universitario "Agostino Gemelli"-IRCCS, Rome, Italy

Objective: p62/SQSTM1/Sequestosome-1 is an autophagic protein that plays a crucial role in cellular metabolism, proliferation, and malignant growth. However, autophagy may influence development and therapy resistance in many types of human cancers[1,2]. In the present pilot study, we have analyzed p62 immunohistochemical pattern in a cohort of IDH1/2 wild-type glioblastomas (GBM), either primary and recurrent, in order to verify the concordance or discordance in these tumors, also in relation to patient's outcome and O^6-methylguanine–DNA methyltransferase (MGMT) status.

Materials and methods: 40 patients with both primary and recurrent IDH1/2 wild-type glioblastoma were included in our study andeach case was histologically reviewed. Clinical parameters such as age, gender, disease free interval and overall survival were collected for each patient. Immunohistochemical analysis for p62 and the assessment of MGMT promoter methylation status by pyrosequencing analysis were performed for each case.

Results: We have found p62 immunoexpression in the nucleus and cytoplasm of neoplastic elements in 45% primary and 55% recurrent GBM. A discordant p62 immunoreactivity was found in 35% of cases, with a variation either with positive or negative conversion. Statistically, p62 expression and MGMT status exhibited a significant value by univariate analysis, while only MGMT promoter methylation status emerged as independent prognostic value in multivariate test. Finally, the most favorable prognosis was documented when the same GBM case was positively concordant for both p62 expression and MGMT methylated status.

Conclusions: Since only few data[3,4] are available regarding the association between p62 expression and MGMT in GBM, we suggest that further investigations may be required to determine if new targeted therapies may be addressed against autophagy-related proteins such as p62.

References

1. Ieni A, Cardia R, Giuffrè G, et al: Immunohistochemical Expression of Autophagy-Related Proteins in Advanced Tubular Gastric Adenocarcinomas and Its Implications. Cancers (Basel). 2019;11(3):389. https://doi.org/10.3390/cancers11030389
2. Ieni A, Pizzimenti C, Giuffrè G, et al: Autophagy-related prognostic signature in HER2 positive gastric carcinomas. Curr Mol Med 22:809-818, 2021. https://doi.org/10.2174/1566524021666211123093532.
3. Jiang T and Wu Z: Immunohistochemical assessment of autophagic protein LC3B and p62 levels in glioma patients. Int J Clin Exp Pathol 11: 862-868, 2018.
4. Tamrakar S, Yashiro M, Kawashima T, et al: Clinicopathological Significance of Autophagy-related Proteins and its Association With Genetic Alterations in Gliomas. Anticancer Res 39: 1233-1242, 2019.https://doi.org/10.21873/anticanres.13233

ID 790
EWSR1-ALTERED ASTROBLASTOMA-LIKE TUMOR: REPORT OF A POSSIBLE NEW ENTITY

A. d'Amati[1,2], M. Antonelli[1], M. Gessi[3], M. Massimino[4], P. Modena[5], M. Giagnacovo[5], E. Maiorano[2], F. Giangaspero[1]

[1] Department of Radiological, Oncological and Anatomo-Pathological Sciences, La Sapienza University, Rome, Italy; [2] Anatomic Pathology Unit, Department of Emergency and Organ Transplantation, University of Bari, Bari, Italy; [3] Neuropathology Unit, Pathology Division, Fondazione IRCCS Policlinico Universitario A. Gemelli, Università Cattolica S. Cuore, Rome, Italy; [4] Pediatric Oncology Unit, Medical Oncology and Hematology Department, Fondazione IRCCS Istituto Nazionale Dei Tumori, Milan, Italy; [5] Genetics Unit, ASST Lariana General Hospital, Como, Italy

A 2 years old boy presented with a supratentorial mass in the left hemisphere. After surgical resection, the tumor showed histologic features suggestive of both ependymoma and astroblastoma. Five mitoses in 10 HPF, necrosis and microvascular proliferation were detected.Immunohistochemistry showed GFAP positivity, Olig2 negativity and dot-like EMA positivity. L1CAM and p65 were negative. Molecular analyses were performed, revealing that fusions involving ZFTA and YAP1 genes were absent, excluding supratentorial ependymoma (ZFTA or YAP1 fusion-positive). MN1 alterations were not detected, excluding also astroblastoma. Array CGH revealed a breakpoint on chromosome 22, in the site of the introne 7 of EWSR1 gene. Histologic features of ependymoma or astroblastoma are not specific and can be seen in other gliomas. Genomics studies revealed that tumors with astroblastoma-like morphology and MN1 gene fusion (usually with BEND2 as partner) define a specific group of circumscribed gliomas, called "astroblastoma MN1-altered", included in the 5th edition of the WHO Classification of the Central Nervous System Tumors. Recent studies reported of other gliomas with astroblastoma-like histology lacking MN1 alterations, instead harboring EWSR1-BEND2 fusion and resolving into a distinct epigenetic cluster. The biologic relationship of these gliomas lacking MN1 alterations compared to

the “astroblastoma MN1-altered” is currently unresolved. This case provides evidence that neoplasms with astroblastoma-like morphology and absence of MN1 alterations may represent a new molecularly-defined entity and suggests that other genes, rather than MN1, may have important roles in tumor pathogenesis.

ID 791
HISTOLOGIC DEFINITION OF FLAIR HYPERINTENSITY ZONE AND CANCER STEM CELL DISTRIBUTION IN GLIOBLASTOMA

M. Failla[1], G. Broggi[1], R. Altieri[2], F. Certo[2], G. Barbagallo[2], J. Farina[1], G.N. Rosano[1], B. Morante[1], R.Caltabiano[1]

[1] Anatomic Pathology, Department “G.F. Ingrassia”, University of Catania, Catania, Italy; [2] Neurological Surgery, Department “G.F. Ingrassia”, University of Catania, Catania, Italy.

Objectives: The fact that higher expressions of cancer stem cell (CSC) markers were reported at the infiltrating tumor edge of glioblastoma (GBM), along with the evidence that GBM recurrences often arose from peritumoral areas, increased the need to pay more attention to “what was going on around the tumor”. Moreover, as little is known about the histopathologic composition of the radiologically-defined FLAIR hyperintensity zone, we investigated the histopathologic features, along with the distribution of SOX2+ and CD133+ CSCs, both in the central core (Enhancing Nodule; EN) and in the FLAIR hyperintensity zone of a series of 33 GBMs, *IDH-wild type*.
Materials and methods: Our cohort included 20 males and 13 females (mean age: 56 years). The inclusion criterion was the intraoperative sampling of EN and FLAIR regions identified by Neuronavigation and by the use of 5-ALA. The immunoexpression of SOX2 and CD133 was semiquantitatively evaluated.
Results: Histologically, EN regions exhibited the conventional GBM morphology with hypercellularity, increased mitotic activity, necrosis and/or microvascular proliferation (MVP) in 29/33 cases; the ENs of 4/33 cases lacked necrosis and MVP and the diagnosis of GBM was mainly molecularly-based. Histological samples from FLAIR regions showed fragments of white matter tissue focally to diffusely infiltrated by GBM cells in 25/33 cases; neither necrosis nor MVP were seen within these samples. FLAIR regions from 5/33 cases exhibited a mixture of white matter with reactive astrogliosis, along with grey matter with neuronal satellitosis. The FLAIR zones from 3/33 cases showed viable tumor tissue with necrosis and MVP. No significant difference in the quantitative distribution of SOX2 and CD133 immunoexpressions between the EN and FLAIR regions was found.
Conclusions: Based on our results, an interesting future perspective of the present study would beto recalibrate both the surgical and radiotherapy target in GBM to improve the local control of the disease.

ID 810
GLIOBLASTOMA, *IDH*-WILDTYPE WITH *FGFR3-TACC3* FUSION: PATHOLOGIC AND MOLECULAR FEATURES OF 4 CASES

G. Broggi[1] , G. Magro[1], E. Tirrò[2], P. Vigneri[2], G.N. Rosano[1], B. Morante[1], G. Barbagallo[3], R. Caltabiano[1]

[1] Anatomic Pathology, Department “G.F. Ingrassia”, University of Catania, Catania, Italy; [2] Center of Experimental Oncology and Hematology, University of Catania, Catania, Italy; [3] Neurological Surgery, Department “G.F. Ingrassia”, University of Catania, Catania, Italy.

Objectives: The *FGFR3-TACC3* gene fusion is a rare, potentially targetable, molecular rearrangement which is encountered in about 3-4% of glioblastomas, *IDH-wild type* (GBMs,*IDHwt*). It has been reported that some unusual morphological features may be seen inGBMs, *IDH*wt with *FGFR3-TACC3* fusion, including bland-looking rounded cells with ovoid nuclei, nuclear palisading, endocrinoid network of “chicken-wire” vessels, microcalcifications and desmoplastic stroma.
Material and methods: We report the clinico-pathologic and molecular features of a series of 4 cases of GBMs, *IDH*wt, exhibiting some of the above-mentioned morphological features and molecularly-proven *FGFR3-TACC3* fusions. Our cohort included 3 males and 1 female (age range: 56-79 years).
Results: Histologically, 1 out of 4 cases exhibited, in addition to the conventional GBM morphology, the following unusual morphologic features: i) rounded monomorphous tumor cells with scant pale cytoplasm (perinuclear halo); ii) thin capillary-like vessels with “chicken-wire” pattern; iii) nuclear palisading; iv) vague perivascular pseudorosettes; v) a spindle cell componentset in a myxoid stroma. While 2 cases showed the conventional GBM morphology with necrosis and microvascular proliferation, the remaining case exhibited low-grade histology (moderate cellularity, few mitoses, low degree of nuclear atypia, absence of necrosis and microvascular proliferation); accordingly, the diagnosis of GBM, *IDHwt* was rendered on the basis of the characteristic molecular signature. Next-generation sequencing analyses were performed and *FGFR3-TACC3* fusion was found in all cases.
Conclusions: As it has been reported that *FGFR3-TACC3* fusion is a potentially targetable molecular alteration, whose presence confers a better prognosis to GBM, *IDHwt*, the present series emphasizes that molecular tests are crucial in the diagnostic approach to adult diffuse gliomas, *IDHwt*, especially in presence of unusual morphologic features.

ID 901
A RECENTLY DESCRIBED CENTRAL NERVOUS SYSTEM TUMOUR ENTITY IDENTIFIED BY DNA METHYLATION-BASED ANALYSIS. A CASE REPORT

S. Ammendola[1], M. Antonelli[2], E. Miele[3], V. Barresi[4], F. Giangaspero[2], P.L. Poliani[1]

[1] Department of Translational and Molecular Medicine, Pathology Unit, University of Brescia, Brescia, Italy; [2] Department of Radiologic, Oncologic and Anatomo Pathological Sciences, University Sapienza of Rome; [3] Department of Pediatric Onco-Hematology and Cell and Gene Therapy, Bambino Gesù Children's Hospital, IRCCS, Rome, Italy; [4] Department of Diagnostics and Public Health, Section of Anatomic Pathology, Verona University Hospital, Verona, Italy

Objectives: Methylation-based molecular analysis has recently emerged as a valuable diagnostic tool for central nervous system (CNS) neoplasms, providing a novel classification approach on a variety of CNS lesions for which the histo-morphological assessment may be insufficient to provide a definitive diagnosis. With the advent of the latest edition of the WHO classification of tumours of the central nervous system, the number of distinct molecularly defined entities is increasing. Here we report the case of a 17 year-old male patient with an intraventricular mass of the right lateral ventricle. The patient presented with a three-day history of headache and diplopia. He was referred to the Neuroradiology Department of our Institution where a magnetic resonance revealed a solitary intra-axial lesion, measuring 3,5x3,4x2,7 cm with lobulated profiles and non-homogeneous contrast enhancement, due to alternating enhancing solid components admixed with cystic non-enhancing areas. The lesion had a complete intraventricular development, involving the ventral aspects of the right ventricle, in close connection with the septum pellucidum. The mass caused a bilateral obstruction of the Monroe foramina with ventricular enlargement and hydrocepahlus. The final neuroradiological diagnosis was neurocytoma. The patient underwent neurosurgery with transcallosal approach and gross total excision of the lesion. The pathology report was followed by the results of methylation analysis. Aim of this report is to describe a complex and unusual lesion of the CNS with ambiguous features, in which methylation analysis contributed to the definition of tumour cell lineage and provided prognostic information.

Materials and methods: Immunohistochemistry was performed on FFPE slides with the following antibodies: GFAP, S100, Olig-2, synaptophysin, CD34, ATRX, p53, BRAF V600E, IDH R132H, Ki-67.

Fluorescent in situ hybridization for 1p/19q codeletion analysis was performed using fluorescent probes targeting 1p36 and 19q13 chromosomal loci.

MGMT promoter methylation analysis, as well as DNA sequencing to identify mutations in *IDH1*/2and BRAFV600E mutation were also carried out. Additionally, FFPE material was submitted to methylation analysis.

Results: Histologic examination revealed a primary CNS lesion composed of elements with glial differentiation, predominantly solid architecture and focal microcystic areas with secretory aspects. The lesion was moderately cellular, with pseudo-nodular areas of increased cellularity. Cytologically, neoplastic cells were small or medium-sized, showing moderately hyperchromatic nuclei, occasional nuclear pseudo-inclusions and scant eosinophilic or clear cytoplasms. Less frequently, elements with moderate pleomorphism and bizarre nuclei, along with scattered multinucleated giant cells were found. The neoplasm showed increased vascularity with vessels lined by plump endothelial cells in the absence of a recognisable microvascular proliferation. Mitotic activity was low (0-1 mitosis/10 HPF of 0.55 mm^2). Prominent intratumoral inflammatory infiltrate was also present, mainly composed by lymphocytes and plasma cells. Perilesional non-neoplastic cerebral parenchyma showed marked reactive gliosis with microglia activation. Tumour cells were immunoreactive for GFAP and S100 and focally for Olig-2 and synaptophysin. CD34 immunostain resulted positive in part of the neoplastic elements, with increased intensity in perivascular tumour cells. Nuclear expression of ATRX was preserved and p53 stained with moderate intensity in a subset of neoplastic nuclei. BRAF V600E and IDH R132H immuno-stains were negative. Ki-67-labeling index was 2-3%, with rare areas in which it reached 4-5%. Molecular analysis did not show IDH 1/2 gene mutations and MGMT gene promoter showed absence of methylation. Fluorescent in situ hybridization did not reveal the presence of 1p/19q codeletion. The overall features of the lesion suggested a low-grade primitive neoplasm with glial and/or neuroepithelial differentiation. Differential diagnoses included subependymoma, chordoid glioma and, due to the focal but strong CD34 immunohistochemical expression in part of neoplastic elements, polymorphous low grade neuroepithelial tumor of the young (PLNTY), despite the unusual intraventricular localization. Methylation profile of the lesion was coherent with a distinct methylation class of neuroepithelial tumours, recently described by Alhalabi et al., characterized by *PATZ1* fusion. This entity has a broad histological spectrum and is molecularly defined by the presence of *PATZ1* fusions with *EWSR1* or alternatively *MN1,* and frequent copy number alterations of chromosome 22, where fusion partners are located. It localizes most commonly in cerebral hemispheres but intraventricular lesions were also rarely described. The lesion does not show sex predilection and the mean age is 11 years. These neoplasms display high expression of markers of neural development and provisional data on patients' survival show a less aggressive behaviour compared to high grade gliomas, in accor-

dance with the low proliferative and mitotic activity of the tumour. Recurrences have though been reported in a subset of patients.
Conclusions: We report the case of a recently described tumour entity, showing neuroepithelial phenotype, a distinct methylation profile and a recurrent genetic alteration in *PATZ1*. Integrated histological, immunohistochemical and molecular diagnosis has dramatically changed the perspectives of neuro-oncology and these new diagnostic tools may improve our current knowledge of tumour biology, with relevant clinical impact in terms of prognostic information and therapeutical strategies for patients with CNS tumors.

Venerdì 14 Ottobre 2022

Sala D 08.40 - 09.00

PATOLOGIA PEDIATRICA

Moderatore: R. Alaggio

ID 932
AN EXPRESSION PROFILE STUDY BY RNA-SEQUENCING IN A SERIES OFINFANTILE FIBROSARCOMAS WITH *ETV6::NTRK3* AND ALTERNATIVE FUSIONS

S. Vallese[1], R. Alaggio[1], G. Milano[2], E. Miele[2], I. Giovannoni[1], A. Stracuzzi[1], S. Barresi[1]

[1] Pathology Unit, Ospedale Pediatrico Bambino Gesù, Rome; [2] Oncology Unit, Ospedale Pediatrico Bambino Gesù, Rome

Objectives: Infantile Fibrosarcoma (IF), is a rare tumor rising within the first 2 years of life with a favorable outcome in more than 90% of cases. Most IF harbor the *ETV6::NTRK3* fusion but in a minority of IF cases, different gene fusions have been identified. The aim of this study is to compare gene expression profiling of IF with *ETV6::NTRK3* fusion and IF with different gene fusions to identify common pathways supporting their classification as one entity and to relate molecular findings with morphological and clinical data.
Materials And methods: 50 of cases diagnosed as IF were collected from the Pathology Department of Ospedale PediatricoBambino Gesù (OPBG). All the cases went through molecular analisys (RT-PCR, NGS Rna panel and/or NGS Rna-sequencing analysis). Differential gene expression profile results were reported in MDS plots and heatmaps, and gene set enrichment analysis were performed.
Results: 36 IF cases were *ETV6::NTRK3* fusion positive. 7 IF had alternative fusion transcripts (*GOLGA4::RAF1*, *CLIP1::RAF*, *LRRFIP2::RAF1*, *MYH10::RET, TPM3::NTRK1 (2) and FGFR1-CK-AP5),* 3 cases had no fusions. In 5 cases, NGS analysis and histologic revaluation led to change in the final diagnosis.
The comparison of genetic expression profile of 10 IF *ETV6::NTRK3* positive and IF with alterative transcripts showed a different segregation on the MDS plot and heatmap, and gene set enrichment analysis showed the upregulation of pathways involved in angiogenesis in *ETV6-NTRK3* positive IF (such as angiogenetic signaling, PDGF, and VEGF pathways).
Conclusions: Rna-sequencing analysis demonstrated a previously undetected fusion in a subset of IF, and gene expression profile highlighted the different pathways between classic IF with *ETV6::NTRK3* fusion and those with other fusions, with involved angiogenetic pathways activation in *ETV6::NTRK3* positive IF, opening new perspectives in classification of these rare entities.

ID 933
ASSESSMENT OF TUMOR MUTATIONAL BURDEN AND MICROSATELLITE INSTABILITY IN PEDIATRIC SOLID TUMORS: A SINGLE CENTER EXPERIENCE

I. Giovannoni[1], A. Stracuzzi[1], C. Tancredi[1], S. Rossi[1], R. Alaggio[1], S.Barresi[1].

[1] Pathology Unit, Bambino Gesù Children's Hospital, IRCCS, Rome, Italy

Background: Over the past decade, the use of Next-Generation-Sequencing (NGS) has allowed significant improvements in the treatment of cancer in children with the identification of novel therapeutic targets.
Rapidly developing on NGS technology makes it possible now to accurately measure and integrate information on key current immuno-oncology biomarkers: Tumor- Mutational-Burden (TMB) and Microsatellite-Instability (MSI) into the mutational analysis offering a comprehensive genomic profiling and a personalizing cancer treatment.
In pediatric oncology, few studies have been published characterizing TMB and MSI biomarkers so far.
Method: In our study, we performed NGS using a commercial panel with 500-genes associated to adult solid tumors and calculated TMB and MSI in 113 tumors from children, adolescents, and young adults, comprising: 71 brain (21 Glioma, ependymoma, 16 Medulloblastoma and Embryonal Tumor, 14 Astrocytoma, 10 Ependymoma, 10 other) and 43 solid soft tumors (11 rhabdomyosarcoma, 21 sarcoma, 11 other).
Results: Our data suggest that 33% of the children in this cohort carry a driver mutation (*BRAF, H3F3A*, *IDH1*, *IDH2*, *PIK3CA*, *TP53, MMR-genes*, *CTNNB1, MYOD1, SMARCA4*).

Of 71 brain tumors, TMB was low (0-6) in 93%, intermediate (6-20) in 6%, and high (> 20) in 1%. The brain tumors with high TMB (31), was a 10 years-old boy with medulloblastoma relapse *AKT1* mutated (p.E17K).
Of 43 soft tumors, TMB was low in 95%, intermediate in 5%, and high in 0%.
MSI score in our cohort was low (> 20%).
Conclusion: Herein, we report a single center experience with commercial NGS panel with 500-genes in 113 pediatric patients with distinct molecular types of cancer (71 brain and 43 soft tumors). In our study, as in other pediatric data, there are several limitations likely the discrimination between primary tumors, recurrent tumors and previously treated tumors.
Pediatric cancers are molecularly different from adult cancers; hence a specific diagnostic World Health Organization (WHO) Classification of childhood tumors was needed. Our data confirmed a low rate of hypermutated samples, low TMB and stable MSI. Therefore, histological and molecular studies is needed to improve knowledge of biological behavior and target therapies.

Venerdì 14 Ottobre 2022

SALA E 8.20 - 09.00

PATOLOGIA PLEUROPOLMONARE 2

Moderatore: L. Righi

ID 813
MEET THE NASTY NEW ENTITY OF THORACIC PATHOLOGY: SMARCA4-DEFICIENT UNDIFFERENTIATED TUMOR

S. Pancetti[1,2], V.Belsito[1,2], S.Zanella[1,2], P.Bossi[2], A.Bonometti[1,2]

[1] Humanitas University, Rozzano, Italy; [2] Pathology Unit, Humanitas Research Hospital, Rozzano, Italy.

Objectives: describe a rare case of thoracic SMARCA4-deficient undifferentiated tumor (SMARCA4-UT) and explore its biological and histopathological features.
Materials and methods: A 40 years old, with a history of heavy smoking, came to our attention to investigate polyarthralgia and nail clubbing. Pneumological examination showed a severe reduction in lung function. Whole-body CT and PET scans showed the presence of an upper lobe lesion in the left lung, suspicious of neoplasia. After an inconclusive biopsy, a lobectomy was performed.
Results: Histologically, the lesion was made of sheets of atypical epithelioid to rhabdoid cells with prominent nucleoli and eosinophilic cytoplasm with hyaline perinuclear inclusion, infiltrating the lung parenchyma and displaying broad areas of necrosis and a brisk mitotic activity.
Immunophenotypically the neoplastic cells result: CK+, EMA+, Synaptophysin+, SALL4+, TTF-1-, p40-,SOX10-, PLAP-, CD34-, BRG-1-. Thus we rendered a diagnosis of thoracic SMARCA4-UT.
Conclusions: SMARCA4-UT is a recently aggressive neoplasm, known in only around 100 cases and affecting mainly young male smokers. The hallmark of this entity is the loss of BRM, encoded by SMARCA2, or BRG1, encoded by SMARCA4. BRG1 is an ATPase that suppresses unscheduled R-loops which cause fork slow-down, DNA asymmetry, and stalling (both causes of double-strand brakes). BRG1 loss induces genome-wide accumulation of R-loops and thus cytogenetic imbalances.
SMARCA4-UT appears as an undifferentiated high-grade neoplasm and enters in differential diagnosis with atypical teratoid/rhabdoid tumor, lung carcinoma, NUT carcinoma, germ-cell tumor, melanoma, and mesothelioma. The neoplastic cells usually express SOX2, CD34, and SALL4. The proliferation index is high with a mean of 79%.
Given its rarity, aggressiveness, and histopathological peculiarities, it may represent a major diagnostic challenge that needs to be solved thanks to an integration of clinical, radiological, and pathological findings.

ID 816
MESOTHELIOMA WITH SMALL CELL COMPONENT: A CLINIC-PATHOLOGIC SERIES OF 11 CASES

Francesca Ambrosi[1], Maria Cecilia Mengoli[2], Giorgia Leone[3], Costantino Ricci[4], Maria Sirolli[4], Chiara Baldovini[5], Alberto Cavazza[2], Giulio Rossi[5]

[1] Pathology Unit, Maggiore Hospital, AUSL Bologna, Bologna, Italy; [2] Pathology Unit, Azienda USL/IRCCS di Reggio Emilia, Reggio Emilia, Italy; [3] Pathology Service, Humanitas Istituto Clinico Catanese, Catania, Italy; [4] Department of Experimental, Diagnostic and Specialty Medicine (DIMES), University of Bologna, Bologna, Italy; [5] Pathology Unit, Fondazione Poliambulanza Hospital Institute, Brescia, Italy

Background. Mesothelioma with small cell features is a rare occurrence that may be confused with other small-cell malignancies of the thoracic region, mainly small cell carcinoma and some sarcomas (e.g., Ewing's sarcoma, desmoplastic small round cell tumor, synovial sarcoma). Small cell component in mesothelioma has been poorly investigated, although it seems related to a very poor prognosis. The purpose of this study is to better characterize the clinical and pathologic features of this challenging mesothelioma.
Materials and methods: Eleven cases of mesothelioma with small cell component were collected from 4 different Institutions. A careful morphologic examina-

tion was performed on all the available hematoxylin-eosin-stained slides. The small cell component consisted of solid nests of closely packed, monotonous, rounded cells with high nuclear/cytoplasmic ratio. The percentage of small cell component was quoted in all cases, including other histologic features. In all cases, a panel of immunohistochemical markers was performed using an automated immunostainer (ULTRA XT, Ventana/Roche) and the following primary antibodies: pan-cytokeratin (AE1/AE3; CK8/18; CK5/6); calretinin, D2-40, WT1, RB1, Claudin-4, BAP1, BerEp4, TTF1, chromogranin, synaptophysin, CD56. One case was analyzed by next-generation sequencing (Ion GeneStudio S5 - Thermo Fisher Scientific) after dissection of the small cell component. Clinical data was obtained from the referring physicians and/or the clinical charts.

Results: There were 8 men and 3 women, with a mean age of 71.6 years (range, 53-86 years). Six patients had an overt history of asbestos exposure (3 unknown and 2 non-exposed). The tumor had a pleural origin in 10 cases (6 on right; 2 on left) and from the peritoneum in 1. Six cases were classified as biphasic mesotheliomas and 5 as epithelioid. The small cell component ranged from 20% to 90% of the entire tumor (mean, 54.5%). All cases were in T3 stage. The diagnosis was performed on 9 thoracoscopic biopsies, 1 pleural surgical resection and 1 peritoneal biopsy. Ten cases were originally diagnosed as mesothelioma, while 1 was misdiagnosed as small cell carcinoma. At immunohistochemistry, small cell mesothelioma stained with pan-CK and RB1, while claudin-4 was always negative and BerEp4 positive in 2 cases. A negative/altered BAP1expression was found in 9 cases (82%). Among mesothelial markers, calretinin was expressed in all but 1, while the other tested markers were variably positive (WT1 in 7, D2-40 in 8, CK5/6 in 6). Chromogranin was negative in all cases, while synaptophysin was positive in 6 cases. NGS analysis revealed a NOTCH4 gene mutation in the tested case, but no differences were noted between small cell and conventional epithelioidcomponents. Eight patients received platinum-based chemotherapy. Nine patients died of disease from 6 days to 29 months after diagnosis. Two cases were alive with disease after 9 and 11 months.

Conclusions: Small cell mesothelioma is a rare variant of epithelioid and biphasic types (1 out of 126 cases, 0.8% in a consecutive series of one of the involved Institution), possibly involving the pleura and the peritoneum. BAP1 and claudin-4 negativity coupled to coordinated expression of RB1 and mesothelial markers are important clues in differential diagnosis, particularly in cases showing some expression with neuroendocrine markers (5 cases). The prognosis is poorer (10.4 months) that than expected conventional epithelioid mesothelioma.

ID 821
AN UNEXPECTED GUEST: A CHALLENGING DIAGNOSIS OF PULMONARY ECHINOCOCCAL CYST

Giuseppe Maggioni[1]; Alessandro Bonis[2]; Marco Schiavon[2]; Chiara Giraudo[3]; Francesca Lunardi[1]; Federica Pezzuto[1]; Fiorella Calabrese[1]

[1] Anatomic Pathology Unit, Department of Medicine, University of Padua, Padua, Italy; [2] Thoracic Surgery Unit, Department of Medicine, University of Padua, Padua, Italy; [3] Department of Medicine, University of Padua, Padua, Italy

Background: Echinococcosis is caused by tapeworm belonging to Echinococcus genus.
Clinically, common infection sites include liver, lungs and spleen. Pulmonary echinococcosis respiratory symptoms vary from mild (chest pain, dyspnea) to severe (cough and hemoptysis with membrane expectoration and anaphylactic reactions) depending on cyst location and structure compression. Imaging is consistent with well-defined homogeneous cysts with smooth, non-calcified walls of variable thickness and fluid content, often without features distinguishing them from malignancy.

Clinical case: A 61-yo Northern African male, with an otherwise unremarkable medical history, sought medical attention for left upper chest pain. No other symptoms or abnormal blood and respiratory function tests were reported. A CT and PET-CT scan showed a voluminous fluid-filled cyst with minor metabolic uptake and thick pleural adherences suspicious of sub-pleural cyst in first instance. The lack of a clear cleavage surface made surgical resection difficult. Intra-operative frozen section together with imprint cytology excluded a malignant lesion or a sub-pleural cyst, suggesting rather an acellular laminar structure coherent with echinococcal exocyst. Final pathological examination confirmed the diagnosis detecting a whitish fibrous capsule and endocystical inner germinative coating with only a single organism consistent with E. Granulosusprotoscolex. The surrounding pulmonary parenchyma showed foci of organizing pneumonia and mixed inflammatory cell infiltration with a few eosinophils.

Discussion and conclusion: Frozen section examination with imprint cytology and the detection of peculiar histopathological characteristics may facilitate a diagnosis that clinically could potentially bear challenges, especially in paucisymptocmatic patients. Despite the rare incidence in our country, all specialists (clinicians, radiologists and pathologists) should be aware of this uncommon entity, especially in populations from endemic areas or with relevant travel or occupational history.

ID 842
TWO CASES OF MULTINODULAR LUNG DISEASE IN FEMALES: DIAGNOSTIC PATHWAY AND DIFFERENTIAL DIAGNOSIS

Q. Zhang[1], M. Chiappetta[2], L. Petracca[2], A. Cancellieri[1]

[1] Section of Anatomic Pathology, Department of Life Sciences and Public Health, Università Cattolica del Sacro Cuore, Fondazione Policlinico Universitario A. Gemelli, IRCCS, Rome, Italy; [2] Thoracic SurgeryUnit,Università Cattolica del Sacro Cuore, Fondazione Policlinico Universitario A. Gemelli, IRCCS, Rome, Italy

Objectives: two cases of multinodular lung disease are presented together with their clinico-radiological features.
Case 1: a 62-year-old female, current smoker, was admitted to our hospital complaining chronic cough. Pulmonary function tests were unremarkable. The CT scan recognized multiple, sub-centimetric, bilateral, both centrilobular but also evenly (random) distributed, high density and often calcific nodules wich wereassociated with mosaic pattern suggesting air trapping. A larger, 1.5-cm, sharp-edged nodule was present in the right upper lobe. A trans-bronchial biopsy of the nodule was performed under general anesthesia and echo-fluoroscopy guidance, showing cytologic and histologic features of a carcinoid tumor. A surgical resection of the right upper lobe confirmed the diagnosis of NET G1, together with both tumorlets and linear neuroendocrine hyperplasia, consistent with DIPNECH. Constrictive bronchiolitis, responsible for air trapping, was rarely observed. In addition, several randomly distributed nodules were present, consistent with previous chicken-pox lung infection. A detailed anamnestic investigation confirmed the diagnosis.
Case 2: a 75-year-old female underwent two wedge resections from upper and lower right lobes, respectively, to make a diagnosis on her multinodular and bilateral lung disease and obtain material for eventually prognostic molecular studies. No malignancy was referred in the anamnesis or found at a total body CT scan. Histologically, nodules were 1-2 mm in size and mainly in a peri-lobular (perivenular) distribution. They consisted in a multifocal proliferation of spindle, bland-looking cells, with immunophenotypical features of meningothelial-like nodules. Further investigation revealed that the patient suffered from chronic hypoxia.
Conclusion: a multinodular lung pattern can be due to different diseases. Before histology, a detailed anamnestic and HRCT evaluation usually contribute to define the correct setting of the disease.

Venerdì 14 Ottobre 2022

Sala F 08.00-09.10

PATOLOGIA GINECOLOGICA 2

Moderatore: G. L. Taddei

ID 879
GASTRIC TYPE ENDOCERVICAL ADENOCARCINOMA: A RETROSPECTIVE STUDY ON INCIDENCE AND DIAGNOSIS IN A SINGLE INSTITUTION

E. Albertini[1], A.G. Corradini[1], F. Rosini[1], A. Costantino[1], S. Coluccelli[1], T. Maloberti[1], D. de Biase[1], G. Tallini[1], D. Santini[1], A. De Leo[1]

[1] Pathology Unit, University of Bologna Medical Center, Bologna, Italy

Objectives: Gastric type adenocarcinoma of the uterine cervix (GAS) is a rare variant of endocervical adenocarcinoma not etiologically associated with human papillomavirus (HPV) infection. These tumors are reported to have worse prognosis than usual HPV-associated endocervical adenocarcinoma (UEA). The objectives of our study were to determine the incidence and clinical-pathological characteristics of GAS in a single institution.
Materials and Methods: A retrospective review of GAS was performed from the pathology database from 2017 to 2022 including only the diagnosis of invasive adenocarcinomas. Immunohistochemistry(IHC) was performed including antibodies for p16, estrogen and progesterone receptors, PAX8, HER2. Clinical and pathological data was retrieved from pathology reports and charts.
Results: Using the International Endocervical Adenocarcinoma Criteria and Classification criteria, 40cases (85,1%) were classified as UEA, 5 cases (10,6%) as GAS, one case of neuroendocrine carcinoma (2,1%) and one case as clear cell carcinoma (2,1%). The mean age at time of GAS diagnosis was 48years (range, 34 to 58 years). At presentation, 100% were advanced stage (FIGO II–IV), 60% had lymph node metastases, 40% had ovarian involvement, and 40% had abdominal disease. The metastatic sites included lymph nodes, adnexa, omentum, bowel, peritoneum, abdominal wall, bladder, and vagina. Four tumours were high-grade and all showed a pattern of invasion type C according to Silva system. All the tumours were negative for HPV infection. By IHC, 4/5 cases were negative for p16 and PAX8. P16 was overexpressed in one case (but HPV negative). Abnormal p53 (overexpression or null) wasseen in 60% of cases. ERBB2 amplification was detected in one case.
Conclusions: GAS represents a distinct, biologically

aggressive type of endocervical adenocarcinoma. In this study, GAS accounted for 10.6% of all cervical adenocarcinoma with high prevalence of destructive invasion, extrauterine spread, and advanced stage at presentation.

ID 887
GANGLIOGLIOMA ARISING IN A OVARIAN TERATOMA

L.J. Marin Torres[1], A. Santoro[1], G. Scaglione[1], P. Tralongo[1], N. D' Alessandris[1], F. Policardo[1], D. Arciuolo[1], M. Valente[1], M. Gessi[1], G. Zannoni[1]

[1]Unit of General Pathological Anatomy, Department for women's Health Sciences, Children and Public Health, Fondazione Policlinico Universitario "A. Gemelli" IRCCS, Rome, Italy.

Objectives: The correct select of immunohistochemistry in distinguishing the spectrum of Central Nervous System Tumors (CNS-type tumors) arising in the ovary is helpful to discriminate awide variety of histological entities.
Materials and methods: we analysed the case of a 23 years-old woman, primi-gravida, who presented a mass in the left ovary of 12x56x82mm during a planned gynecological us check-up. Serum level of CA19.9 was high, while CA125, CEA, and HE4 were within normal ranges. The patient had also a germline BRCA1 mutation. After pregnancy, she underwent a surgical laparoscopic procedure in order to remove the left adnexa. Multiple peritoneal and omental biopsies and a peritoneal washing were done.
Results: The lesion consisted of heterogenous tissue comprising epithelial tissue, scattered foci of cartilage and mature, architecturally disorganized nervous tissues, with small parts of choroid plexus. More cellular areas were also present and composed of a proliferation of glial astrocytic elements with abudant, dense and eosinophilic cytoplasm. By immunohistochemistry these elements were intensely positive for GFAP, vimentin, S100. Another cell population cell with neuronal normo-phenotype were observed (Synaptophysin+, Chromogranin A+ and Neurofilament protein+. $BRAF^{V600E}$ was wild type. No significant mitotic activity or areas of necrosis were detected. The histopathological and the immunohistochemical findings fit with the diagnosis of mature cystic teratoma, associated to a glioneuronal neoplasm compatible with ganglioglioma. Peritoneal biopsies and washing were negative.
Conclusions: A careful examination of nervous tissue in the context of a mature teratoma is mandatory to exclude, along to a clearly malignant neuroctodermal neoplasm, also a low gradeneoplasm In the case of a patient with neuroectodermal derived tumour arisen within a mature teratoma, a multidisciplinary approach for a correct management should be recommended, due to the complexity of the pathological background and the rarity of such neoplams.

ID 900
UTERINE LEIOMYOMAS WITH FH DEFICIENT MORFOLOGY. THE IMPORTANCE OF MORPHOLOGICAL-BASED SCREENING

M.G. Mastrosimini[1-2], G. Settanni[2], V. Viassolo[3], M. Ceccaroni[4], G. Mantovani[4], P.Antonini[1], G.Zamboni[1-2], A. Pesci[2]

[1] Department of Diagnostics and Public Health, Section of Pathology, University and Hospital Trust of Verona, Verona, Italy; [2] Unit of Anatomic Pathology, IRCCS Ospedale Sacro Cuore-Don Calabria, Negrar di Valpolicella, Verona, Italy; [3] Genetic IRCCS Ospedale Sacro Cuore-Don Calabria, Negrar di Valpolicella, Verona, Italy; [4] Gynecology IRCCS Ospedale Sacro Cuore-Don Calabria, Negrar di Valpolicella, Verona, Italy.

Objectives: Hereditary leiomyomatosis renal cell carcinoma (HLRCC) syndrome is caused by germline mutation of Fumarate Hydratase (*FH*) gene. This autosomal dominant disease predisposes to develop high grade renal cell carcinoma (RCC), cutaneous and uterine leiomyomas. FH deficient(FH-d) leiomyomas have well described morphological features: high cellularity, chain-like arrangements of tumor cells, palisading nuclear distribution, macro-nucleoli surrounded by a halo, cytoplasmic eosinophilic globules, staghorn-shaped vessels, and alveolar-pattern edema. All these aspects can be shared by FH-d syndromic and sporadic leiomyomas, the latter carry a somatic mutation in the *FH* gene. The aim was to determine the incidence of FH-d leiomyoma in our cohort and identify somatic and germline mutations.
Materials and methods: Between 2015 and 2022, 18 uterine leiomyomas with FH-d morphology were selected. All caseswere tested with FH immunohistochemistry (FH-IHC) (SantaCruz Biotechnologies). Patients were invited to mutation analysis of tumor tissue using NGS technologies. The detection of *FH* mutationdirected the patients to genetic counselling and germline test to find the same alteration.
Results: 15/18 cases had FH-ICH lost, 16/18 patients were enrolled for molecular studies: 5/16 had *FH* somatic mutation (4/5 had FH-IHC lost, 1/5 FH-IHC retained) and 1/16 had mutation in *PIK3CA*gene. 1/5 harbors double somatic mutations but one of them was detected on the germline test. 1/5 had no germline mutation, 1/5 denied the genetic test and 2/5 are still pending.
Conclusions: Pathology-based screening is important to select, among uterine leiomyomas, those cases for molecular studies. The survey of somatic mutation in sporadic cases can trigger the genetic counselling and focus the germline analysis on the known somatic variant.

ID 902
INTEGRATED CLINICOPATHOLOGICAL AND MOLECULAR ANALYSIS OF ENDOMETRIAL CARCINOMA: PROGNOSTIC IMPACT OF THE NEW ESGO-ESTRO-ESP ENDOMETRIAL CANCER RISK CLASSIFICATION

A. De Leo[1], M. Grillini[1], A. G. Corradini[1], F. Rosini[1], M. Ruscelli[1], M. Errani[1], T. Maloberti[1], S. Coluccelli[1], G. Tallini[1], D. Santini[1], C. Ceccarelli[1], D. de Biase[1]

[1] Pathology Unit, University of Bologna Medical Center, Bologna, Italy

Objectives: The ESGO/ESTRO/ESP committee has been recently proposed a new risk stratification system for endometrial cancer (EC) patients incorporating both clinicopathological and molecular characteristics.The study aims to compare the ESGO/ESTRO/ESP risk classification system with the previous 2016 risk classification.
Materials and Methods: The cohort included 187 consecutive patients with endometrial carcinoma. Immunohistochemistry (IHC) and Next-Generation Sequencing (NGS) were used to assign TCGA molecular EC subgroups: *POLE* mutant (POLE), mismatch repair deficient (MMRd), p53 mutant (p53abn), and no specific molecular profile (NSMP).
Results: TCGA class assignment of EC cohort: 7% *POLE* group, 31% MMRd group, 23.5% p53abn group, 38.5% NSMP group. In the 2020 risk classification system, 39.1% of patients were allocated to low riskcompared with 22.6% in the 2016 risk classification system, mainly due to reclassification of patientspreviously classified especially as high-intermediate risk. The recent 2020 guidelines revealed a total of 61 patients (32,6%) with a change in risk group in relation to the 2016 classification system: the shift was due to p53abn, *POLE* alterations and lymph vascular invasion. The application of the 2020 risk stratification system shows Kaplan-Meier curves with a more significant difference between the groups throughout survival.
Conclusions: In our cohort, the application of the new 2020 risk classification integrating clinicopathological and molecular parameters provided a more accurate identification of low-risk and high-risk patients, potentially allowing a more specific selection for post-operative adjuvant therapy. Integrated molecular classification is a promising tool for a better therapeutic management of patients.

ID 908
MORPHOLOGICAL AND IMMUNOHISTOCHEMICAL APPROACH TO THE DIAGNOSIS OF VULVO-VAGINAL STROMAL TUMORS

G. Angelico[1], S. Spadola[1], B. Morante[2], L. Salvatorelli[2], S. Marletta[3], G.Magro[2]

[1] Pathology Unit, Cannizzaro Hospital, 95126 Catania, Italy; [2] Department of Medical, Surgical Sciences and Advanced Technologies "G.F. Ingrassia", Anatomic Pathology, University of Catania, 95123 Catania, Italy; [3] Department of Diagnostics and Public Health, Section of Pathology, University and Hospital Trust of Verona, 37134 Verona, Italy

Objectives: The category of the "stromal tumors of the lower female genital tract" encompasses a wide spectrum of lesions with variable heterogeneity, which can be nosologically classified as deep (aggressive) angiomyxoma (DAM), cellular angiofibroma (CAF), angiomyofibroblastoma (AMFB) or myofibroblastoma (MFB). The present article focuses on the diagnostic clues of the stromal tumours of the lower female genital tract to achieve a correct classification.
Materials and Methods: A large series of surgically-resected vulvo-vaginal stromal tumors was retrospectively collected from the files of the University of Catania. The following immunohistochemical stainings were applied: vimentin, desmin, smooth muscle actin, h-caldesmon, HGMA, CD34 and S100. FISH analysis for the detection of FOX1, located on 13q14.11, was performed.
Results: Based on morphological and immunohistochemical features, the following tumors were identified: 36 cases of DAM; 10 cases of CAF; 11 cases of AMFB; 5 cases of myofibroblastomas(MFB). Based on our results, we suggest following recommendations: (i) the diagnosis should be mainly based on histological features in combination with clinical and macroscopic features; (ii) immunohistochemical analyses may be misleading for correct tumour classification due to the non-specific results in the different histotypes; (iii) in cases with ambiguous morphological and immunohistochemical features, FISH. analysis showing a 13q14 deletion is helpful in ruling out the diagnosis of DAM and AMFB; similarly, the detection of *HMGA2* rearrangements is helpful for the diagnosis of DAM.
Conclusion: Awareness by pathologists of the wide morphological and immunohistochemical spectrum exhibited by these tumours is crucial to achieve correct diagnoses and to avoid confusion with reactive conditions or other benign or malignant entities.

ID 916
ENDOMETRIAL GIANT CELL CARCINOMA: A SHOT IN THE DARK?

G. Scaglione[1], A.Travaglino[1], S. Sfregola[1], N. D'Alessandris[1], M. Valente[1], A. Santoro[1], F. Policardo[1], S. Paquini[1], D. Arciuolo[1], G.F.Zannoni[1]

[1] Unit of General Pathological Anatomy, Department for Women's Health Science, Children and Public Health. Fondazione Policlinico Universitario "Agostino Gemelli" IRCCS, Rome, Italy

Objectives: To describe the morphological and molecular features of a series of endometrial giant cell carcinoma (EGCC).

Materials and methods: Three patients (55 to 76 yo) with previous diagnosis of high-grade endometrial endometrioid carcinoma (stage Ia-Ib1, FIGO 2019), underwent to hysterectomy. The lesions had similar diameter (average 7 cm) and polypoid appearance.
Results: Histologically the main pattern consisted of dyscohesive, pleomorphic multinucleated giant cells with eosinophilic nucleoli and occasionally nuclear pseudoinclusion. The mitotic count was high, up to 80mitoses/10HPF and lymphovascular space invasion was present. All cases presented other histological component such as serous, low grade endometrioid and undifferentiated carcinoma with mononucleated cells. Immunohistochemically the giant cells stained for AE1/AE3, EMA, p16 and ER, PR, while PAX8 and E-cadherin were focally positive. αFP, CD68, βHCG were negative. The cases were mismatch repair-proficient, one tumor had p53 aberrant staining. No POLE mutation was detected.
All patients received adjuvant treatment (carbotaxol and external beam radiation therapy). After 12, 8 and 3 months of follow up they were alive without recurrences.
Conclusions: Giant cell component in endometrial cancer was previously described in different reports, although the current WHO does not consider it as a separate entity. Generally, the EGCC is associated with conventional neoplastic areas, serous or low grade endometrioid carcinoma. These features force the pathologists to rule out other entities, mostly endometrial dedifferentiated/undifferentiated carcinoma (UDEC/DDEC) and carcinosarcoma (ECS). Themorphological features combined with the molecular signatures should draw to the correct diagnosis. While histologically our data are too limited to better define this tumor. Clinically as stated by literature and according to the morphological feature we observed, EGCC seems to have an aggressive behavior and should be managed as UDEC/DDEC and ECS.

ID 943
LEIOMYOADENOMATOID TUMOR OF THE UTERUS: DESCRIPTION OF SEVEN CASES

E. Olmeda[1], L. Runza[1], A. Palicelli[2], S. Ferrero[1,3], E. Kuhn[1,3]

[1] Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico, Milano, Italy; [2] Pathology Unit, Azienda USL-IRCCS di Reggio Emilia, Reggio Emilia, Italy; [3] Dipartimento di Scienze Biomediche, Chirurgiche ed Odontoiatriche, Università degli Studi di Milano, Milano, Italy

Objectives: Adenomatoid tumors are benign neoplasms of mesothelial origin that occur more frequently in the genital tract of both women and men. Rarely, the adenomatoid tumor is closely associated with a leiomyomatouscomponent and is called leiomyoadenomatoid tumor.
This study aims to describe the clinical and immunomorphological features of leiomyoadenomatoid tumors.
Materials and methods: All cases of concurrent leiomyoma and adenomatoid tumor of the uterus between 2010-2021 were retrospectively collected from the Pathology archive of our Institution. All available slides were reviewed, and clinico-pathological features were collected. Immunohistochemical stainings for caldesmon, actin, cytokeratin pool, and calretinin were performed.
Results: A total of seven leiomyoadenomatoid tumors were identified. Patient age ranged from 24 to 46 years, median 40 years. All patients underwent surgery for fibroids. Specifically, 5 patients underwent nodulectomy and 2 hysterectomy. Macroscopically, all cases were described as white, fibrous nodules, with a hard-elastic consistency, a cut surface fasciculated. The nodules measured between cm 1.2 and cm 8.5 of major axis (median cm 2.8). Microscopic examination revealed a dominant benign smooth muscle proliferation compatible with leiomyoma. Moreover, an intermingled component formed by tubular and/or microcystic structures lined by bland cuboidal or flat epithelioid cells with clear cytoplasm was present. No significant cytological atypia and mitotic figures were found. The ratter component was a minority. The immunohistochemistry confirmed the mesothelial differentiation of the epithelioid component (cytokeratin/calretinin+) and the smooth muscle differentiation of the main component (caldesmon/actin+).
Conclusions: Leiomyoadenomatoid tumor is a rare and benign lesion characterized by an adenomatoid proliferation interspersed by a prominent smooth muscle component. It can be overlooked and diagnosed as a leiomyoma or overestimated as a malignant tumor, due to the pseudoinfiltrative appearance. Most authors believe that it represents a variant of adenomatoid tumor that penetrates normal or hyperplastic smooth muscle fascicles, which conceal the mesothelial tumor. On the other hand, it could be a collision tumor, namely the compresence and fusion of two benign neoplasms, an adenomatoid tumor and a leiomyoma. Further studies are needed to clarify the biological nature of this lesion.

Sabato 15 Ottobre 2022

Sala A 08.30 - 09.00

PATOLOGIA SPERIMENTALE

Moderatore: M. Ponzoni

ID 812
A NEW PROTOCOL FOR HISTOPATHOLOGICAL ANALYSIS OF HUMAN LUNG TUMOROIDS: A PROOF-OF-CONCEPT

M. Mandarano[1], C. Floridi[1], L. Annese[1], C. Pelliccia[1], S. Ceccarelli[2], G. Bellezza[1], F. Puma[2], T. Zelante[3], A. Sidoni[1]

[1] Section of Anatomic Pathology and Histology, Department of Medicine and Surgery, University of Perugia, Italy;[2] Section of Thoracic Surgery, Department of Medicine and Surgery, University of Perugia, Italy; [3] Section of Pathology, Department of Medicine and Surgery, University of Perugia, Italy

Objectives: tumoroid is an innovative 3D cell culture-based technology to study primary tumor epithelial cells simulating in vivo condition.
Although the morpho-functional evaluation of tumoroids can be based on various methods, obtaining paraffin-embedded preparations for reliable histopathological and immunohistochemical analyses is still far from being standardized.[1]
For this reason, we have developed a proof of concept attempts to produce a satisfactory and reproducible protocol using lung carcinoma derived tumoroids.
Materials and Methods: four neoplastic samples from resected human lung carcinomas were collected to obtain tumoroids. After 8, 14 days from the beginning of the culture and 14 days after a first tumoroids splitting, they were harvested, rapidly fixed in neutral buffered formalin for 30', gently centrifuged at 200 rcf/g for 5'. The cell precipitate obtained was resuspended in PreservCyt® solution and processed according to Cellient® (Hologic) protocol, to automated prepare cell blocks.
Afterwards, 4-µm-thick sections were stained with H&E and immunohistochemically for cytokeratin 7, TTF-1, p53 on fully automated immunostainer BOND-III (Leica Biosystems).
Results: tumoroids of 2 (50%) patients were viable after the predetermined days from the initial culturing. Microscopically, the obtained tumoroids are well organized (H&E) and showed positivityfor TTF-1 and cytokeratin 7, as the original "donor" tumors.
Conclusions: using Cellient®, histological sections are faster and more standardized obtained than agar-or gel-based protocols.
These results appear very promising in order to study larger series of tumoroids for an *ex vivo*reconstruction of the biological characteristics of human tumors.

References

1. Zhang SW, Chen W, Lu XF, et al. An efficient and user-friendly method for cytohistological analysis of organoids. J Tissue Eng Regen Med 2021;15(11):1012-1022. https://doi.org/10.1002/term.3248

ID 910
PREVALENCE OF VESSELS ENCAPSULATING TUMOR CLUSTERS (VETC) IN HUMAN CANCERS

C. De Carlo[1,2], R. Akpinar[3], M. Coto-Llerena[4,5], L.M. Terracciano[1,2], S. Piscuoglio[4,5], L. Di Tommaso[1,2], S.L. Renne[1,2]

[1] Department of Pathology, IRCCS Humanitas Clinical and Research Hospital, Rozzano, Italy; [2] Humanitas University, Department of Biomedical Science, Pieve Emanuele, Italy; [3] Department of Pathology, KanuniSultan Süleyman Training and Research Hospital, Istanbul/Türkiye; [4] Visceral Surgery and Precision Medicine Research Laboratory, Department of Biomedicine, University of Basel, Basel, Switzerland; [5] Institute of Medical Genetics and Pathology, University Hospital Basel, Basel, Switzerland

Objectives: VETC shields tumor-cell clusters, promoting free circulation in the bloodstream. This mechanism of metastasization is an alternative to the epithelial-to-mesenchymal transition process and has been recently described in hepatocellular carcinoma (HCC), however the prevalence of VETC among cancers is unknown.
Materials and methods: We assessed the VETC prevalence on a retrospective series of 1861 neoplastic samples from 1796 patients comprising 24 different histologies. VETC was defined as continuous endothelial covering of neoplastic clusters and highlighted by CD34 immunohistochemistry. Analysis was performed on a tissue microarray by experienced pathologists blinded to clinical and pathological data.
Results: VETC was present in 73/1769 cases (4%) and in 10/23 histotypes (44%). The histotypes that had VETC+ cases were renal cancer (RCC) 29/115 (25%), HCC 23/102 (23%), prostate cancer (PRC) 7/42 (17%), mesotheliomas 2/17 (12%), pancreatic cancer 4/84 (5%), esophageal cancer 2/72 (3%), and gastric cancer 3/158 (2%). Colorectal, extrahepatic bile duct, and urinary bladder cancer had one VETC+ case each. Cases with VETC+ tended to be associated to the intermediate histologic grade (G2; $p < 0.0001$). Age, size, survival, and status were similar between the two groups.
Conclusions: VETC is present across different histotypes and might represent an alternative mechanism of metastasis. Even if present in a minority of cases, VETC was peculiar of some histotypes (especially HCC, RCC, PRC), possibly representing a histotype-specific angiogenetic mechanism. Since VETC predicts response to tyrosine kinase inhibitors, further studies are warranted to assess the prognostic and predictive value of this peculiar vascularization type.

ID 950
A NEW TOOL FOR INVESTIGATION PLATELET ACTIVATION IN ENDOMETRIOSIS PATIENTS

A. Mangogna[1], B. Belmonte[2], B. Bortot[1], G. Zito[1], D. Vacca[2], A. Romano[3], F. Valle[4], F. Zanconati[3,5], G. Ricci[1,5] and S. Biffi[1]

[1] Institute for Maternal and Child Health, IRCCS Burlo Garofolo, Trieste, Italy; [2] Tumor Immunology Unit, University of Palermo, Italy; [3] UCO Anatomia e Istologia Patologica, ASUGI, Hospital of Cattinara, Trieste, Italy; [4] CNR, Istituto per lo Studio dei Materiali Nanostrutturati, Bologna, Italy; [5] Department of Medical, Surgical and Health Science, University of Trieste, Italy

Objectives: Endometriosis (EM) is a gynecological disease characterized by chronic inflammation, due to the interaction of inflammatory cells with ectopic endometrium [1]. Platelets (PLTs), recruited by procoagulant factors released from endometriotic stromal cells, secrete angiogenetic factors and induce overexpression of genes involved in pro-survival/anti-apoptotic propensity, inflammationand extracellular matrix remodeling [2]. We aimed to develop a tool to measure PLT activation (by small extracellular vesicles, s-EVs) in EM peritoneal fluids, as a potential predictive marker of EM severity.
Materials & methods: S-EVs were isolated from EM peritoneal fluids and characterized with imaging (Atomic Force Microscopy; AFM) and protein expression analyses (Western blot, WB) [3]. We explored gene expression in peritoneum and EM lesions using EndometDB [4].
Results: We demonstrated the presence of s-EVs isolated from EM peritoneal fluids by liquid AFM, as showed by contact angle *vs* diameter scatterplot (Fig.1A-B), and by WB detecting the s-EV markers CD63, CD9, and TSG101 (Fig.1C). Using EndometDB, we highlighted the differentially expressed genes between control and EM peritoneum samples (Fig.1D). The protein expression of a panel of biomarkers of PTL in s-EVs was further confirmed by WB (Fig.1E).
Conclusions: We propose applying s-EV research to EM investigation, generating a novel biochemical tool for PLT activation assessment and for the development of new diagnostics and therapies.

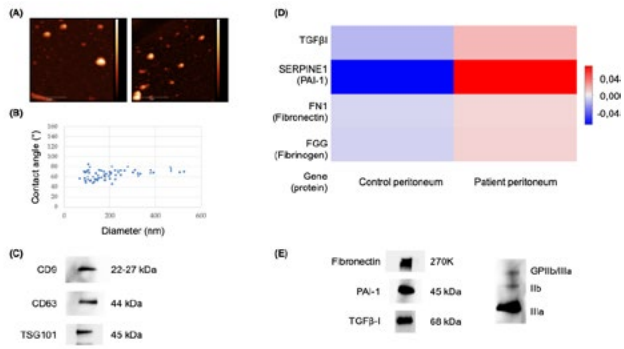


Figure 1. A) Representative liquid AFM micrographs of s-EVs isolated from the peritoneal fluid of an EM patient. B) Contact angle vs diameter scatterplot of s-EVs. Each circle represents one individual s-EVs as measured via AFM imaging in liquid. C) CD63, CD9, and TSG101 were detected by WB in s-EVs samples. D) Heatmap visualizing the expression of genes across the control and EM patient's peritoneum samples. Data are collected from the EndometDB. E) Protein expression of a panel of biomarkers in s-EVs.

References

1. Agostinis C, Balduit A, Mangogna A, et al. Immunological Basis of the Endometriosis: The Complement System as a Potential Therapeutic Target. Front Immunol 2020, 11, 599117, https://doi.org/10.3389/fimmu.2020.599117.
2. Ding D, Liu X, Duan J et al. Platelets are an unindicted culprit in the development of endometriosis: clinical and experimental evidence. Hum Reprod 2015, 30, 812-832, https://doi.org/10.1093/humrep/dev025.
3. Bortot B, Apollonio M, Rampazzo E, et al. Small extracellular vesicles from malignant ascites of patients with advanced ovarian cancer provide insights into the dynamics of the extracellular matrix. Mol Oncol 2021, 15, 3596-3614, https://doi.org/10.1002/1878-0261.13110.
4. Gabriel M, Fey V, Heinosalo T, et al. A relational database to identify differentially expressed genes in the endometrium and endometriosis lesions. Sci Data 2020, 7, 284, https://doi.org/10.1038/s41597-020-00623-x.

Sabato 15 Ottobre 2022

Sala D 08.00 - 09.00

DIGITAL PATHOLOGY

Moderatore: F. Fraggetta

ID 756
AI-BASED SOLUTION FOR SUPPORTING PRIMARY DIAGNOSIS OF PROSTATE BIOPSIES IN ROUTINE PRACTICE

G. Mallel[1], D. Raoux[2], G. Sebag[1], M. Vecsler[1], M. B. Amin[3], E. Comperat[4], C. Linhart[1], J. Sandbank[1,5]

[1] Ibex Medical Analytics, Tel Aviv, Israel; [2] MediPath, Frejus, France; [3] Department of Pathology, University of Tennessee Health Science Center, Memphis, USA; [4] Department of Pathology, Medical University of Vienna, Vienna, Austria; [5] Institute of Pathology, Maccabi Healthcare Services, Rehovot, Israel

Objectives: We aimed to clinically validate the use of an AI-based tool by pathologists for reviewing and reporting prostate core needle biopsies (PCNBs) as compared with Standard of Care review on microscope, also assessing improvements in efficiency and turnaround time.

Design: A two-arm study in which the standard of care arm (using a microscope) was compared with an arm in which pathologist conducted the reporting using an AI-based solution workflow. Eight pathologists from six different sites in a large network of pathology labs participated and reported on 3,196 slides from 420 prostate core needle biopsies. Each case was reported twice, both with microscope and with the AI-based solution randomized between pathologists. Both arms were compared to ground truth (GT) established by consensus of two subspecialist uropathologists. Rates of major discrepancies between each arm and GT, as determined by an adjudicating expert uropathologist, were compared. Detailed time measurements were taken for each step and compared between study arms, as was TAT.
Results: The major discrepancy rates of the microscope and of the AI-based solution arms against GT were 7.15% and 4.84%, respectively. The AI solution demonstrated very high performance on prostate cancer detection with AUC=0.99, sensitivity of 95.5% and specificity of 96.2%. Pathologists with the AI-based solution demonstrated a decrease in review and reporting time, leading to 37% efficiency gains and a significant decrease of 1-2 days in TAT. Other endpoints included sensitivity and specificity of both arms on cancer detection and grading.
Conclusion: Here we reported on a large-scale study, in which pathologists performed full primary diagnosis with the support of an integrated AI-based solution in a real world-like clinical setting, in a large network of pathology laboratories. Importantly, diagnostic accuracy improvements were observed in addition to significant efficiency gains for the pathologists reviewing and reporting with the support of the AI solution.
Moreover, the overall user experience, as reported by pathologists, was markedly better with the AI solution compared to a microscope.

ID 829
RECURRENCE-FREE SURVIVAL PREDICTION THROUGH A DEEP LEARNING MODEL USING MELANOMA WHOLE SLIDE IMAGES

C. Traversi[1], M. C. Comes[2], S. Bove[2], L. Fucci[1], R. Massafra[2], F. Mele[1], S. Strippoli[3], I. Trotti[1], M. Guida[2], A. Zito[1]

[1] Unità Operativa Complessa di Anatomia Patologica, I.R.C.C.S. Istituto Tumori "Giovanni Paolo II", Bari, Italy; [2] Unità Operativa di Fisica Sanitaria, I.R.C.C.S. Istituto Tumori "Giovanni Paolo II",Bari,Italy; [3] Unità Operativa di Tumori Rari e Melanoma, I.R.C.C.S. Istituto Tumori "Giovanni Paolo II", Bari, Italy

Objective: Whole-slide histological images (WSIs) can provide insights for clinical and basic science investigations of melanoma tumors[1]. The identification of quantitative imaging biomarkers for the prediction of recurrence-free survival (RFS) in melanoma patients is crucial to improve and accelerate patient management.
Methods and materials: In this study, we developed an artificial intelligence-based method that can recognize prognostic biomarkers from WSIs of primary melanoma tumors with the aim to predict RFS in melanoma patients. A total of 43 WSIs of primary melanoma tumors from the Clinical Proteomic Tumor Analysis Consortium Cutaneous Melanoma (CPTAC-CM) public database[2] with known 1-year RFS (31 RFS cases, 12 non-RFS cases) were segmented into 12575 crops. These crops were used to train and validate the proposed model by means of a 5-fold cross validation scheme for 5 rounds.
Results: The best Area Under the Curve (AUC) and accuracy were achieved by combining quantitative imaging biomarkers extracted by Convolutional Neural Networks (CNNs) composing the proposed model with some clinical data. An AUC value of 70.1 3.0% and an accuracy value of 72.7 6.8%, were reached respectively.
Conclusions: Overall, promising results for the RFS prediction task were obtained by extracting information from WSIs directly. This study provides a valuable basis for future research investigation on wider cohorts of patients registered at our Institute.

References

Brinker TJ, Kiehl L, Schmitt M, et al; Deep learning approach to predict sentinel lymph node status directly from routine histology of primary melanoma tumours. Eur J Cancer 2021,154:227–234. https://doi.org/10.1016/j.ejca.2021.05.026

https://wiki.cancerimagingarchive.net/display/Public/CPTACCM#33948224bcab02c187174a288dbcbf95d26179e8

ID 832
QUANTIFYING THE IMPACT OF COLOR NORMALIZATION IN COLORECTAL CANCER HISTOLOGY CLASSIFICATION

S. De Summa[1*], M. Caputo[1*], T.M. Marvulli[1*], B. Prencipe[2], A. Brunetti[2], V. Bevilacqua[2], E. Mattioli[1], N.Altini[2], S. Tommasi[1], F.A. Zito[1]

[1] IRCCS-Istituto Tumori "Giovanni Paolo II", Bari, ITALY; [2] Polytechnic University of Bari, Bari, ITALY

Objectives: The digitalization of health data involves the managing and development of algorithm able to support the clinicians. In particular, digital pathology has led to the advent of a new omic science, the so-called "pathomics".
By applying deep-learning (DL) methods to histopathology in colorectal cancer (CRC), we investigated how DL can support the pathologists in recognizing different tissue types on Whole slide images (WSI). An important obstacle to overcome is stain color variations of the standard hematoxylin and eosin (H&E) sections of FFPE samples among different laboratories.

In this work, we addressed this issue by comparing classic image processing and DL methods with respect to the downstream task of CRC tissue multi-class classification.
Materials and methods: The task of multi-tissue classification involved the segmentation of the following histotypes: tumorepithelium, stroma and smooth muscle tissue, immune-cell conglomerates, debris and mucus, normal mucosa, adipose tissue and background.
A publicly available dataset from NCT-CRC-HE-100K dataset[1] (n = 77,805) has been exploited for training the multi-class classification models. External validation has been conducted on an internal dataset collected and annotated by an expert pathologist (n = 15,856).
The image processing methods considered for normalization are Macenko, Reinhard, Vahadane and Khan. DL methods, based on Generative Adversarial Network (GAN) include CycleGAN, GcGAN, CUT, FastCUT and AIFFPE.
To train GANs we used 150k tiles both from the TCGA domain and the internal cohort, distributed in 100k for training and 50k for testing in both domains.
To perform classification, transfer learning has been used by comparing three different pre-trained CNN architectures (InceptionV3, DenseNet201, VGG16) to extract features, that were the inputs for Support Vector Machines.
Results: The results show that the DL method FastCUT outperformed the classic image processing methods in classification tissue task, reaching an overall accuracy of 0.8420 using DenseNet201 features.
Conclusions: GANs could represent a method able to overcome the staining variations. Blinded discrimination of fake/real images, generated by GANs, is currently ongoing by the support of two expert pathologists.

References

1. J.N. Kather, N. Halama, A. Marx (2018). 100,000 histological images of human colorectal cancer and healthy tissue (v0.1) https://doi.org/10.5281/zenodo.1214456

ID 850
ESTABLISHING QUANTITATIVE IMAGE ANALYSIS METHODS FOR TUMORMICROENVIRONMENT EVALUATION

C. Ercan[1,2], M. Coto-Llerena[1,2], S. Piscuoglio[1,2], L. M. Terracciano[1,3]

[1] Institute of Pathology, University Hospital Basel, Switzerland; [2] Visceral Surgery and Precision Medicine Research Laboratory, Department of Biomedicine, University of Basel, Switzerland; [3] Unit of Anatomic Pathology, Humanitas University, Pieve Emanuele, Milan, Italy

Objective. Tumor microenvironment (TME) evaluation requires a combination of cell type and spatial information. Computational methods such as artificial intelligence(AI)-based tools, may expedite the detection and classification of thousands of different cells on immunohistochemistry (IHC) slide images, expanding our understanding of the TME. Here, we aim to develop an AI based image analysis pipeline to define morphological and immunological characteristics of liver tumor TME.
Materials and Methods: We collected 50 HCC samples from liver resections. Tumorslides were stained with a panel of TME markers by IHC. The slides were digitalised and whole slide images were used for the quantification. For the quantification of immune cells, we trained two separate convolutional neural networks (CNN): cell detection and tumor-stroma segmentation. Cell nucleus instance segmentation was achieved using StarDist package. We trained a model with our slides and tested over the pretrained model. Immune cells were classified using an artificial neural network (ANN) classifier in QuPath. Finally, we trained another CNN in UNET architecture for semantic segmentation of tumor tissue into parenchyma, stroma, and debris classes, by fastaideep learning library.
Results: The cell segmentation model reached 87% accuracy, 92% F1-Score, 92% true positive, 94% true negative rates on IHC slide images, while the pretrained model had only 66% accuracy. The ANN classifier annotated immune cells at 98% accuracy. The CNN tumor segmentation model classified tumor regions into parenchyma, stroma, and debris at 96% accuracy, 93% dice, 86% Jaccard index.
Conclusions: In this study, we developed a pipeline implementing open-source software to quantify IHC slides. The use of this semi-automatized computational pathology workflow can provide robust information in regard to the TME composition augmenting the discovering tumour specific TME features and pave the way for the discovery of novel prognostic and therapeutic targets.

ID 927
AN AI-BASED APPROACH TO AUTOMATICALLY CLASSIFY OSCC HISTOPATHOLOGY IMAGES.

F. Merolla[1], G. Ilardi[2], D. Russo[2], S. Varricchio[2], V. Strazzullo[2], R. Alfano[2], Francesco Martino[3],Stefania Staibano[2]

[1] Department of Medicine and Health Sciences "V. Tiberio", University of Molise, Campobasso, Italy; [2] Department of Advanced Biomedical Sciences, Pathology Unit, University of Naples "Federico II", Naples, Italy; [3] Dedalus HealthCare, Division of Diagnostic Imaging IT, Vienna, Austria

Objectives: Oral squamous cell carcinoma (OSCC) is the most common malignant tumor of the head and neck district. To the best of our knowledge, few studies are reported in the literature concerning the use

of AI-based approaches for the analysis of the histological samples of these tumors [1]. We developed an AI tool to classify WSIs for the purpose of prioritizing cases of OSCC.
Materials and methods: We selected 390 OSCC images from the TCGA image database and the archives of Federico II Pathology Unit in Naples. The selected images were cropped and scaled to 228px x 228px with an RGB color profile using OpenCV. Following data augmentation, we splitted the dataset in Train, Validation and Test set, then a model was developed to classify neoplastic from non-neoplastic lesions. The metric used was Area Under Curve. We built the model on normal and histogram matching normalized images, following a data augmentation step.
Results: The results show that, although the normal RGB images trained model had a better AUC, training the network on normalized images produced a more robust model because it was less affected by overfitting.In order to improve the metrics of our model, we contaminated the dataset with squamous cell carcinomas of other anatomical sites (skin, cervix). Our preliminary results demonstrated a significant increase in sensitivity. Final results will be discussed.
Conclusions: The CNN obtained represents an AI tool that efficiently prioritizes OSCC cases starting from normalized images. We successfully employed dataset contamination to improve the ability to correctly classify tumorcontaining samples. As a computer-aided diagnostic tool, we envisage its employment in a pathologist's daily routine to prioritize the work allowing the pathologist to focus first on more urgent cases.

References

1. Alabi RO, Youssef O, Pirinen M, et al.. Machine learning in oral squamous cell carcinoma: Current status, clinical concerns and prospects for future-A systematic review. Artif Intell Med. 2021 May;115:102060. https://doi.org/10.1016/j.artmed.2021.102060. Epub 2021 Mar 26. PMID: 34001326.

ID 951
COMPARATIVE EVALUATION BETWEEN LIGHT MICROSCOPY AND DIGITAL IMAGING OF LUNG CANCER WITHLOW IMMUNOHISTOCHEMICAL EXPRESSION OF PD-L1

F. Crivelli[1], F. Sessa[2], C. Patriarca[3], G. Arpa[4], S. Zannella[3], M. Cerati[2], G. Lo Bello[3], A.F. Ussia[3]

Department of Pathology [1] ASST Valle Olona, Busto Arsizio; [2] ASST Sette Laghi, Varese; [3] ASST Lariana, Como; [4] ICS Maugeri, Pavia

Within the Project "Use of Digital Pathology in histological diagnosis in the territory of ATS Insubria", in order to verify the quality and feasibility of digital imaging in the evaluation of PD-L1 positivity in lung cancer with a low immunohistochemical expression (TPS ≤ 1%), 30 cases with the above features were isolated from the archives of the Pathological Anatomy wards of ASST Lariana and ASST Sette Laghi, digitized using 20x Hamamatsu Nanozoomer S360 scanner and made available for digital viewing.
Eight pathologists of the above-mentioned institutions, including one from ASST Valle Olonaand one from ICS Maugeri co-opted as partners, performed a double-blinded comparative evaluation of the selected histological slides on light microscopy at first and, subsequently, on digital imaging.
The results demonstrated a good agreement in the evaluation of PD-L1 expression on light microscopy ($\kappa = 0.98$); on the contrary, the assessment of PD-L1 immunostaining on digital slides resulted more problematic, mostly due to some issues regarding the quality of digitized images.
In the light of these results, it is considered necessary (i) to evaluate mainly by microscopy lung carcinomas having a low percentage of PD-L1 reactivity, as, in these cases, digitization processes may prevent an optimal visualization of the histologic section, and (ii) to focus on training the pathologist in the interpretation of digital slides. This will make, in the future, both microscopy and digital imaging comparable and equally reproducible.

Sabato 15 Ottobre 2022

Sala E 08.00 - 09.10

PATOLOGIA TESSUTI MOLLI E DELL'OSSO

Moderatore: G. Pennelli

ID 802
EPITHELIOID SARCOMA: A MORPHOLOGICAL AND IMMUNOHISTOCHEMICAL EVALUATION OF 22 CASES

N. Narducci[1], M. Sbaraglia[1,2], A. P. Dei Tos[1,2]

[1] Department of Medicine, University of Padua School of Medicine, Padua, Italy; [2] Department of Pathology, Azienda Ospedale-Università Padova, Padua, Italy

Objectives: Epithelioid sarcoma is an extremely rare sarcoma, occurring predominantly in adolescents and young adults. Epithelioid sarcoma is classified in two variants: distal and proximal type, that are characterized by distinct morphology, anatomic distribution and prognosis[1]. For patients with metastatic disease treatment options are rather limited. Very recently in single cases activity of immunotherapy have been reported. We herein investigate, immunohistochemi-

cally in both subtypes of ES, the presence of immune infiltrate. Furthermore, as the expression of GATA3 has been observed in sarcomas, including ES, we explored its potential role as diagnostic marker[1].
Materials and methods: Twenty-two cases of ES were retrieved from the consultation files of one of the authors (ADT). Only INI1-deficient cases were included. Based on morphology, the cases were subclassified in proximal ES (9) and classic ES (13). Whole sections from formalin-fixed paraffin-embedded material were immunostained for CD20, CD3, CD4, CD8, CD68, PD-L1 (22C3) and GATA3. The immune infiltrate was evaluated by an Artificial Intelligence-driven software (Visiopharm).
Results: An increased number of lymphocytes was seen in the classic variant of ES, mostly composed of CD3+/CD8+ elements, with an average of lymphocytes CD3+/mm^2 in classic ES of 550/mm^2 vs. 257/mm^{2}in proximal ES. Focal positivity for PD-L1 in neoplastic cells and/or lymphocytes was seen in only 2 cases of proximal type ES. All classic ES were negative for PD-L1. Strong and diffuse nuclear expression of GATA3 was seen in proximal ES (5/8 cases).
Conclusions: Classic and proximal ES feature a distinct immune-landscape, with distal ES showing a greater activation of inflammatory response, opening opportunity for immune therapy. Furthermore, with the limits of small sample, expression of GATA3 may represent a helpful diagnostic marker to separate distal and classic ES.

References

1 Frezza AM, Sbaraglia M, Lo Vullo S et al. The natural history of epithelioid sarcoma. A retrospective multicentre case-series within the Italian Sarcoma Group. Eur J Surg Oncol. 2020 Jul;46(7):1320-1326 https://doi.org/10.1016/j.ejso.2020.03.215.

ID 805
A VERY RARE CASE OF SPINDLE CELL SARCOMA WITH *MEIS1:NCOA2* FUSION GENE AND REVIEW OF THE LITERATURE

B. Del Forno[1,2], M. Sbaraglia[1,2], F. Cappello[1,2], I. Tortorelli[3], A. Brunello[3], A. Dei Tos[1,2]

[1] Department of Pathology, Azienda Ospedale Università Padova, Padua, Italy; [2] Department of Medicine, University of Padua School of Medicine, Padua, Italy; *[3] Oncology 1 Unit, Department of Oncology, Veneto Institute of Oncology IOV-IRCSS, Padua, Italy*

Objectives: In the NGS' era several "new entities" are emerging. Many cases previously misdiagnosed or not recognized have been molecularly reclassified. Very recently a new group of sarcomas with *MEIS1::NCOA1/2* fusion gene were described. To date only eight cases arising in the urogenital tract have been reported[1,2]. Clinically, these neoplasms are characterized by local recurrences, whereas distant metastases have not been reported.
Materials and methods: We report a case of a low-grade spindle cell sarcoma of the pararectal region retrieved from the consultation cases of one of the authors (ADT), in which NGS-based molecular analysis identified a *MEIS1::NCOA2* fusion transcript.
Results: Our patient was a 33-year-old pregnant female with a 10 cm mass involving the pararectal region that MRI described as partially cystic. A marginal resection of the tumor was performed without previous biopsy. Histologically, the tumor was composed of plump spindle cells arranged in a storiform pattern and set in a myxoid stroma. The immunophenotype was not distinctive. Molecular analyses detected a *MEIS1::NCOA2* gene fusion. A local recurrence occurred at six months after surgical resection, nevertheless no distant metastasis occurred after two years.
Conclusions: We described an extremely rare case of low-grade spindle cell sarcoma with *MEIS1::NCOA2* gene fusion showing an indolent clinical behavior. Proper recognition of these rare emerging entities is crucial to better understand their clinical behavior and define the best therapeutic approach.

References

[1] Argani P, Reuter V, Kapur P, et al. Novel *MEIS1-NCOA2* Gene Fusions Define a Distinct Primitive Spindle Cell Sarcoma of the Kidney. Am J Surg Pathol 2018;42(11):1562-1570. https://doi.org/10.1097/PAS.0000000000001140

[2] Kao Y, Bennet J, Suurmeijer A, et al. Recurrent *MEIS1-NCOA2/1* fusions in a subset of low-grade spindle cell sarcomas freqeuently involving The genitourinary and gynecologic tracts. Mod Pathol, 2021;34(6); 1203-1212. https://doi.org/10.1038/s41379-021-00744-7.

ID 860
AN APP TO PREDICT MITOTIC COUNT ON THE SURGICAL SPECIMEN.

S.L. Renne[1,2], M. Cammelli[1], I. Santori[1], M. Tassan-Mangina[1], L. Ruspi[3], S. Federico[3], P. Colombo[1,2], V. Quagliuolo[3], L.M. Terracciano[1,2], F. Cananzi[1,3]

[1] Department of Biomedical Sciences, Humanitas University, Pieve Emanuele MI, Italy; [2] Pathology Unit, IRCCS Humanitas Research Hospital, Rozzano MI, Italy; [3] Sarcoma Unit, IRCCS Humanitas Research Hospital, Rozzano MI, Italy.

Objectives: Preoperative risk stratification in Gastrointestinal Stromal Tumors (GISTs) is important in determining the indication to neoadjuvant therapy. Size and site can be defined by imaging, whereas mitotic count is done on biopsy. The latter is burdened by some limitations mainly due to tumor heterogeneity and sampling bias. We aim to build an App that, accounting for these limitations, could allow a more accurate stratification of patients in preoperative settings.

Materials & methods: As modeling strategy, we used a multilevel hierarchical model; to select variables to train the model, we created a Directed Acyclic Graph (DAG) of the available clinico-pathological variables. Given the DAG, to estimate the mitotic count we identified as variables: tumor size, tumor site, mitotic count on biopsy, surface area assessed on biopsy andtumor response to therapy – if performed. We performed prior predictive simulations, tested the model on a mock dataset and finally trained on real GIST cases with paired biopsy and surgery (n = 80) from IRCCS Humanitas Research Hospital (a total of 160 cases) after mitotic count re-evaluation and measurement of the biopsies' surface.
Results: Model diagnostics were satisfactory. To choose the best model we estimated model performance through Widely Applicable Information Criteria and Pareto Smooth Important Sampling Leave-One-Out Cross-Validation Criteria. We selected the model with the lower deviance in out of sample performance. Posterior predictive check was used to highlight model performance against ground truth. We built an app that used the posterior probability generated by the model to dynamically compute the number of mitoses on the surgical specimen given tumor size, site and surface of, and mitotic count on, biopsy.
Conclusions: We created an App that predicts mitotic count on the surgical specimen. This allows a better preoperative risk stratification of GISTs, leading to a tailored treatment approach.

ID 864
PROGNOSTIC VALUE OF VESSELS ENCAPSULATING TUMOR CLUSTERS (VETC) IN SARCOMA

D. Corbitt[1], M. Tassan-Mangina[1], M. Cammelli[1], L. Ruspi[3], S. Federico[3], P. Colombo[1,2], V. Quagliuolo[3], Di Tommaso L[1,2], F. Cananzi[1,3], S.L. Renne[1,2]

[1] Department of Biomedical Sciences, Humanitas University, Pieve Emanuele MI, Italy; [2] Pathology Unit, IRCCS Humanitas Research Hospital, Rozzano MI, Italy; [3] Sarcoma Unit, IRCCS Humanitas Research Hospital, Rozzano MI, Italy

Objectives: How sarcoma metastasize is unknown. VETC has been described as an epithelial-to-mesenchymal- transition independent process of metastasis: endothelium covers neoplastic clusters allowing tumor dissemination. Our aims are to assess the presence of VETC in sarcoma, and to model its prognostic role.
Materials & methods: The study was retrospective. We selected 54 cases of sarcomas (6DDLPS, 10 GIST, 6 LMS, 9 MLPS, 8 MPNST, 10 SFT, 5 UPS); of them 31 were metastatic (M1 group), 23 were not (M0 group, defined as least 5 years of negative follow-up). VETC was assessed with CD31 immunohistochemistry and defined as a continuous endothelial lining around tumor clusters. We used probabilistic modeling for the analysis.
Results: Within each histology, the two groups (M0 & M1) were substantially homogeneous: most (89%CI) of posterior probability (PP) difference – i.e. the contrast CPP – included 0 for sex, age, size, and grade. VETC in SFT was basically only expressed in M1, with almost all the CPP mass above the 0. Also, in UPS and GIST, VETC was more probable to be in metastatic diseases with 79% and 78% respectively of the CPP mass above 0. VETC was prognostic of metastasis free survival in SFT and UPS with a coefficient of 2.42 (CI 0.73–4.65) and 1.94 (CI 0.16–3.67); only UPS reached median survival of 65 months(mo) (standard deviation, SD:74 mo) for VETC- Vs 11 mo (SD:14 mo) for VETC+.
Conclusions: VETC was present in all the investigated histotypes but two (MLPS, MPNST). VETC was prognostic of disease-free survival in UPS and SFT. These findings warrantconfirmations on a larger series. Moreover, in some carcinomas VETC has been shown to be predictive of tyrosine-kinase-inhibitors (TKI) response; our results prompt us to verify if this is also true for SFT, where TKI are often used in clinical practice.

ID 885
A SINGLE INSTITUTION EXPERIENCE WITH SS18-SSX AS DIAGNOSTIC IMMUNOMARKER MARKER OF SYNOVIAL SARCOMA. FUSION-SPECIFIC ANTIBODY IN THE WORKUP TO DISTINGUISH SYNOVIAL SARCOMA (SS) FROM HISTOLOGICAL MIMICS

E. Bellan[1], M. Sbaraglia[1,2], A. P. Dei Tos[1,2]

[1] Department of Medicine, University of Padua School of Medicine, Padua, Italy; [2] Department of Pathology, Azienda Ospedale-Università Padova, Padua, Italy

Objectives: SS is an aggressive tumor, which accounts for 5-10% of soft tissue sarcomas and classically presents in the limbs of young adults. Histologically, SS is classified as biphasic, monophasic and poorly differentiated. Over 95% of cases harbor the characteristic t(X;18) (p11;q11) involving gene *SS18* on chromosome 18 and either of two genes on chromosome X: *SSX1* or *SSX2*[1]. Recently, SS18-SSX (E9X9V) breakpoint-specificantibody was shown to be highly sensitive (95%) and specific (100%) for SS[2]. Herein, we present a single-center experience with the newly introduced antibody.
Materials and methods: From 2020 to 2022, we performed SS18-SSX (E9X9V) immunostaining on 250 cases retrieved from the archives of Padua University Hospital and the consultation cases of one of the authors (ADT). Only strong and diffuse nuclear expression was considered positive. All cases of SS were confirmed molecularly (FISH and RT-PCR).
Results: Of the 250 cases included in the study, SS was the rendered diagnosis in 34 cases (14%). All but two casesof SS stained positively for SS18-SSX. One of the two negative cases showed poor tissue fixation. Genetic evidence of *SS18::SSX* gene was detected in

all SS cases. Non-synovial sarcomatous neoplasms (216 cases) that potentially entered in the differential diagnosis were negative. Overall the specificity and sensitivitywere 100% and 94%, respectively.
Conclusions: We concluded that SS18–SSX antibody is highly sensitive and extremely specific for SS diagnosis. Therefore, it represents a valid surrogate for molecular analysis and an extremely helpful tool for all Pathology Departments, even ones not equipped for molecular testing.

References

[1] McBride MJ, Pulice JL, Beird HC, et al. The SS18-SSX Fusion Oncoprotein Hijacks BAF Complex Targeting and Function to Drive Synovial Sarcoma. Cancer Cell. 2018;33(6):1128-1141.e7. https://doi.org/10.1016/j.ccell.2018.05.002.

[2] Baranov E, McBride MJ, Bellizzi AM, et al. A Novel SS18-SSX Fusion-specific Antibody for the Diagnosis of Synovial Sarcoma. Am J Surg Pathol. 2020;44(7):922-933. https://doi.org/10.1097/PAS.0000000000001447.

ID 893
RARE MESENCHYMAL TUMORS OF THE BREAST: CLINICO-PATHOLOGICAL FEATURES OF 11 CASES

T. Buzzanca[1], L. Salvatorelli[1], G. Broggi[1], S. Poidomani[1], M. Mazzucchelli[1],G. Magro[1]

[1] Department of Medical and Surgical Sciences and Advanced Technologies, G.F. Ingrassia, University of Catania, Catania, Italy

Objectives: The aim of this work is to report the clinical and pathological features of aseries of 11 unusual cases of mesenchymal tumors arising primarily in the breast parenchyma. Given the rarity of these lesions in the breast, diagnostic difficulties maybe encountered in rendering the correct diagnosis.
Materials and methods: We retrieved from the files of the pathology archives of Anatomic Pathology Section of University of Catania 11 diagnostically challenging cases of soft tissue tumors occurring the breast parenchyma. The age of patients ranged from 42 to 77 years. For each single case, clinicoradiologic features and paraffin blocks were available for immunohistochemical analyses.
Results: The following cases were studied: i) 1 case of low grade myofibroblastic sarcoma; ii) 1 case of myxoma; iii) 1 case of fibroma; iv) 1 case of intraparenchymal leiomyoma; v) 2 cases of solitary fibrous tumors; vi) 6 cases of desmoid-type fibromatosis. Although the diagnosis was histologically based in all cases, immunohistochemistry was crucial for the diagnosis of solitary fibrous tumors, revealing a diffuse nuclear expression of STAT6 as surrogate of *NAB2–STAT6* gene fusion.
Conclusions: Apart from the more common stromal tumor, i.e. myofibroblastoma, awareness of the possibility that other unusual soft tissue tumors may also arise in the breast parenchyma is crucial for pathologist. The diagnostic approach is similar to that applied in the more common counterpart lesions arising in the soft tissues. In our opinion the diagnosis is mainly based on morphology and supported by suitable immunohistochemical features.

ID 898
SOFT TISSUE TUMORS IN PARENCHYMAL ORGANS: A SERIES OF 10 CASES

F. Gibilisco[1], L. Salvatorelli[1], G. Broggi[1], T. Buzzanca[1], G.M. Vecchio[1], G. Magro[1]

Department of Medical and Surgical Sciences and Advanced Technologies, G.F. Ingrassia, University of Catania, Catania, Italy.

Objectives: The aim of this work is to report the clinico-pathological features of a series of 10 cases of soft tissue tumors arising from unusual sites, namely the uterus, kidney, lung and liver. Given the rarity of these tumors in such locations, the cases herein presented can be viewed as "*diagnostically challenging cases*".
Materials and methods: We have collected a series of 10 cases of soft tissue tumors occurring in parenchymal organs, archived at the Anatomic Pathology Section of the Department of Medical and Surgical Sciences and Advanced Technologies "G. F. Ingrassia" of the University of Catania. Each case was accompanied by clinical and radiological information and paraffin blocks were available for immunohistochemical analyses.
Results: By analyzing the different soft tissue tumors arising primarily from parenchymal organs, we were able to identify the following lesions: i) 3 solitary fibrous tumors of the kidney (one case exhibited nodular area of sarcomatous dedifferentiation); ii) 2 malignant PEComas of the uterus; iii) 2 epithelioid hemangioendotheliomas of the lung; iv) 1 embryonal rhabdomyosarcoma of the uterus; v) 1 myxofibrosarcoma of the kidney; vi) 1 malignant pigmented PEComa of the liver. Their diagnosis was mainly based on the recognition of their morphological features and supported by suitable immunohistochemical profiles.
Conclusions: Pathologists should be aware that soft tissue tumors are not exclusive to soft tissues proper but can also arise from the stromal connective tissue supporting parenchymal organs. In our opinion, it is the unusual site and the unfamiliarity with soft tissue pathology that make the final diagnosis difficult.

CITOLOGIA

ID 748
TISSUE MICROARRAY FROM CELL BLOCK MATERIAL IN THE PREDICTIVE PATHOLOGY ERA: THE PD-L1 EXPERIENCE

G. Acanfora, A. Iaccarino, P. Pisapia, U. Malapelle, C. Bellevicine, G. Troncone and E. Vigliar

Department of Public Health, University of Naples Federico II, 80131 Naples, Italy

Objective: Historically, predictive biomarker tests have been clinically validated on histological formalin-fixed, paraffin-embedded (FFPE) samples; however cytological samples have emerged as a useful tool in predictive testing, also in the immuno-oncology scenario. Despite the promising results reported [1-3], their full implementation in routine clinical practice is still affected by several issues such as protocols standardization, cyto-histological correlation and inter-observer agreement. The aim of this report was to explore the possibility of implementing a large-scale validation of predictive biomarker testing on cytological material, evaluating the technical feasibility of PD-L1 assessment on a cell block (CB) derived tissue microarray (TMA).
Material and methods: Consecutive and unselected CBs prepared from metastatic lymph node fine-needle cytology (FNC) samples were retrospectively collected and used for TMA construction. TMA contained 33 CB-derived cores and 20 sections were hematoxylin and eosin (H&E) stained. PD-L1 immunocitochemistry was carried out using the companion diagnostic kit SP263 assay on 5 sections.
Results: Overall, 29 (88%) samples were visible at least in one H&E-stained slide. Four cases (4/29, 13.8%) showed PD-L1 positivity in neoplastic and/or immune cells.
Conclusions: Although our study was based on a limited and non-selected series, our findings provide a proof of concept for the use of cbTMA in predictive biomarker testing on cytological material in large-scale post-clinical trial validation studies, multicenter studies, and quality control programs.

References

1. John R. Gosney et al, Cytology for PD-L1 testing: A systematic review, Lung Cancer 2020, 41, 101-106, https://doi.org/10.1016/j.lungcan.2020.01.010.
2. Pepe F et al, Tumor mutational burden on cytological samples: A pilot study. Cancer Cytopathol. 2021;129(6):460-467. https://doi.org/10.1002/cncy.22400.
3. Jacobi EM et al, Evaluating Mismatch Repair/Microsatellite Instability Status Using Cytology Effusion Specimens to Determine Eligibility for Immunotherapy. Arch Pathol Lab Med. 2021 1;145(1):46-54. https://doi.org/10.5858/arpa.2019-0398-OA.

ID 835
CYTOLOGICAL DIAGNOSIS OF THYROID INVOLVEMENT IN LANGHERANS CELL HISTIOCYTOSIS: A CASE REPORT

F. Dello Iacovo[1], C. Bellevicine[2], G. Troncone[2], E Vigliar[2]

[1] Department of Advanced Biomedical Sciences, University of Naples "Federico II", Naples, Italy; [2] Department of Public Health, University of Naples "Federico II", Naples, Italy

Objectives: Langerhans cell histiocytosis (LCH) is a rare disease caused by Langerhans cell clonal proliferation. It can involve several organs and tissues, with a predominance of bone (80 % of cases) followed by pituitary gland, which represent the most common endocrinological manifestation[1]; thyroid involvement in LCH is rare, with only 109 cases being reported in literature[1,2]. We present a case of LCH of the thyroid diagnosed on fine needle cytology (FNC) sample.
Materials and methods: A 43-years-old woman with history of LCH presented with thyroid nodule. FNC was performed under ultrasound guidance; direct smears were prepared for Diff-Quick and Papanicolaou staining. Due to the history of the patient an additional pass was performed for cell block (CB) preparation.
Results: Microscopic examination showed mononuclear cells with moderate-to-abundant cytoplasm intermingled with eosinophils. The nuclei were enlarged, and some cells had convoluted nuclei, with cells exhibiting nuclear grooves and "coffee-bean" incisures. Immunocytochemicalstaining performed on CB, showed positivity for S100 and CD1a, confirming the diagnosis of thyroid involvement in LCH.
Conclusions: Thyroid involvement in LCH is rare, with only a small number of documented cases diagnosed on FNC. Microscopic evaluation may be challenging due to the risk of misdiagnosing as benign goiters, anaplastic carcinoma, or lymphoma and due to the rarity of this entity. However, thepossibility should always be taken into consideration in patients with thyroid nodules and history of LCH; therefore an additional pass for CBpreparation is advisable to carry out ancillary studies.

References

1. D. K. Patten et al; Solitary Langerhans Histiocytosis of the Thyroid Gland: A Case Report and Literature Review. Head Neck Pathol. 2012 Jun; 6(2): 279–289. https://doi.org/10.1007/s12105-011-0321-8
2. J. Zhang et al. Spontaneous Thyroid Hemorrhage Caused by Langerhans Cell Histiocytosis: A Case Report and Literature Review. Front Endocrinol (Lausanne). 2021; 12: 610573. https://doi.org/10.3389/fendo.2021.610573

ID 836
FINE-NEEDLE ASPIRATION OF BREAST CLASSIFIED WITH THE IAC YOKOHAMA REPORTINGSYSTEM: FOCUS ON ATYPICAL CASES

M. Salatiello[1], P. Pisapia[1],V. Romeo[1],E. Vigliar[1],A. Brunetti[1],G. Troncone[1],C. Bellevicine[1].

[1] Università degli Studi di Napoli Federico II, Naples, Italy

Objectives: The International Academy of Cytology proposed a new reporting system for breast FNA, the Yokohama System (IACYRS), which aims to standardize the reporting system and the management of the patients with worrisome breast lesions. However, the atypical category (AC), associated with an intermediate risk of malignancy (ROM), still poses diagnostic challenges to cytopathologists. This study aim is twofold: to evaluate, in a prospective series of breast FNAs classified according to the IACYRS, both the ROM and the microscopic features associated with malignancy in AC.
Materials and methods: The database of our Institution was searched for breast FNA specimens performed by interventional cytopathologists between April 2021 and May 2022 and classified according to the IACYRS, and the FNAs diagnosed as atypical were selected; histological follow-up availability was searched as well. Original descriptive reports were reviewed to extract recurrent atypia qualifiers.
Results: Overall, 113 breast FNA were retrieved and classified as inadequate 19 (16,81%), benign 47 (41,6%), atypical 26 (23,1%), suspicious for malignancy 5 (4,42%), and malignant 16 (14,16%). Out of 113 cases collected, 70 (62%) had corresponding histology. Considering the FNA classified as atypical, the histological follow-up was available in 16/26 cases with ROM of 37.5%. The microscopic architectural atypia qualifiers most frequently reported were papillae, cribriformpattern and branching groups whereas the cytology atypia qualifiers were pleomorphism, nucleoli, hyperchromatic nuclei and intracytoplasmic vacuoles; the absence or paucity of myoepithelial cells were mentioned in 9/26 (%) cases. The atypia qualifiers more frequently associated with a subsequent malignant diagnosis were cribriform architectural pattern, nuclear atypia (in particular prominent nucleoli and pleomorphism), and scant myoepithelial cells.
Conclusions: In the present series AC is associated with a moderate ROM (37.5%); we argue that specific atypia qualifiers such as cribriform architecture, nuclear pleomorphism with prominent nucleoli and a scant myoepithelial cell component are potentially helpful for the identification of a subset of higher-risk AC.

ID 894
BRAF MOLECULAR EVALUATION IN MELANOMA METASTASES ON CELL SUSPENSION BY FNAC SAMPLES

E. Carraturo[1], A. Ronchi[1], M. Montella[1], E. Clery[1], F. Zito Marino[1], M. Amato[1], G. Zannini[1], R. Franco[1], I. Cozzolino[1]

[1] Pathology Unit, Department of Mental and Physical Health and Preventive Medicine, University of Campania Luigi Vanvitelli, Naples, Italy

Objectives: Currently, the BRAF molecular assessment on cytological samples of melanoma metastases implies the loss of one or more diagnostic smears, or the execution of one or more passes to obtain dedicated cytological samples[1]. The present study aims to investigate the BRAF molecular status on cell suspensions, to obtain a rapid combined cytological diagnosis of lymph node melanoma metastasis and a molecular assessment of BRAF molecular status. These results can be integrated into the final cytology report.
Materials and methods: All patients were enrolled at the University Hospital "L. Vanvitelli" of Naples from November 2019 to April 2022. FNAC was performed by an interventional cytopathologist. In case of melanoma metastases diagnosed by ROSE, the needle was flushed in a vial containing 350µL of nuclease-free water for molecular evaluation. A second pass was then collected in formalin to set up a cell-block (CB) for immunocytochemical evaluation.
Results: The study included 31 cases of lymph node melanoma metastases diagnosed by FNC. For each case, a cell suspension was prepared. The molecular evaluation on the suspension was performed in all cases. The evaluation was adequate in 29 cases. Overall, BRAF mutations (V600E, V600R and V600K) were detected in 8 out of 29 cases.
Conclusions: Lymph node FNAC is a minimally invasive and inexpensive procedure that is reliably used for the diagnosis of metastatic melanoma. The assessment of BRAF molecular status on cell suspension, obtained by flushing the residual material in the hub of the needle, is an effective technique, allowing to spare biological material.

References

Ronchi A, Montella M, Zito Marino F, Caraglia M, Grimaldi A, Argenziano G, Moscarella E, Brancaccio G, Troiani T, Napolitano S, Franco R, Cozzolino I. Predictive Evaluation on Cytological Sample of Metastatic Melanoma: The Role of BRAF Immunocytochemistry in the Molecular Era. Diagnostics (Basel). 2021 Jun 18;11(6):1110. https://doi.org/10.3390/diagnostics11061110.

ID 918
COMPARISON BETWEEN CONVENTIONAL AND LIQUID-BASED THYROID CYTOLOGY IN THE PANDEMIC ERA: ONE YEAR EXPERIENCE IN A SINGLE INSTITUTION

M. Franchina[1], C. Pizzimenti[2], G.B. Militi[3], A.M. Bonanno[1], A. Ieni[1], M. Martini[1], G. Fadda[1]

[1] Department of Human Pathology in Adult and Developmental Age "Gaetano Barresi", Section of Pathology, University of Messina, Messina, Italy; [2] Department of Biomedical, Dental, Morphological and Functional Imaging Sciences, University of Messina, Messina, Italy; [3] Department of Health Promotion, Mother and Child Care, Internal Medicine and Medical Specialties, Human Pathology Section, University of Palermo, Palermo, Italy

Objectives: Cytology is the pivotal technique for the diagnosis of the nodules of the thyroid gland. We have studied some parameters of efficacy of this technique during two time periods of one year each,using the Liquid Based Cytology (LBC) method (2021-2022) and the conventional method (2019).
Materials and methods: Comparing the exclusive use of the two different methods in the "Gaetano Martino" University Hospital of Messina, we have noticed that a significant decrease of the rate of nondiagnostic (-4,5%) and indeterminate (-2%) diagnoses in the post-pandemic time compared to the first pre-pandemic time. Although the number of fine-needle biopsies in the pre-pandemic time is higher than the ones in the post-pandemic time (498 vs 167), the efficacy of the technique seems to be improved also with the application of immunocytochemical techniques to the material stored in Hologic PreservCyt.
Results: The cyto-histological comparison shows that 100% of the cytological diagnoses are confirmed at histological examination.
Conclusions: The study confirms that LBC is a useful method for the diagnosis of thyroid nodules and its efficacy is increased by the possibility of application of molecular techniques.

References

Vigliar E, Cepurnaite R, Alcaraz-Mateos E, et al. Global impact of the COVID-19 pandemic on cytopathology practice: Results from an international survey of laboratories in 23 countries. Cancer Cytopathol. 2020;128(12):885-894. doi:10.1002/cncy.22373

ID 923
ONCOCYTIC DILEMMA FOR CYTOPHATOLOGIST HEAD AND NECK PATHOLOGY

G. Pini[1], L. Moneghini[2], U. Gianelli[3,4]

[1] Pathology Unit, ASST Santi Paolo e Carlo, Milan, Italy; [2] Pathology Unit, ASST Santi Paolo e Carlo, Milan, Italy; [3] Pathology Unit, ASST Santi Paolo e Carlo, Milan, Italy; [4] Department of Pathophysiology and Trasplantation, University of Milan, Milan, Italy

Introduction. Diffuse hyperplastic oncocytosis (DHO) is an extremely rare benign non-neoplastic condition characterized by a complete replacement of normal salivary gland by oxyphilic cells without alteration in the pre-existing acinar structure. Only ten cases have been previously reported in the literature, representing the most uncommon oncocytic entity. Cytologic evaluation alone is not enough to achieve a definitive diagnosis of DHO, because the findings overlap with those of other oncocytic and oncocytoid lesions.
Objectives. Herein we present cytological findings that should inspire oncocytosis to be included in the differential diagnosis among various oncocytic salivary gland disorders. We also characterize histopathological distinctive features between oncocytic hyperplasias and oncocytoma, its benign neoplastic counterpart.
Materials and methods. A 73-year-old female patient presented with a gradually enlarging right parotid swelling over the preceding year. Fine-needle aspiration citology (FNAC) and parotid gland surgical excision were subsequently performed, revealing diffuse hyperplastic oncocytosis. We describe the findings of cytological and histological evaluations.
Results. On cytology evaluation DHO classically demonstrates a uniform low cellularitypopulation of bland oncocytes, often in cluster, in absence or with few lymphocytes and nobasophilic cells.
Conclusions: **.** The predominance of oncocytes only on citology assessment is a diagnostic challenge due to the lack of unique features, despite some specific clues may be veryhelpful. Hstopathology examination remains the gold standard to discern hyperplastic change from neoplastic conditions.

ID 960
COMPARISON BETWEEN CORE NEEDLE BIOPSY AND FINE NEEDLE ASPIRATION IN SALIVARY GLAND MASSES: A RETROSPECTIVE STUDY

G.B. Militi[1], C. Pizzimenti[2], M. Franchina[3], A.M. Bonanno[3], A. Ieni[3], M. Martini[3], G. Fadda[3]

[1] Department of Health Promotion, Mother and Child Care, Internal Medicine and Medical Specialties, Human Pathology Section, University of Palermo, Palermo, Italy; [2] Department of Biomedical, Dental, Morphological and Functional Imaging Sciences, University of Messina, Messina, Italy; [3] Department of Human Pathology in Adult and Developmental Age "Gaetano Barresi", Section of Pathology, University of Messina, Messina, Italy

Objectives: Among the exams utilized to study salivary gland masses, the core needle biopsy (CNB) and the fine needle aspiration (FNA) have proved to have high specificity and diagnostic accuracy in diagnosing malignancies. The aim of our study is to

clarify the role of these techniques as “first line” in the diagnosis of the nodules of the salivary glands.
Materials and methods: We examined 46 FNA and 20 CNB of salivary gland nodules, for a total of 66 cases, in a period of 5 years (2017-2021), in “Gaetano Martino” University Hospital of Messina. We classified thesecases according to the “The Milan System for Reporting Salivary Gland Cytopathology”(MSRGC), a new reporting system that standardizes and submits the salivary gland lesions in six categories.
Results: We have observed that the neoplastic lesions diagnosed with CNB and FNA were respectively 75% and 52,17%, while the non-neoplastic lesions were 10% and 21,74%, respectively. Furthermore, at the histopathological examination of surgical samples, the CNB diagnosis were confirmed in 80% of cases while the FNA diagnosis were confirmed in 46,15% of the cases, due to the late introduction of MSRSGC and the poor performance in some FNA cases.
Conclusions: According to our study, despite the low concordance of FNA, these two techniques can be used in synergy to improve the diagnostic accuracy of gland salivary lesions in the clinical practice.

References

H.-J. Eom, J.H. Lee, M.-S. Ko, et al. Comparison of Fine-Needle Aspiration and Core Needle Biopsy under Ultrasonographic Guidance for Detecting Malignancy and for the Tissue-Specific Diagnosis of Salivary Gland Tumors. AJNR Am J Neuroradiol 2015 Jun;36(6):1188-93. https://doi.org/10.3174/ajnr.A4247

C. Kala, S. Kala, and L. Khan. Milan System for Reporting Salivary Gland Cytopathology: An Experience with the Implication for Risk of Malignancy. J Cytol. 2019 Jul-Sep; 36(3): 160–164.https://doi.org/10.4103/JOC.JOC_165_18

DERMATOPATOLOGIA

ID 823
SARCOMATOID MELANOMA IS A REALLY TYPE OF MELANOMA?

M. Mureddu[1], G. Cerrone[1], D. Fanni[1], G. Faa[1], L. Pilloni[1]

[1] Division of Pathology, Department of Medical Sciences and Public Health, University of Cagliari, AOU Cagliari, Cagliari, Italy

Background and objective: Spindle cell melanoma is a form of melanoma with pseudosarcomatosus features. Histologically, it ischaracterized by fusiform melanocytes with elongaded nuclei[1]. Desmoplastic melanoma (DM) is a form of spindle cell melanoma. It is classified as pure or combinated. Tumour cells are immunoreactive for S100 and SOX10, but do not express HMB45 and MelanA [2]. DM tends to be diagnosed at a more advanced local stage because it can mimic various benign and malignant nonmelanocytic lesions. However, they tend to have a better prognosis than other subtypes.[3]
Case Report: We report a case of DM with sarcomatoid dedifferentiation in men aged 80 years, that presented with a nodule in the scalp for about 10 years. Histologically, at the dermal-hypodermal junction, a biphasic proliferation was observed. A minor component of the neoplastic nodule was characterized by an amelanotic proliferation of spindle cells in a background of fibrotic stroma and lymphocytic aggregates. The majorcomponent of the proliferation was characterized by a higher cellular density and spindle globular pleomorphic cells, with marked cytonuclear atypia associated with frequent atypical mitotic figures. Neoplastic cells were immunoreactive for SOX10 and S100 and did not show any expression of HMB45 andMelanA.
Conclusions: Spindle cell can be seen in all forms of invasive melanoma. In literature, no pure cases of sarcomatoidmelanomas have been described, whereas all reported cases were mixed with both desmoplastic and sarcomatoid component. According to literature and to the best of our knowledge, pure sarcomatoidmelanomas may not exist, but instead they might represent areas of dedifferentiation of a DM. The development of sarcomatoid area have been reported to be related to a poorer progniosis.[2]

References

1.G. Massi and PE LeBoit Histological Diagnosis of Nevi and Melanoma © Springer-Verlag Berlin Heidelberg 2014, p.457.

2.Kiuru M, McDermott G, Berger M, et al. Desmoplastic melanoma with sarcomatoid dedifferentiation. Am. J. Surg. Pathol. 38, 864–870 (2014).

3.Licata, G. et al. Diagnosis and Management of Melanoma of the Scalp: A Review of the Literature. Clin. Cosmet. Investig. Dermatol. 14, 1435–1447 (2021).

ID 824
NEVOID MELANOMA STRONGLY RESEMBLING A COMMON ACQUIRED NEVUS: A CASE REPORT AND LITERATURE REVIEW

A. Selvaggini, R. Salmaso, A. P. Dei Tos

Anatomic Pathology Unit, Padua University Hospital

Objectives: Nevoid melanoma is an uncommon variant of melanoma characterized by a somewhat nested growth with a variable amount of small to medium sized melanocytes that may lack prominent atypia, especially in the deeper aspects of the lesion. It can arise in a wide age range and it can develop anywhere on the body[1]. Herein we report a case of nevoid melanoma which was particularly tricky to recognize.
Materials and methods: A 44 year old man performed a dermatologic check-up and a nevus with

atypical features was identified and excised by the dermatologist. Macroscopically, the formalin-fixed sample comprised a 0,3 x 0,2 cm, evenly pigmented lesion with sharp border. Sections were paraffin-embedded and H&E stain was performed along with immunohistochemical staining for Sox10/melanA, HMB45, β-catenin, p16, S100 and BRAF.
Results: Low power microscopic examination revealed a 0,7 mm thick, well circumscribed melanocytic proliferation centered in the papillary and superficial reticular dermis with a negligible amount of inflammation, without an apparent junctional component, in the setting of a slightlyatrophic epidermis. A closer look showed mildly atypical melanocytes in the superficial aspects of the lesion with gradual nests and melanocytes size decrease towards the base, which we now refer to as "paradoxical" maturation. There was no evidence of metaplastic reaction. A single, deep mitosis was found in one section. HMB45 and P16 were irregularly expressed, mainly in the superficial half of the lesion. BRAF mutation was confirmed by both IHC and PCR. Our final diagnosis was nevoid melanoma, maturating type.
Conclusions: Nevoid melanoma is matter of concern for the pathologist. The case we reportedfeatured a very small lesion that displayed an exceptional resemblance with a common nevus and could be easily missed by a non-specialized pathologist. Aim of this study is to raise attention on these misleading lesions along with a review of the literature to help distinguish them from common nevi.

References

[1] *Cook MG, Massi D, Blokx WAM et Al. New insights into naevoid melanomas: a clinicopathological reassessment. Histopathology. 2017 Dec;71(6):943-950. https://doi.org/10.1111/his.13317.*

ID 852
ENDOCRINE MUCIN-PRODUCING SWEAT GLAND CARCINOMA (EMPSGC): A CASE REPORT

D. Anconelli[1], N. Santoro[2], G. Pieretti[2], C. Mazzini[2], D. Massi[2]

[1]*Section of Pathology, Department of Health Sciences, University of Florence, Florence, Italy;* [2] *Unit of Ocular Oncology, AOU Careggi, Florence, Italy*

Objectives: Endocrine mucin-producing sweat gland carcinoma (EMPSGC) is a rare low-grade neuroendocrine neoplasm of sweat gland origin; it is considered as the cutaneous counterpart of solid papillary carcinoma of the breast due to its morphophenotipic resemblance, as well as a true precursor of primary adnexal mucinous carcinoma with neuroendocrine differentiation.
Materials and methods: We present a case of a 73-year-old female with a small erythematous nodule on the right upper eyelid of 4 years duration. Past clinical history included a breast cancer diagnosed 10 months before for which the patient was treated with neoadjuvant chemotherapy and hormonotherapy. During hormonotherapy the eyelid nodule had progressively reduced in size. Nodule biopsy was then executed for morphological and immunohistochemical evaluation. Immunohistochemistry for synaptophysin, chromogranin, neuron-specific enolase (NSE), CD56, CK7, GCDFP-15, ER, PR, GATA-3, Ki-67 and HER-2 was performed. Meanwhile, haematoxylin and eosin slides of the previous breast cancer was reviewed.
Results: Histopathological examination showed a well-demarcated expansile tumour with cribriform architecture surrounded by fibrous stroma composed by uniform, polygonal, small to intermediate-sized ductal cells, with round to oval nuclei, finely stippled chromatin andinconspicuous nucleoli. Accurate distinction with a potential breast metastasis was based on different cyto-architectural and immunohistochemical features. Neoplastic cells showed positivity for synaptophysin, chromogranin, NSE, CD56, CK7, GCDFP-15, ER and PR. GATA-3 displayed diffuse and strong nuclear positivity. Ki-67 was positive in less than 5% of cells. HER-2 was negative. These findings were consistent with the diagnosis of endocrine mucin-producing sweat gland carcinoma.
Conclusions: A scant deposition of mucin was observed in absence of infiltrating nests of atypical cells in lakes of extracellular mucin and thus the final classification may be open to discussion. The association of mammary carcinoma and EMPSGC and the clinical framework is intriguing and reinforces the potential relationship of EMPSGC with other hormonally sensitive tumors.

ID 869
c-MYC AND FGFR4 DEREGULATION IN CUTANEOUS ANGIOSARCOMAS

S. Lucà[1], A. Ronchi[1], A.G.M. Cossu[2], R. Caltabiano[3], A. De Chiara[4], G. Palmieri[5], G. Savarese[6], L. Circelli[6], R. Franco[1]

[1]*Pathology Unit, Department of Mental and Physical Health and Preventive Medicine, Universitàdegli Studi della Campania "L. Vanvitelli", Naples, Italy;* [2] *Department of Medical, Surgical, and Experimental Sciences, University of Sassari, Sassari, Italy ;* [3] *Department of Medical Surgical Sciences and Advanced Technologies "G.F. Ingrassia", University of Catania, Catania, Italy;* [4] *Pathology Unit, Department in Support of Oncology Paths, Istituto Nazionale dei Tumoril.R.C.C.S. Fondazione "Pascale, Naples, Italy;* [5] *Institute of Genetic and Biomedical Research (IRGB), National Research Council (CNR), Sassari, Italy;* [6] *AMES, Centro PolidiagnosticoStrumentale srl, Naples, Italy*

Objectives: The molecular profile of cutaneous angiosarcomas (cAS) is largely unknown[1,2]. The aim of this study is to investigate the status of C-MYC and

FGFR4 in primary and secondary cASs.
Materials and methods: The series included twenty-nine cASs. c-Myc and FGFR4 expression were investigated by immunohistochemistry. C-Myc rearrangements and amplifications were investigated by FISH. NGS was performed to investigating 500 genes, including FGFR.
Results: Twenty-one cases (72.4%) were c-myc positive. Evaluation by FISH showed that nine (9/17, 53%) cases showed amplification and/or rearrangement. C-MYC rearrangement significantly correlated with secondary AS ($p < .05$). Ten cases showed a moderate or strong FGFR4 expression. Nine cases showed FGFR4 G388R mutation, which significantly correlated with weak immunohistochemical expression ($p < .05$).
Conclusions: c-MYC rearrangements are more frequent in secondary cASs. FGFR4 mutations are relatively frequent in cASs and correlated with a weaker expression of the protein.

References

1) Requena C, Rubio L, Lavernia J, et al. Immunohistochemical and Fluorescence In Situ Hybridization Analysis of MYC in a Series of 17 Cutaneous Angiosarcomas: A Single-Center Study. Am J Dermatopathol. 2018 May;40(5):349-354. https://doi.org/10.1097/DAD.0000000000001053.
2) Ronchi A, Cozzolino I, Zito Marino F, et al. Primary and secondary cutaneous angiosarcoma: Distinctive clinical, pathological and molecular features. Ann Diagn Pathol. 2020 Oct;48:151597. https://doi.org/10.1016/j.anndiagpath.2020.151597.

ID 906
A RARE CASE OF SUBCUTANEOUS SYNOVIAL SARCOMA

S.Squillaci[1], G.De Biasi[2], M.Chiudinelli[1], P.Balzarini[3], F. De Trovato[1], M.Piccolomini[1],R.Marchione[1]

[1] Unit of Pathology and [2] Surgery, Hospital of Esine (BS), Italy; [3] Dept. of Pathology, University of Brescia, Italy

Objectives: Synovial sarcoma (SS) is a rare soft tissue malignancy affecting mostly young patients which generally occurs in a deep location of the lower extremities. In the current medical literature there are only 14 cases of superficial SS, 11 of the subcutis, one in the dermis, and two involving both the dermis and subcutaneous fat[1].
Materials and methods: A 56 year old man presented in April 2022 with a left buttock mass evolving for a year, clinically resembling a lipoma.
Results: Grossly a 4,2x4x3 cm solid circumscribed whitish/grey mass was seen. Microscopically, it was composed of two different types of neoplastic cells, epithelioid and fusiform in a biphasic arrangement. The first component was made up of columnar to cuboidal cells with round vesicular nuclei and ample eosinophilic cytoplasm arranged in cords, well-formed glands and irregular spaces with eosinophilic granular material within the lumina. The fusiform component consisted of spindle cells arranged in dense cellular sheets, fascicles or herring-bone pattern. The cells were fairly uniform with hyperchromatic nuclei and inconspicuous nucleoli. No necrosis was seen. Mitoses: 25/10 HPF. Resection margins were not free. Immunohistochemistry revealed positivity in the epithelioid component for AE1/AE3 and Cam5.2, CK7, EMA, E-Cadherin, synaptophysin, CD99 (faint), whereas staining for vimentin, CD56, calponin and bcl-2 was seen in the fusiform cells. No expression of SMA, desmin, SOX-10, S-100, CD34 and CEA was observed. FISH break apart showed SS18 gene rearrangement.
Conclusions: The differential diagnosis comprised mainly cutaneous and adnexal carcinosarcoma, including other spindle cell tumors as MPNST, cellular schwannoma and FS-DFSP for monophasic SS[1]. Treatment of choice is wide excision followed by local radiotherapy. Based on previously reported cases, superficial SS appears to act in a relatively indolent fashion with no evidence of metastasis after complete excision.

References

1) Li P, Laskin W, Wang W-L, et al. Primary superficial synovial sarcoma: clinical and histopathological characteristics in eight cases with molecular confirmation. J.Cutan.Pathol.2021;48:263-268 https://10.1111/cup.13887

DIGITAL PATHOLOGY

ID 782
SETTING A DIGITAL PATHOLOGY NETWORK FOR REAL-WORLD VIRTUAL DISCUSSION. SUBDIAGNOSTIC ACCURACY AND TIMING OF DIGITAL PATHOLOGY IN A MULTI-CENTRIC GROUP

A. Del Gobbo[1], L. Bandiera[2], D. Lorenzini[3], A. Di Bernardo[4], A. Pellegrinelli[5], M.Nebuloni[5,8], S. Ferrero[1,6], A. Parafioriti[4], G. Pruneri[3,7], E. Bonoldi[2]

[1] Division of Pathology, Fondazione IRCCS Ca' Granda – Ospedale Maggiore Policlinico, Milan, Italy; [2] Division of Pathology, ASST Grande Ospedale Metropolitano Niguarda, Milan, Italy; [3] Department of Pathology and Laboratory Medicine, Fondazione IRCCS Istituto Nazionale dei Tumori, Milan, Italy; [4] Division of Pathology, ASST Pini-CTO, Milan, Italy; [5] Division of Pathology, ASST Fatebenefratelli-Sacco, Milan, Italy; [6] Department of Biomedical, Surgical and Dental Sciences, University of Milan, Milan, Italy; [7] Department of Oncology and Hemato-Oncology, University of Milan, Milan, Italy; [8] Department of Biomedical and Clinical Sciences, DIBIC, University of Milan, Milan, Italy

Objectives: Validation studies have been carried out to investigate the relevance of whole slide image ex-

amination for primary diagnostic reporting and review, secondary consultation and frozen sections evaluation. Although pathology departments are steadily implementing a digital transformation in different countries, regulatory requirements are still under study. Digital pathology and telepathology has gained momentum upon the COVID19 pandemic, since it ensures remote diagnostic activity and sign-out.
The aim of this study was to evaluate the operational feasibility of a digital pathology system connecting the pathology departments of five big academic hospitals.
Materials and methods: 316 slides from 38 fully anonymized cases, representative of routine inflammatory and neoplastic pathology, were scanned and discussed collectively by experienced pathologists during 10 weekly virtual meetings. For each case, the following features were recorded: type of material, topography, number of slides discussed, level of agreement (concordance, minor and major discordance) among pathologists, quality of the images and requirement of glass slide examination.
Results: Among the 38 cases discussed (13 men and 15 women, mean age 55 years, range 13-83), 20 were represented by biopsies and 18 by surgical samples. Haematopoietic pathology (9 cases) was the most frequent disease, followed by bone and soft tissue (6 cases), skin (5 cases), gastrointestinal and pulmonary pathology. The mean number of virtual slidesanalyzed was 8 (range 1 to 51). In all cases, a diagnostic agreement was achieved: in 6 cases, additional investigations were required to confirm the initial diagnosis (immunohistochemical stainings in three cases; histochemical and immunohistochemical staining in one case; clinical information in two cases). The average discussion time was 15 minutes and the virtual image quality was always rated as excellent (score 5 in a tier from 1 to 5).
Conclusions: Our study confirms that digital pathology is a reliable tool for virtual histopathology discussion, confirming it may significantly reduce turnaround time by avoiding patient transfer for second opinion and expediting second level phenotypical and molecular analysis.
A larger series of cases is needed to strengthen these preliminary considerations. We plan to prospectively validate our data on a larger series of cases.

ID 917
EVALUATION OF KI67 PROLIFERATION INDEX IN PMP: MANUAL COUNT AND DIGITAL IMAGE ANALYSIS COMPARED

C.M. Biatta[1], G. Zalgelli[1], S. Bigolin[2], D. Tormen[1], I.M. Montagner[1], A. Sommariva[1], A. Scapinello[1]

[1] Pathology Unit, Veneto Institute of Oncology IOV-IRCCS, Padua, Italy; [2] University of Padua, Padua, Italy

Objectives: Pseudomyxoma peritonei (PMP) is a clinical syndrome in which a mucinous neoplasm grows within the peritoneal cavity causing mucinous ascites and peritoneal implants. Most cases are of appendiceal origin. The PSOGI expert panel classified PMP in low-grade mucinous carcinoma peritonei (LG), high-grade mucinous carcinoma peritonei (HG) and high-grade mucinous carcinoma peritonei with signet ring cells (SRC).
Materials and methods: We retrospectively reviewed 17 cases of PMP of appendiceal origin, surgically treated at IOV, in the period May 2018 – March 2022. Among them, 7 LG, 5 HG and 5 SRC. For every case, we selected three implants with higher cellularity, on which we performed immunohistochemical staining with Ki67 antibody. In cases where we reported only two percentages, it was not possible to identify three implants with sufficient cellularity (at least 200 cells). For each Ki67 stained specimen, we identified and evaluated multiple areas with higher proliferation index, reporting the most representative percentage, with a count of at least 500 cells. The Ventana DP200 slide scanner has been used at ×20 magnification to digitize the slides. The platforms QuPath (open-source software) was used to assess the percentage of Ki67 positive cells on the stained slides employing the *positive cell detection* algorithm based on Nucleus DAB optical density (OD) mean of DAB positive cells with optimized settings. The analysis was run on all the ROI defined by a pathologist and output as a global Ki67 score (%).
Results: Globally, a significant difference ($p < 0.0001$) between manual and digital Ki67 evaluation was observed, especially in HG, due to more architectural complexity and nuclear stratification. LG values moderately correlate. In 4 of them, QuPath could score Ki67 where we didn't identify sufficient epithelial component (2 SRC consisting of focal neoplastic cell dispersed in mucinous material).
Conclusions: Digital image analysis is a useful tool to increase accuracy in Ki67 scoring in PMP, especially in HG cases. According with morphology, its standardized application may help stratifying PMP as a future perspective.

EMATOPATOLOGIA

ID 772
A RARE CASE OF UNUSUAL VARIANT OF MANTLE CELL LYMPHOMA WITH COINCIDENT FOLLICULAR LYMPHOMA IN SITU

R. Carbone[1], C.Vignale[1], P.Quilici[1], N.A. Pala[1], A. Testa[1], M.Mora[2], R. Brizio[1].

Pathology Unit, Hospital of Imperia (IM), Italy [1]; Pathology Unit, Policlinico San Martino (Ge), Italy [2]

Objectives: Follicular lymphoma in situ (FLIS) is a rare, recently described entity, which has several peculiar features, such as eventual coexistence of a simultaneous disseminated follicular lymphoma, risk of subsequent over follicular lymphoma and association with a second subtype of lymphoma, such as mantle cell lymphoma (MCL). Herein we describe a really rare case of mantel cell lymphoma in conjunction with follicular lymphoma in situ
Material and Methods: A 76-year-old man was admitted at Civil Hospital of Imperia, for splenomegaly and thrombocytopenia. It was also observed a monolateral inguinal lymphadenopathy. The patient underwent excisional biopsy of the lymphnode on suspicion of malignancy.
Results: Gross examination showed a nodular structure of 1,2 cm of maximum size, with white-grayish color cut surface. The lymphnode architecture was predominantly nodular in pattern and most of the nodules were composed of a monotonous population of small to intermediate lymphoid cells with round and irregular nuclei and incospicious nucleoli. This population express B cell-markers such as CD20 and PAX5 and typical mantle cell lymphoma cyclin-D1 and CD5: a diagnosis of mantel cell lymphoma, small cell variant,was done. In the context of this proliferation, moreover, it was identified a single follicle whose germinal center showed complete and strong positivity for BCL2, BCL6 and CD10 with not entirely preserved follicular dendritic network by CD23 antibody, consistent with follicular in situ lymphoma. Subsequently, bone marrow evaluation showed marrow involvement, by interstitial and paratrabecular infiltrates of medium size lymphoid cells with irregular nuclei, expressing CD20, CD79a, and usual mantle cells lymphoma markers CD5 and Cyclin-D1.
Conclusions: Follicular lymphoma in situ is a relatively recent described entity defined as the presence within a lymphnode of a subset of aberrantly BCL2 protein positive follicles with the t(14;18)(q32;q21) IGH/BCL2 traslocation characteristic of follicular lymphoma[1]. FLIS has been found in association with other lymphoma: to our knowledge only two other cases of FLIS were concurrent with a MCL[2,3]. MCL is an aggressive and incurable small B cell lymphoma, part of CCND1 neoplasms group and have a wide variety of morphology which have to be identified in order to guide appropriate therapies and predict clinical outcome: specifically, small cell variant constitutes a specific subset of indolent lymphoma with t (11;14)(q13;q32) characterized by bone marrow involvement, splenomegaly and a low Ki67 proliferation index[4]. The coexistence of FLIS with another B cell lymphomasuggests some hypothesis such as FLIS may be a marker of increased risk to develop lymphoid malignancies due to underlying molecular abnormalities or, simply, a relatively common incidental finding[5]. Our understanding of these entities and their relationship is stil growing and debated.

References

1. Swerdlow SH, Campo E, Harris NL, et al. WHO classification of Hematopoietic and Lymphoid tissue, Revised 4th ed. Lyon, France: IARC Press; 2017.
2. Roullet MR, Martinez D, Ma L, Fowler MH, McPhail ED, Judkins A, Arber DA, Bagg A (2010). Coexisting follicular and mantle cell lymphoma with each having an in situ component:a novel, curious and complex consultation case of coincidental, composite, colonizing lymphoma. Am J Clin Pathol 133:584-591. doi:10.1309/AJCP5RT4MRSDGKSX.
3. Carbone A, Gloghini A (2011). Coexisting follicular and mantle cell lymphoma with each having an in situ component. Am J Clin Pathol 136:481-483. https://doi.org/10.1309/AJCPDSG2J5IBEYFB.
4. Fratoni S. Unusual variants of mantle cell lymphoma. Hematopathology. 2017;2(1):31-49
5. Wu, W., Grimm, K., Hummel, J. et al. Follicular lymphoma in situ with coincident low-grade B cell lymphoma: a case series. J Hematopathol 7, 95–101 (2014). https://doi.org/10.1007/s12308-014-0212-5

ID 826
HISTOLOGICAL SPECTRUM OF INFECTIOUS MONONUCLEOSIS

Rachele Biancotti[1], Marco Pizzi[1,2], Federico Scarmozzino[1], Luisa Santoro[1], Angelo Paolo Dei Tos[1,2]

[1] Department of Pathology, Azienda Ospedale-Università Padova, Italy; [2] Department of Medicine, University of Padua School of Medicine, Italy

Objectives: Infectious mononucleosis (IM) is a major diagnostic pitfall in haematopathology. While the histology of IM has long been described, few studies have addressed the heterogeneity of its morphology and phenotype. To this aim, we present a single center series of clinically annotated IM, focusing on relevant diagnostic clues.
Materials and methods: All cases were retrieved from the Pathology Department of Azienda Ospedale-Università Padova. The following clinical-pathological features were considered: (i) patients' age and demographics; (ii) clinical presentation; (iii) site of diagnostic samples; (iv) growth pattern and cytology of the lymphoid infiltrate; (v) phenotype of the lymphoid cells; (vi) percentage of EBV-positive cells (EBER *in situ* hybridization).
Results: This case series included 4 males with median age at diagnosis of 48.5 years (range: 12-78). The clinical diagnosis was IM in 1/4 (25%) case and suspected lymphoid malignancy in 3/4 (75%) cases. Diagnostic samples included cervical lymph nodes (2/4 [50%]), adenoids (1/4 [25%]) and the spleen (1/4 [25%]). All cases showed at least partial architectural effacement by sheets of medium-to-large atypical blasts with scattered plasma cells, histiocytes and small lymphocytes. Rare Hodgkin/Reed-Sternberg-like cells and foci of necrosis were present in 3/4 (75%) cases. Phenotypic studies showed a predomi-

nance of T cells with partial loss of pan-T cell antigens (CD5 and CD7) in 3/4 (75%) cases. Atypical B cells expressed CD79a and MUM1, with variable loss of CD20 and no light chain restriction. EBER positivity ranged from 7% to 40% of cells and it was mostly documented in B lymphocytes with different degrees of differentiation. A minor (yet consistent) component of EBER-positive T cells was documented in 2/4 (50%) cases.
Conclusions: IM has heterogenous histology and may mimic aggressive B and T cell lymphomas. Clues for the correct diagnosis include: (i) limited architectural effacement, (ii) morphological heterogeneity of EBV-positive cells, (iii) EBV positivity in both B and T cells. However, integration of clinical and histological features together with close follow-up are mandatory in most cases.

ID 862
MYELOID/LYMPHOID NEOPLASMS WITH EOSINOPHILIA: REPORT OF TWO CASES WITH DIFFERENT CLINICAL PRESENTATION

E.Merenda[1], K.Paciaroni[2], E. Scalzulli[3], M. Breccia[3], S.Licci[4], C. Giordano[1], E.Rullo[1]

[1] Department of Radiologic, Oncologic and Pathologic Sciences, Sapienza University of Rome, Italy; [2] Hematology Unit, San Filippo Hospital, Rome Italy; [3] Department of Cellular Biotechnology and Hematology BCE, Sapienza University of Rome, Italy; [4] Pathology Unit, San Filippo Hospital, Rome Italy

Objectives: Myeloid/lymphoid neoplasms with eosinophilia (MLN-eos) and rearrangements of PDGFRA, PDGFRB, FGFR1 and PCM1-JAK2 include rare and heterogeneous clinical-pathological entities with some similarities, not always associated with peripheral eosinophilia. Accurate diagnosis and demonstration of thespecific genetic substrate has important implications since target therapy is available. We report two cases showing similar bone marrow features but different clinical presentation.
Materials and methods: Bone marrow biopsies were stained with Hematoxylin-Eosin, Giemsa and Gomori. Immunohistochemical stains for E-cadherin, Myeloperoxidase, CD15, CD61, CD34, CD117, Tryptase, CD20, CD3, CD30 and CD138 were performed.
Results: Case 1: Male, 57-years old, presenting with mild anemia (Hb 11 g/dl) and splenomegaly. Case 2: Male, 65-years old, presenting with weight loss, night sweats, leukocytosis (20500/mm^3) with eosinophilia (15800 /mm^3) and hepato-splenomegaly. In both cases, bone marrow was hypercellular (90% of hematopoietic component) with a prominent proliferation of immature eosinophilic granuloblasts. The latter were organized in large nodules displacing residual hematopoietic cells (case 1) and were associated withareas of necrosis (case 2). Dyserythropoiesis and dysmorphic features of megakaryocytes were evident. Blast count (CD34+, CD117+) was < 5%. A significant increase of atypical, spindle shaped mast cells, isolated or in small loosely cohesive groups accounting for 15% of bone marrow cellularity, was observed. Recognition of eosinophilic granuloblasts prompted genetic analysis that showed PDGFRB (case1) and PDGFRA (case2) gene rearrangements.
Conclusions: Diagnosis of MLN-eos may be challenging. Pathologists may be the first professionals to suspect the disorder and should be aware of the therapeutic implication. Accurate BOM marrow evaluation with a panel of immunohistochemical reactions, and specific molecular analyses are required for proper diagnosis.

ID 915
EBV+ LARGE CELLS AND HYPERVASCULARITY IN LYMPH NODE OF HIV INFECTED PATIENTS. ALWAYS A MALIGNANT LYMPHOMA?

Paparesta F, Pepi L, Santarelli M, Mazzucchelli R, Filosa A, Goteri G.

Pathology Unit, Marche Polytechnic University, Ospedali Riuniti di Ancona, Ancona, Italy

Objective. EBV reactivation mainly occurs in immunocompromised individuals. EBV-associated lymphoproliferation (LPD) in adults usually arise from reactivation of the virus from EBV-infected memory cells. Such reactivation is generally considered to consist in an escape from the normal immunosurveillance and preferentially occurs at sites representing the physiological reservoir of EBV-infected B cells, such as the lymphatic tissues and mucosa. EBV-related LPDs range from reactive manifestations to malignant lymphomas.
Material and methods: We report a true challenging case of a 52-years old female patient with a recent diagnosis of HIV, who presented persistent diffuse superficial lymphadenopaties since three months, without other constitutional symptoms. Clinicians excised an inguinal lymph node with the hypothesis of a malignant lymphoma.
Results: Pathologic examination showed an overall preservation of the normal nodal architecture, with a prominent harborizing vasculature and a marked expansion of the interfollicular region with a mixture of small lymphocytes, plasma cells, and immunoblasts. Several residual germinal centers were present. The CD4+ T cell were slightly decreased, but T lymphocytes did not exhibit abnormal T cell antigen loss. Immunohistochemcal analysis showed polyclonal plasma cells, and no detectable presence of human herpesvirus–8. Atypical large Hodgkin-like cells were observed in the paracortical area: they resulted CD20+, CD30+, CD15- and EBV positive by in situ hybridization. Paracortical expansion and the complex vasculature raised the suspicion of a T cell

lymphoma. Molecular analysis did not show any B or T clonal population. The overall picture suggests the diagnosis of a polymorphic EBV-LPD.
Conclusions: EBV-related polymorphic LPDs should be taken in mind in HIV positive patients with diffuse lymphadenopaties. Paracortical expansion, EBV positive large cells along with a rich vasculature could represent a reactive aspect and should not be misdiagnosed as a malignant lymphoma such a peripheral T cell lymphoma or an angioimmunoblastic lymphoma. Molecular analysis represent an important ancillary tool in such cases.

References

1. Kim H. Composite lymphoma and related disorders. Am J Clin Pathol 1993;99:445-451. https://doi.org/10.1093/ajcp/99.4.445
2. Taverna J, Nair A, Yun S, et al.. A rare presentation of in situ mantle cell lymphoma and follicular lymphoma: a case report and review of the literature. Case Rep Hematol 2014;2014:145129. https://doi.org/10.1155/2014/145129
3. Hsi ED. 2016 WHO Classification update-What's new in lymphoid neoplasms. Int J Lab Hematol. 2017; Suppl 1:14-22.

IMMUNOISTOCHIMICA

ID 937
COMPOSITE LYMPHOMA COMPRISING FOLLICULAR LYMPHOMA WITH *IN SITU* MANTLE CELL NEOPLASIA: A CASE REPORT

A.Lapenna[1], A. Bianchi[1], O. Annibali[2], V. Tomarchio[2], S. M. Rossi[1], G. Perrone[1]

[1] Unit of Anatomic Pathology, Fondazione Policlinico Universitario Campus Bio-Medico, Rome, Italy; [2] Unit of Hematology, Stem Cell Transplantation, Fondazione Policlinico Universitario Campus Bio-Medico, Rome, Italy

Objectives. Composite lymphoma is an uncommon condition consisting of two or more distinct lymphomas within the same anatomic site[1]. Among them, combination of follicular lymphoma (FL) and *in situ* mantle cell neoplasia (ISMCN) is extremely rare, with only 7 cases reported[2].
Materials and methods. A 55-year-old gentleman presented to the Hematology Unit of our hospital with a single inguinal adenopathy without any other clinical symptoms or signs. An excisional lymph node biopsy was performed.
Results. Pathologic evaluation showed a nodular infiltrate characterized by the proliferation of centrocyte-like and centroblast-like lymphoid cells, which stained positive for CD20, CD10, bcl-2 and bcl-6. Unexpectedly, immunostaining for Cyclin D1 highlighted the presence of small mantle cell-like lymphocytes, and some of them co-expressed SOX11. Based on these findings, the diagnosis of grade 2 B-cell follicular non-Hodgkin lymphoma, with coexisting, in situ mantle cell neoplasia (ISMCN) was made, according to the 2017 WHO criteria[3].
Conclusions: While in some cases the FL and ISMCN components of CL are clonally related, in others they represent the "collision" of two clonally unrelated tumors[2]. Their finding is incidental, with the ISMCN component generally not evolving in an aggressive disease, and the treatment driven by the progression of the sole FL component[2]. This is typically low-grade, bcl2 positive and harbors the t(14;18) translocation, whereas the ISMCN component shows an in situ mantle-zone growth pattern, it is Cyclin D1 positive and harbors the t(11;14) translocation[2].

ID 758
DIFFERENTIAL HLA CLASS I SUBUNIT EXPRESSION LEVELS IN NORMAL TISSUES

F. Ugolini[1], A. Szumera-Ciećkiewicz[2,3], G. Baroni[1], G. Nesi[1], M. Mandalà[4], S. Ferrone[5], D.Massi[1]

[1] Department of Health Sciences, Section of Pathological Anatomy, University of Florence; Florence, Italy; [2] Department of Pathology, Maria Sklodowska-Curie National Research Institute of Oncology, Warsaw, Poland; [3] Diagnostic Hematology Department, Institute of Hematology and Transfusion Medicine, Warsaw, Poland; [4] Unit of Medical Oncology, University of Perugia, Perugia, Italy; [5] Department of Surgery, Massachusetts General Hospital, Harvard Medical School, Boston, Massachusetts

Objectives: Human leukocyte antigen (HLA) class I subunit expression level in primary and metastatic lesions has been characterized in many cancer types utilizing formalin-fixed and paraffin-embedded (FFPE) tissue sections as substrates in immunohistochemical reactions. The evaluation of the results of these studies has been hampered by the scant information about HLA class I subunit expression level in normal tissues. To address this unmet need, we have analyzed HLA class I subunit expression level in FFPE sections of normal tissues.
Materials and methods: Two tissue microarray (TMA) blocks were constructed from archived FFPE tissue samples of a wide number of human normal organs/tissues. The expression level of HLA-A heavy chains, HLA-B, C heavy chains and β_2-microglobulin (β_2-M) was evaluated by IHC staining. The staining was scored according to its intensity.
Results: According to their staining patterns with the three mAbs tested, normal tissues can be divided into four groups. (i) Tissues displaying moderate/strong staining patterns with the three mAbs; (ii) tissues displaying barely detectable staining patterns with the three mAbs; (iii) tissues displaying differential staining patterns with the three mAbs and (iv) tissues with no detectable staining by the three mAbs. Ubiquitous expression pattern for HLA-A, B, C heavy chain and β2-M was found only at the endothelial level, the stroma was negative except for fibroblasts in all the tissues analyzed.

Conclusions: Our data suggest that contrary to the general postulate, HLA class I subunit expression is not detectable in all nucleated cells in FFPE normal tissues. This information provides a useful background to evaluate changes in HLA class I subunit expression associated with malignant transformation of cells.

NEUROPATOLOGIA

ID 797
MUSCLE BIOPSY IN THE DIAGNOSTIC WORK-UP OF NEUROMUSCULAR DISORDERS: A CASE REPORT

F. Nozzoli[1], G. Baroni[1], R. Nassini[2], D. Massi[1], F. Massaro[3], P. Palumbo[3]

[1] Section of Anatomic Pathology, Department of Health Sciences, University of Florence, Florence, Italy; [2] Department of Health Sciences, Clinical Pharmacology Unit, University of Florence, Florence, Italy; [3] Neurology Unit, Santo Stefano Hospital, Prato, Italy

Objectives: Limb–girdle muscular dystrophy (LGMD) refers to a heterogeneous group of non-congenital, genetic muscle disorders with variable age of onset, primarily causing weakness and wasting of the proximal limb and caused by pathogenic genetic variants producing abnormal protein synthesis in various components of the muscle fiber.
Materials and methods: We present a case of a 58-year-old man showing mobility impairment since nearly eight years. None of family members reported known genetic muscle diseases. Serum CK was significantly elevated. Echocardiogram revealed a moderate hypertrophic cardiomyopathy. Muscle MRI identified hypotrophy and fatty replacement of muscle tissue mostly affecting biceps and upper legs muscles. Muscle biopsy was then performed to support the differential diagnosis between several conditions characterized by the possible coexistence of skeletal muscle and cardiac damage such as LGMD1A, 1B, 1C, 1E, 2A,2C, Becker Dystrophy and Pompe Disease.
Results: Haematoxylin and eosin stain shows a marked variation in fiber shape and size with angular fibers, increase in internal nuclei, and adipose replacement of muscle tissue. Gomori Trichrome better demonstrates lobulated fibers separated by excess ofendomysial connective tissue. PAS stain excludes glycogen storage. A customimmunohistochemical panel was performed, including the following antibodies: anti-Dystrophin, anti-Myotilin, anti-Laminin, anti-Caveolin-3, anti-Desmin, anti-Calpain-3 andanti α-, β-, γ-, δ-Sarcoglycan. Calpain-3 was completely absent in the sarcoplasm, while the expression of all other proteins was retained. Muscle biopsy made the diagnosis consistent with a calpainopathy (LGMD2A).
Conclusions: A new tailored neuromuscular immunistochemical panel suitable for FFPE tissue has demonstrated high reliability in resolving a challenging clinical differential diagnosis, identifying the specific muscle disease subtype, thanks to biomarkers targeting proteins implicated in the dystrophic pathogenesis and delivering a non-mandatory role to genetic testing.

ID 952
CASE OF SUBCUTANEOUS MENINGIOMA WITH EQUIVOCAL PATHOGENESIS

T.Ientile[1], G.De Lisi[1], A.G.Giannone[1], A.Bordonali[1], A.M.Florena[1], E.Orlando[1]

[1] Anatomic Pathology, Department of Promotion of Health and Mother and Child Care, Internal Medicine and Medical Specialties, University of Palermo, Italy

Introduction: Meningioma is the most common intracranial tumor. Subcutaneous meningiomas are extremely rare and available data is based only on case reports.
We present a case of subcutaneous meningioma in a patient with history of brain trauma without signs of skull fracture.
Materials and methods: A 40-year-old woman presented with a large (5x4 cm), slowly growing and asymptomaticsubcutaneous lesion of the frontal region, covered by intact skin, firmly fixed both to the skin and to the underlying bone, that progressively occurred 2 years before. Clinical history revealed a domestic accident in the same region, above 3 years before. No CT scan at the time of the trauma is available.
The patient performed skull CT-scan and brain MRI-scan that confirmed the doubtful nature of the subcutaneous lesion associated with focal thickening of the cortical inner section of the bone. The underlying dura mater showed incremented thickness (18x7 mm).
Radiological features suggested the presence of a neoplasm of the dura mater (en-plaque meningioma radiologically supposed) without contiguity of the bone table. An excisional biopsy of the subcutaneous lesion was performed.
Results: Histopathological examination revealed a proliferation of round-to-oval cells arranged in lobules and nests intermingled with slightly pleomorph spindle cells.
The neoplasm showed infiltration of the adjacent tissues. No mitoses were seen.
At immunohistochemistry, tumour cells showed positivity to vimentin, EMA, progesteronereceptor, and Ki67 proliferation index was 2%.
S100, MDM2, CD34, p63 and Cytokeratins were negative, thus excluding a soft tissue neoplasm or an undifferentiated squamous carcinoma or adnexal tumour.
Given morphological and phenotypic characteristics,

the diagnosis was subcutaneous meningioma (WHO grade I).
Conclusions: In our case, it's unlikely that the neoplasm could be caused by a primary location of meningothelial cells in the subcutis, and their localization is probably caused by a post-traumatic implantation.
The patient suffered from an occulted dural meningioma, years before the trauma. During this event, a non-detected skull trauma, probably promoted the implantation of meningothelial cells, and their subsequent growth.

PATOLOGIA APPARATO DIGERENTE

ID 840
ESOPHAGEAL BURIED ADENOCARCINOMA WITH PAGETOID CELLS, A DIAGNOSTIC CLUE

M. Pocci[1], R. Alfieri[2], S. Realdon[3], M. Fassan[4], A. Scapinello[1]

[1] Anatomy and Histology Unit, Istituto Oncologico Veneto, IOV-IRCCS, Castelfranco Veneto, TV, Italy; [2] Surgical Oncology Unit, Istituto Oncologico Veneto IOV IRCCS, Padova, Italy; [3] Gastroenterology Unit, Istituto Oncologico Veneto, IOV-IRCCS, Padua, Italy; [4] Department of Medicine (DIMED), Surgical Pathology & Cytopathology Unit, University of Padua, Padua, Italy

Esophageal adenocarcinoma is the most common esophageal cancer in western countries. It occurs at the distal esophagus in nearly all cases from a Barrett Esophagus. When dysplasia onset, an endoscopic therapy such as radiofrequency ablation, photodynamic therapy or endoscopic mucosal resection should be provided. After these treatments the esophagus heals with the development of a neosquamous epithelium (NSE). One concern in the medical literature is the possibility for the NSE to grow over residual glands that becomes "buried". These buried glands can have intestinal metaplasia but also can be dysplastic or even cancerous. Beneath the NSE they are invisible at endoscopy and may not be biopsied during the follow-up. Furthemore, a superficial biopsy that includes only squamous epithelium can miss the buried glands in the deeper lamina propria. The possibility that a carcinoma is invisible at endoscopy is worrisome. In a systematic review only 34 patients with a subsquamous neoplasia were identified, 31 after PDT and the other after APC or laser ablation. After RFA this possibility is even more rare with only 13 cases reported, 7 with an adenocarcinoma and 5 with a HGD. We observed an additional case of buried adenocaricnoma after a RFA. In this case with a normal appearing endoscopy the possibility of a carcinoma was suspected at histological examination of the biopsy specimen by the observation of atypical cells with a pagetoid fashion in the NSE. Pagetoid cells in the esophageal squamous epithelium were found strongly associated with the presence of an underlying adenocarcinoma and can be a diagnostic clue even in superficial biopsies. Moreover, we observed in the resection specimensadditional histological features that were claimed to be associated with the presence of a carcinoma if found in the biopsies and can have value also when buried neoplasia occous.

ID 859
FEASIBILITY OF DNA EXTRACTION AND MOLECULAR PROFILING IN ENDOSCOPIC BIOPSIES OF PANCREATIC CANCER

V. Mourmouras[1], G. Zalgelli[1], A. Fantin[2], G. Peserico[2], L. Moserle[3], M. Curtarello[3], M.Tessari[4], M.R. Biasin[1], A. Scapinello[1]

[1] Anatomy and Pathological Histology, Veneto Institute of Oncology, IOV-IRCCS, Padua, Italy; [2] Gastroenterology, Veneto Institute of Oncology, IOV-IRCCS, Padua, Italy; [3] Immunology and Molecular Oncology Unit, Veneto Institute of Oncology IOV, IRCCS, Padua, Italy; [4] Gastroenterology Unit, San Bortolo Hospital AULSS8, Vicenza (VI), Italy

Objectives: This study evaluated the correlation between morphometric parameters and quantity and quality of DNA extracted from formalin-fixed paraffin-embedded (FFPE) endoscopic ultrasound fine needle biopsies (EUS-FNB) of pancreatic ductal adenocarcinoma (PDAC) samples, aiming at a complete molecular profiling.
Materials and methods PDAC specimens from 48 patients were analyzed by means of a digital image analysis software (QuPath) in order to categorize epithelial neoplastic tissue, desmoplastic stroma, normal pancreatic parenchyma, necrosis and other parameters. Upon selection of the areas of interest and tumoral DNA extraction, both nucleic acid quantity and quality were assessed.
Moreover, samples were profiled for Microsatellite Instability (MSI) status.
Results The mean area of the samples was 17 mm², mainly represented by neoplastic cells (41,6%), followed by desmoplastic stroma (40,4%). DNA extraction was feasible in all samples with a mean DNA concentration of 14,82 ng/ml and a mean DNA Integrity Number (DIN) of 3,12 ± 1,07. There was a statistically significant linear correlation between amount of DNA extracted and total area and cellularity of the neoplastic component. MSI status was stable in 44 of 48 patients, instable-low in 2 and not feasible in other 2 patients.
Conclusions: EUS-FNB is an optimal method to achieve histological diagnosis, as well as to collect adequate quantity and quality of DNA in PDAC. Digital image analysis software is a useful tool for an objective identification of morphometrical parameters, as well as the quantification of their area and cellularity in PDAC biopsy samples. Area and cellularity

of neoplastic component might predict total amount of DNA extracted from biopsies. Molecular profiling, assessed by MSI status, was feasible in 96% of patients. Therefore EUS-FNB can be recommended to allow precision medicine in pancreatic cancer.

ID 877
UPPER GASTROINTESTINAL INVOLVEMENT IN PAEDIATRIC ULCERATIVE COLITIS: A CASE SERIES

M. C. Sibillo[1], P. Cretella[1], R.A. Franca[1], M. Martinelli[3], E. Miele[3], M. D'Armiento[2], A.Staiano[3]

[1] Section of Anatomic Pathology, Department of Biomorphological and Functional Sciences, Federico II University, Naples, Italy; [2] Section of Anatomic Pathology, Department of Public Health, Federico II University, Naples, Italy; [3] Section of Paediatrics, Department of Translational Medical Science, Federico II University, Naples, Italy

Objectives: Ulcerative Colitis (UC) is typically characterized by a chronic mucosal inflammation, which involves the intestinal tract starting from the rectum and proceeding continuously toward proximal segments. Nevertheless, in children the diagnosis of UC may be more challenging due to the existence of atypical phenotypes such as the upper gastrointestinal (UGI) involvement whichresulted to be the most frequent atypical UC phenotype. Therefore, the ESPGHAN revised Porto criteria recommended that esophagogastroduodenoscopy (EGD) should be performed in all children at the initial evaluation of inflammatory bowel disease (IBD) irrespective of the UGI symptoms and/or signs and the lower endoscopic appearance.
Materials and Methods: This is a retrospective analysis of three cases of severe gastritis in paediatric UC observed between May 2021 and October 2021 at the paediatric IBD centre of the University of Naples "Federico II".
Results: Three cases diagnosed with UC and history of bloody diarrhoea associated with abdominal pain and a moderate disease activity score were observed. Laboratory investigations revealed increased inflammation indices. Lower endoscopy was performed and revealed the presence of severe active pancolitis, consistent with a diagnosis of UC. EGD showed the presence of severe gastritis, confirmed by histologic examination, which revealed chronic active and erosive gastritisand no evidence of Hp infection (Warthin Starry negative staining). During follow-up with induction and maintenance therapy, a progressive improvement was observed, reaching an endoscopic and histological remission of the severe gastritis in 2/3 cases.
Conclusions: The involvement of the UGI tract as atypical phenotype in paediatric UC togetherwith histopathologic findings observed underlines the importance of performing EGD in all children with suspected IBD irrespective of presence or absence of UGI symptoms.

ID 883
PER-PASS PERFORMANCE CHARACTERISTICS OF EUS-FNB IN PANCREATIC MASSES AND NEW PROTOCOLS IN FNB MANAGEMENT

F. Motta[1], G. Bonanno[2], C. Bonanno[1], F. Romeo[2], A. Giorlandino[1], G. Di Mauro[1], M. M. Baron[1], E. Cavallo[1], A. Rizzo[1]

[1] Unit of Anatomic Pathology, Humanitas – ICC Catania; [2] Unit of Endoscopy, Humanitas – ICC Catania

Objectives: Compare results obtained over the last four years in relation to introduction of new aquisition protocols and customized management in EUS-FNB specimen of pancreatic solid masses.
Evaluate the diagnostic accuracy of each sample obtained by EUS-FNB procedure separately, in order to establish, if possible, a minimum sufficient number of passes to perform to have diagnostically representative specimen
Materials and methods: We assessed 220 EUS-FNB procedures from pancreatic lesions, performed with and without macroscopic on-site evaluation (MOSE), between 2018 and 2021. Specific laboratory protocols were made up in order to obtain the most suitable specimens for each diagnostic challenges (i.e. neuroendocrine neoplasms, pancreatic ductal adenocarcinoma in advanced stage, metastasis). Each pass performed in the last two years were individually valuated by two pathologists indipendently in order to obtain separate performance characteristics.
Results: The average reporting time about last four years EUS-FNB procedures has been reduced from 8,5 days, in 2018, to 1,8, in 2021; also diagnosis of malignancy was improved from 49%, in 2018, to 83%, in 2021. Regarding per-pass performance characteristics of EUS-FNB performed in the last two years, initial pass has diagnostic accuracy of 90%, while second pass 94%.
Conclusions: According to our results a single pass would be sufficient for the diagnosis, nevertheless the maximal diagnostic yield of EUS-FNB is reached after 2 needle passes. That allows to minimize the number of passes obtained and may also potentially reduce procedure time and related adverse events. Using Macroscopic On Site Evaluation (MOSE) during EUS-FNB tissue acquisition allow to provide real-time information in order to assesses tissue's adequacy, furthermore introducing specific laboratory protocols we have increased diagnostic accuracy and reduced the number of repeated procedures.

ID 884
GASTRIC CALCIFYING FIBROUS TUMOR, A CASE REPORT

G. Attanasio[1], C. Colarossi[1], G. Magro[1], G. Broggi[1], G. Tinnirello[1], F. Ginilisco[1], L. Memeo[1].

[1] Department of Medical and Surgical Sciences and Advanced Technologies, G.F. Ingrassia, University of Catania, Catania, Italy; [2] Pathology Unit, Department of Experimental Oncology, Mediterranean Institute of Oncology, Catania, Italy

Objectives: Calcifying fibrous tumor (CFT) is a rare pseudotumoral/fibro-inflammatory lesion that occurs most commonly in the gastrointestinal tract, especially in the stomach and the small and large bowel. It tipically affects young adult females, but can occur virtually at any age. The pathogenesis is unknown, but it has been hypothesized that CFT can be associated with IgG4-related disease, Castlemann disease, Sclerosing angiomatoid nodular transformation of the spleen (SANT), trauma and other fibro-inflammatory lesions. The differential diagnosis of gastric CFTs includes Inflammatory myofibroblastic tumor, sclerosing variant of GIST, leiomyoma, schwannoma, plexiform fibromyxoma, solitary fibrous tumor, desmoid fibromatosis, sclerosing mesenteritis and reactive nodular fibrous pseudotumor.
Materials and methods: A 30-years-old female presented with recurrent low-grade fever for two years, and was admitted in our hospital for the evaluation of an asymptomatic gastric lesion detected incidentally. Endoscopy revealed an exophytic subserosal mass in the greater curvature. The gross examination showed a 2.9 x 2.2 cm sized well-circumscribed, unencapsulated and lobulated mass. Sectioning revealed a firm homogeneous, gritty, white cut surface.
Results: Histologically, the mass was characterized by a paucicellular bland-looking spindle cell proliferation, embedded within a dense hyalinized stroma, psammoma bodies, and lymphoplasmocytic infiltrate with some scattered activated lymphoid follicles. Immunohistochemical analysis shows positivity for CD34 and negative staining for DOG-1, ALK, SMA, CD45.
Conclusions: These histological findings suggest the diagnosis of gastric CFT, which should been included in the differential diagnosis between all mesenchymal neoplastic and pseudotumoral lesions of the GI tract, especially in the stomach of young adult females.

ID 888
PRIMARY HEPATIC SARCOMATOID CARCINOMA: A CHALLENGING DIAGNOSIS

G. Pignata[1], R.A. Franca[1], P. Cretella[1], C. Sibillo[1], M. Salatiello[1], R. Montalti[2], R. Troisi[2], M. D'Armiento[3]

[1] Section of Anatomic Pathology, Department of Biomorphological and Functional Sciences, Federico II University, Naples, Italy; [2] Section of hepato-bilio-pancreatic, mini-invasive, and robotic surgery, Department of Public Health, Federico II University, Naples, Italy; [3] Section of Anatomic Pathology, Department of Public Health, Federico II University, Naples, Italy

Objectives: Primary hepatic sarcomatoid carcinoma (PHSC) is a rare but aggressive malignant tumour (~0.2 % of primary malignant hepatic tumours) containing a mixture of hepatic carcinomatous epithelial and sarcomatous elements. Both the terms sarcomatoid carcinoma (SC) or carcinosarcoma (CS) are used to define these tumours. However, a diagnosis of SC is preferred when sarcomatous component is predominantly spindled-shaped, but the epithelial cells are still identifiable. The term CS should be used in tumours with heterologous sarcomatous tissues.
Materials and methods: Clinicopathological feature of a patient diagnosed with PHSC have been revised and described at our Department.
Results: A 54 years-old man with recurrent pain at the right side underwent CT and NMR exams which revealed the presence of a liver mass, centrally colliquated. Solid lung nodules referable to lung metastases were detected. After an US-guided liver biopsy revealing a mesenchymal neoplasm hepatic resection was performed. Gross examination revealed a gray-whitish mass of cm 17,2 maximum diameter, with irregular margins and solid areas. On H&E staining there was a neoplasm composed of pleomorphic cells both of spindled-shaped and epithelioid appearance in addition withmultinucleated neoplastic cells and tumour necrosis. On IHC tumour cells showed positivity for CKAE1/AE3, CK8/18, vimentin, CD68 and WT1 (focally). They were negative for EMA, CK19, CK5/6, CK7, calretinin, SMA, MSA, caldesmon, ALK1, calponin, desmin, c-kit, β-catenin, S100, HepPar1, GPC3, FVIII, ERG, CD34, podoplanin, Bcl-2, CD23. These findings were consistent with a primitive epithelial poorly differentiated malignant neoplasm having a spindle cell component associated. A diagnosis of PHSC was made.
Conclusions: To make a PHSC diagnosis, extensive sampling, histologic and IHC studies areneeded.

ID 903
THE INTRATUMOR HETEROGENEITY OF HER2 AMPLIFICATION IN MIXED-TYPE GASTRIC CANCER

M. Amato[1], R. Sabetta[1], R. Ghirelli[1], I. Panarese[1], M. Accardo[1], F. Ferraraccio[1], F. Zito Marino[1], R. Franco[1]

[1] Pathology Unit, Department of Mental and Physical Health and Preventive Medicine, University of Campania "L. Vanvitelli", Naples, Italy

Objectives: The clinicopathological features of gastric cancer (GC) with mixed-type histology (differentiated and undifferentiated) are incompletely under-

stood. The morphological and molecular intratumor heterogeneity in most human cancers is considered a great limitation. HER2 is overexpressed in approximately 10%-20% of GC and it is a predictive biomarker for targeted therapy. Our study aims to determine whether mixed-type GC could be affected of HER2 overexpression/amplification heterogeneity compared to other histotypes.
Materials and methods: A series of 65 GC, including 31 intestinal, 14 diffuse and 20 mixed types, was analyzed through IHC and FISH to evaluate HER2 overexpression/amplification. IHC was evaluated as follows: score 0/1+: loss or low expression; score 2+: moderate staining; score 3+: intense staining. FISH was positive in cases showing HER2 signals/cell > 6 or ratio HER2/Chr17≥ 2.
Results: In the intestinal-type subset, 11 out of 31 cases were negative; 7 positive; 13 were IHC 2+of which only 2 were FISH positive. In the diffuse-type subset, 7 out of 14 cases were negative; 4 positive;3 were IHC 2+ and FISH negative. In the mixed-type subset, 10 out of 20 cases were negative;5 positive; 5 were IHC 2+ of which only 3 were FISH positive. All intestinal and diffuseGCs showed a homogeneous overexpression/amplification of HER2, whereas 5 out of 10 positive mixed GC showed a heterogeneous HER2 status. Particularly, HER2 overexpression/amplification was associated exclusively with the solid-signet pattern.
Conclusions: Our data suggest that HER2 amplification is homogeneously expressed in intestinal and diffuse types, on the contrary, it is heterogeneous in mixed-type. Furthermore, a strict correlation between HER2 amplification and solid-signet pattern was observed. The intratumor heterogeneity of HER2 amplification in mixed types could lead to a possible impact on the correct diagnosis and the therapeutic responses.

ID 953
GALLBLADDER SQUAMOUS CARCINOMA SECONDARY TO MASSIVE MUCOSAL SQUAMOUS METAPLASIA

G.De Lisi[1], A.G.Giannone[1], A.Bordonali[1], E.Formisano[1], T.Ientile[1], A.M.Florena[1], V.Rodolico[1]

[1] Anatomic Pathology, Department of Health Promotion Sciences Maternal and Infantile Care, Internal Medicine and Medical Specialities, University of Palermo, Italy

Squamous metaplasia is a rare non-neoplastic lesion of the gallbladder. The most frequent types of gallbladder epithelial metaplasia are intestinal and gastric type. Squamous metaplasia was observedin < 0.1% of the cholecystectomy specimens and approximately 12% of gallbladder cancer hadsquamous metaplastic cells evident in adjacent mucosa. We describe a case of clinical and radiological preoperative diagnosis of acute cholecystitis resulted, after pathological examination, in a rare case of primary gallbladder squamous cell carcinoma arising from a xantho-granulomatous cholecystitis with massive squamous metaplasia of the gallbladder mucosa.
Material and methods: A 74 year old man presented with pain in right hypochondrium. Computed tomography demonstrated a condition of acute on chronic cholecystitis with abscessualization. At laparotomy, the gallbladder showed adhesion with duodenum, colonic flexure, and renal fascia; a partial resection was performed.
Results: Microscopy showed almost complete replacement of the gallbladder mucosa by a multi-layered squamous epithelium, characterized by hyperparakeratosis and papillomatosis. Only focal areas of mono-stratified columnar epithelium were found, free from atypia, referable to residual biliary epithelium placed in direct contiguity with the squamous epithelium. Under the epithelial liningthere was a fibrous tissue extensively affected by an intense chronic xantho-granulomatous inflammatory process.
In an isolated area, the metaplastic mucosa showed high-grade dysplasia with some foci of infiltrating carcinoma associated with desmoplasia. Immunohistochemistry showed *en-block* positivity for PanCK, p63, p40, p53 and Ki67.
Conclusions: Primary squamous cell carcinoma is an aggressive gallbladder neoplasm rarely reported in literature only by “case reports”. Multiple evidences suggest that the squamous metaplasia of the gallbladder epithelium may present in close link to squamous neoplasia. As in other settings, the chronic and persistent irritation may lead differentiation of gallbladder epithelial cells into squamous cells,probably triggering a metaplasia-dysplasia-carcinoma sequence.

ID 956
HEPATIC METASTATIC PAGET DISEASE: A VERY UNUSUAL CASE OF VULVAL EXTRA-MAMMARY PAGET DISEASE

S. Battista, G. De Maglio, M. Battiston, L. Caggiari

Unit of Anatomic Pathology, Department of Laboratory Medicine,“S.Maria della Misericordia” Hospital, Udine (Italy)

Background: Vulval extramammary Paget disease (vEMPD) is an uncommon epithelial malignancy usually develops in apocrine gland-bearing areas, may arise within the vulva (primary vEMPD), scrotum and penis or represent vulval skin involvement by a noncutaneous carcinoma (secondary vEMPD). We report an interesting and very unusual case of metastatic Paget of the vulva to the liver, lung, lymph nodes and bone.
Matherials and methods: the material received from hepatic needle biopsy was processed, embedded in paraffin and subsequently dissected with the microtome. The sections were staining with hematoxylin/

eosin and immunohistochemical examinations have been set up to characterize the lesion according to the clinical-anamnestic data (CK7, GCDFP-15, androgen receptor AR and GATA-3).
Discussion: a 72-year-old woman with history of vulvar neoplasia in 2008, access to the emergency room for reported progressive worsening of general conditions, associated with abdominal pain for about 10 days, radiating to the lumbar region, with dyspepsia, nausea and weight loss (about 10 kg in the last few months), worsening of known visual deficits. Blood chemistry tests show alteration of hepatic indices of cytonecrosis. CT abdomen highlighed enlarged liver (DL right lobe 17 cm); the hepatic parenchyma is entirely occupied by multiple hypervascularized oval lesions, with a tendency to confluence, the largest in the VIII segment to form a parenchymal mass with lobulated margins of 85 x 50 mm (ApxLL). Morphologically, the liver parenchima is replaced by an atypical epitheliomorph proliferation with solid growth pattern consisting of single vacuolated cellular elements, sometimes forming pseudo-glandular structures. Immunohistochemical examinations revealed that these cellular elements shows positivity for CK7, GCDFP-15, androgen receptor (AR), and GATA-3 morpho-immunophenotipically coherent with vulvar lesion in anamnesis (positive more often in primary than secondary).
Conclusions: we report an interesting and very unusual case of liver metastasis of extra-mammary Paget (Paget's) disease.

PATOLOGIA CARDIOVASCOLARE

ID 817
A TEN-YEAR STRAIN ANALYSIS ON CARDIAC MAGNETIC RESONANCE IN ENDOMYOCARDIAL BIOPSY PROVEN MYOCARDITIS

S. Rizzo, S. Gazzea, A. Cecere, C. Basso, M. Perazzolo Marra

Department of Cardiac,Thoracic and Vascular, University of Padua, Padua, Italy

Objectives: To evaluate the potential role of global strain cardiac magnetic resonance (CMR) in the differentiation of acute, chronic and chronic active myocarditis in patients with endomyocardial biopsy (EMB)-proven myocarditis.
Materials and methods: The study population consisted of 50 patients with an EMB diagnosis of myocarditis and CMR between 2010 and 2021. For each patient, clinical history, symptoms at presentation and tests were searched for. Based on EMB findings, the population was divided according to the stages of the disease in acute, chronic and chronic active myocarditis. On each CMR, global circumferential (GCS), longitudinal (GLS) and radial peak (GRS) strains were calculated in the left ventricle. The data were compared between patients with acute and chronic and with acute and chronic active myocarditis, with ejection fraction (EF)≥ 40% and < 40%.
Results: Twenty-six patients had acute myocarditis, 13 chronic myocarditis and 9 chronic active myocarditis. Among patients with EF≥40%, GLS, GCS and GRS peak values were -13,1±3,71%, -15,3±4,53% and 25,2±10,78% respectively and they were not statistically different between patients with acute and chronic myocarditis and between acute and chronic active myocarditis. GLS, GCS and GRS of patients with EF < 40% measured -6,06±2,33%, -7,21±3,38% and 9,52±4,15%, with no difference among the three groups.
Conclusions: In patients with an EMB diagnosis of myocarditis, feature tracking analysis of GLS, GCS, and GRS does not differentiate stages of the disease, which remains feasible only through EMB.

ID 822
STABLE AND UNSTABLE ATHEROSCLEROTIC PLAQUES. HOW A NEW SCORING SYSTEM COULD SIMPLIFY THE HISTOPATHOLOGICAL DIAGNOSIS

E. Noto[1], G. Cerrone[1], M. Piras[1], F. Cau[1], D. Fanni[1], G. Faa[1], L.Saba[2], C. Gerosa[1]

[1] Division of Pathology, Department of Medical Sciences and Public Health, University of Cagliari, AOU Cagliari, Cagliari, Italy; [2] Department of Radiology, University Hospital of Cagliari, Cagliari, Italy

Objectives: Atherosclerosis is a pathological process characterized by endothelial dysfunction and plaque formation in the intima of the arterial wall. In atherosclerotic plaques we can observed multiple elementary lesions that have heterogeneous features and different prognostic significance. These elementary lesions have heterogeneous features and different prognostic significance. For example, calcifications are a common lesion of the atherosclerotic plaque but the type of calcification is strictly associated with prognosis:calcifications in sheets are typical of the stable plaque, whereas fragmentated or microcalcifications are more typical of the unstable plaque[1].
Materials and methods: One hundred consecutive patients treated with carotid endarterectomy were retrospectively analyzed to assess elementary lesions that characterize stable and unstable plaques.
Results: Unstable plaques are at risk for rupture; they are characterized by a thin fibrous cap, microcalcifications, numerous inflammatory cells, intraplaque hemorrhage, neoangiogenesis, hemosiderosis and, occasionally, organized thrombi. We propose a new simplified Instability Scoring System (ISS), in which an arbitrary numerical value isassigned to every elementary lesion. A final score < 3 should indicate a

stable plaque; a final score > 6 should suggest the diagnosis of unstable plaque.
Conclusions: According with our preliminary data, ISS is a simple histological scoring system,reproducible even among non-experienced pathologists, that would help to unify and standardize the histologic report of the atherosclerotic plaque. Further studies will clarify the correlations between our scoring system and the radiological and clinical data, in order to better stratify patients at higher risk for further complications on the basis of the histological analysis of any carotid plaque.

References

1. L.Saba, N. Agarwal, R. Cau, et al. Review of imaging biomarkers for the vulnerable carotid plaque. JVS Vasc Sci. 2021 May 15;2:149-158 https://doi.org/10.1016/j.jvssci.2021.03.001. PMID: 34617065; PMCID: PMC8489200.

ID 844
HYPERTROPHIC CARDIOMYOPATHY: A PATHOLOGY STUDY OF THE MITRAL VALVE APPARATUS AND THE MITRO-AORTIC FIBROUS CONTINUITY

M. De Gaspari, M. Mazzuccato, M. Bueno Marinas, S. Rizzo, C. Basso

Department of Cardiac, Thoracic, Vascular Sciences and Public Health, University Hospital, Padova, Italy

Objectives: Hypertrophic cardiomyopathy (HCM) is a inherited myocardial disease mostly due to sarcomericgene mutations at risk of sudden cardiac death (SCD) and heart failure even requiring heart transplantation (HT). Recently, a "muscular discontinuity" of the mitro-aortic intervalvular fibrosa has been reported during surgery in obstructive HCM.[1] The aim of our study is to validate these findings through a detailed pathological analysis of full heart specimens.
Material and methods: Inclusion criteria were septal asymmetric HCM from SCD, HT or other causes of death. Heartswithout a diagnosis of HCM served as controls. Both gross and histological analysis of the mitral valve (MV) apparatus and the mitro-aortic continuity was performed.
Results: A total of 30 HCM hearts (10 SCD, 10 HT, and 10 with other causes of death; median age 29.5 years; 15 males) and 10 controls (median age 54; 7 males) were studied. In HCM hearts, a thickening of the anterior MV leaflet was present in 56.7% and an anomalous insertion of papillary muscle in 10%. The septal bulging was evident in 80%, and the endocardial plaque in 63.3%. All cases but one (96.7%) revealed a posterior muscular layer overlapping the mitro-aortic fibrous continuity, corresponding to the left atrial myocardium. A negative correlation between the length of the left atrial myocardium layer and both the patients' age and the anterior MV leaflet length was found. Comparing the length of the "discontinuity" in HCM vs controls, no significant difference between the two groups is evident after age adjustment.
Conclusions: Pathology study of septal asymmetric HCM demonstrates that the mitro-aortic "muscular discontinuity" is due to the overlap of left atrial myocardium along the fibrous continuity. The length of this layer is decreasing with age. Anterior MV leaflet thickening, septal bulging, and endocardial plaque are frequent findings, while papillary muscle abnormalities are rare.

References

1. Ferrazzi P, Spirito P, Binaco I, et al. Congenital Muscular Mitral-Aortic Discontinuity Identified in Patients With Obstructive Hypertrophic Cardiomyopathy. J Am Coll Cardiol. 2020;76(19):2238-2247

ID 897
INFERIOR VENA CAVA AND INTRACARDIAC INVASION OF HEPATOCELLULAR CARCINOMA: A CASE REPORT

V. Nistri[1], P. Stefano[2], G. Batignani[2], F. Marra[2], G. Nesi[1]

[1] Section of Pathology, Department of Health Sciences, University of Florence, Florence, Italy; [2] Department of Experimental and Clinical Medicine, University of Florence, Florence, Italy

Objectives: Cardiac metastases are reported in 12-25% of patients with widespread malignancy. By contrast, solitary metastases to the heart are highly unusual. Tumor invasion can occur through multiple mechanisms, including direct contiguous extension, hematogenous spread, lymphatic spread, or transvenous extension.
Materials and methods: We present a case of a 65-year-old Caucasian man who had an orthotopic liver transplant for HCV-related cirrhosis and hepatocellular carcinoma (HCC) in 2018. In 2022, a follow-up whole body CT scan demonstrated thrombosis of the inferior vena cava as far as the right atrium. Echocardiography showed the right atrium almost entirely occupied by a roundish mass of 4 x 5 cm in size. MRI and CT scans of the heart confirmed the presence of a polylobate mass with inhomogeneous contrast enhancement at the level of the right atrium, inferior cavoatrial junction and intrahepatic tract of the inferior vena cava. The lesion was surgically removed.
Results: Microscopy revealed an encapsulated tumour with trabecular-sinusoidal pattern and polygonal cells with abundant clear cytoplasm and central round nucleus. At immunohistochemistry, neoplasticcells were positive for Hepatocyte Specific Antigen (HSA), Glypican-3 and CAM 5.2, and negative for CK20, CK7 and PAX-8. Final diagnosis was intracavitary metastasis of HCC to the right atrium.

Conclusions: Although HCC has a tendency to spread to the venous system, intracardiac involvement is extremely rare and has a dismal prognosis. HCC with inferior vena cava and right atrium thrombus accounts for approximately 1.4-4.9% of cases. Patients may be asymptomatic, as in our case, or present with non-specific symptoms. Early diagnosis is therefore crucial and improved HCC follow-up is mandatory for better patient life expectancy.

ID 909
CARDIAC DESMOPLASTIC SMALL ROUND CELL TUMOR (DSRCT) IN ADULT WOMAN: A UNIQUE CASE

N. D'Alessandris[1], F. Addante[2], G. Scaglione[1], S. Sfregola[1], F. Vegni[1], F. Policardo[1], R. Benvenuto[1], A. Carlino[1], A. Santoro[1], A.M. Mulé[1].

[1] Unit of General Pathological Anatomy, Department for Women's Health Science, Children and Public Health. Fondazione Policlinico Universitario "Agostino Gemelli" IRCCS, Rome, Italy; [2] Department of Emergency and Organ Transplantation, Section of Anatomic Pathology, School of Medicine, University "Aldo Moro", Bari, Italy

Objectives: Correct use of immunohistochemistry in distinguishing suspicious lesions in patients with personal history of previous malignancy, especially in cases with unusual presentation.
Materials and methods: We analysed the case of a 64-year-old woman diagnosed with breast cancer in 2011. During the standard follow-up program at our Department, in 2021, imaging incidentally revealed a mass in the right atrium. Patient had no symptoms at that time.
Successively, she underwent further instrumental evaluations which confirmed the presence of a 53x34 mm retrocardiac mass in the right atrium. The final diagnosis was assessed by median sternotomy and concurrent biopsy.
Results: Microscopically, we observed a malignant neoplasia composed by solid nests and cords of uniform small "blue" cells, with scant cytoplasm and prominent cell borders. These small blue cells were separated by desmoplastic stroma and necrosis. Rosette-like structures were occasionally observed. The mitotic index was 15 mitoses per 10 high power fields (HPFs). Immunoistochemistry revealed diffuse positivity for AE1/AE3 (dot-like), desmin (dot-like), focal positivity for CK20 and 17, vimentin, CD99 and GATA3 and negativity forp63, chromogranin, synaptophysin, NCAM, TTF1, CD45, CD138, S100, MyoD1, FLI1 and TLE1. After excluding a possible breast metastasis, we focused on the differential diagnosis of the so called "small blue cell tumors" with a final diagnosis of DSRCT further confirmed by molecular studies showing the presence of the EWSR1-WT1 fusion gene.
Conclusions: DSRCT diagnosis can be challenging especially in cases of unusual presentation and in cases of patient positive previous history of malignancy. The application of an optimal immunohistochemical panel together with molecular confirmation are useful to avoid misdiagnoses and to proceed with the correct therapeutic approach.

PATOLOGIA ENDOCRINA

ID 792
L-CELL NET WITHIN A TAILGUT CYST

A. Feraco[1], G. Fornuto[2], C. Cascone[3], C. Rabitti[3], A. Onetti Muda[2], G. Rindi[1]

[1] Section of Anatomic Pathology, Department of Life Sciences and Public Health, Università Cattolica del Sacro Cuore, Roma, Italy; [2] Pathology Unit, [3] Department of Surgery, University Hospital Campus Bio-Medico of Rome, Roma, Italy

Objectives: To investigate the neuroendocrine neoplasia observed in a tailgut cyst.
Materials and methods: A 64-year-old lady with a presacral mass at MRI suspected for tailgut cyst underwent curative surgery. The study comprised: H&E for histology andimmunohistochemistry for chromogranin A, synaptophysin, INSM1, CDX2, TTF1, GATA3, Glucagon (glicentin cross-reacting), PP(PYY cross-reacting), serotonin, SSTR2, Ki67(MIB1).
Results: The lesion was cm10 in size, cystic and multilocular. The histology showed multiple fluid-containing cysts lined by squamous or columnar, ciliated, glandular epithelium with intermingling fibrous-adipose tissue and focal smooth muscle. No atypia was observed. However, a 0,5cm epithelial neoplasia was found in the glandular lined cystic component. The tumor had a well differentiated neuroendocrine neoplasia morphology (1). Tumor cells stained diffusely for cytokeratin AE1/AE3, synaptophysin, Glucagon, PP, INSM-1, SSTR2 and focally for chromogranin A and CDX2; no stain was observed for GATA3, TTF-1 and 5HT. The Ki67 proliferation index was 8%. GATA3 was diffusely positive in the epithelial lining of the glandular lined cystic only. A diagnosis of well differentiated neuroendocrine neoplasia, neuroendocrine tumor, NET(G2) composed by L-cell of a tailgut cyst was rendered.
Conclusions: Tailgut cysts are presacral teratoma. About 30 cases have been described. To our best-knowledge, this is the first and only case of L-cell Type NET described in a presacral teratoma. L-cell NETs are usually observed in the lower gut(colon-rectum) and, exceedingly rare, in the middle ear(1). The observed expression of GATA3 in the overlying epithelial component may suggest a mesonefric origin. Genetics for a Currarino syndrome (sacral anomaly, anorectal malformation and presacral mass) variant is currently investigated.

References

1. WHO Classification of Tumours Editorial Board. Endocrine and Neuroendocrine tumours. Lyon (France):International Agency for Research on Cancer; 2022.(WHO classification of tumours series, 5th ed; vol. 8). Available from: https://tumourclassification.iarc.who.int/chapters/36.

ID 873
ATYPICAL ADENOMA OF THE PARATHYROID: A CASE REPORT

E. Clery[1], M. Accardo[1], L. Scappaticcio[2], K. Esposito[2], R. Franco[1], A. Ronchi[1].

[1] Pathology Unit, Department of Mental and Physical Health and Preventive Medicine University of Campania "L. Vanvitelli", Naples; [2] Department of Medical and Advanced Surgical Sciences, University of Campania "L. Vanvitelli", Naples

Objectives: We describe an unusual case of atypical parathyroid adenoma.
Materials and methods: A 58 -year -old man with serious hypercalcemia due to primary hyperparathyroidism. The patient had complained of pain and oedema located on the right hip, where a prosthesis was implanted in June 2021. Neck ultrasound showed an apparently extra-thyroidal, solid, hypoechoic nodule of 25 mm, in continuity with the lower third of the right lobe. In this area the planar parathyroid scintigraphy (^{99m}Tc-MIBI) waspositive.
Results: Microscopic evaluation showed a neoplasm bordered by a thin fibrotic capsule with organoid and trabecular growth pattern. Cell population showed round nuclei and large cytoplasm, without significant atypia. Necrosis was not observed. There were neither clear images of endovascular neoplastic embolization nor extralesional invasion. Banding fibrosis was present, buried the neoplasm. Mitotic index was < 1 mitosis/10 HPF. Ki-67 appears low (about 3%). Diagnosis of atypical parathyroid adenoma was rendered[1].
Three days after surgery, a marked reduction of serum parathormone (PTH, 19.2 pg/mL) and calcium (ACC,8.6 mg/dL) were observed. At one month after the bone surgery, the fracture fixation was stable and biochemical parameters were good (creatinine 2.3 mg/dL, PTH 56.5 pg/mL, ACC 9.1 mg/dL, potassium 4.2 mEq/L).
Conslusions: A careful analysis of the capsule of parathyroid nodules is fundamental for a correct differential diagnosis between adenoma, atypical adenoma, and carcinoma. This differential diagnosis has important therapeutic and prognostic repercussions. Follow-up is necessary due to the uncertain biological behaviour of these proliferations.

References

1) Erickson LA, Mete O, Juhlin CC, Perren A, Gill AJ. Overview of the 2022 WHO Classification of Parathyroid Tumors. Endocr Pathol. 2022 Mar;33(1):64-89.

ID 890
A RARE CASE OF ECTOPIC PARATHYROID ADENOMA ASSOCIATED WITH ECTOPIC THYMIC TISSUE

M. Cieri[1], C. De Carlo[2], M. Valeri[2], M. Iuzzolino[2]

[1] Pathology Unit, Humanitas Research Hospital, Rozzano, Italy; [2] Humanitas University, Rozzano, Italy

Objectives: Ectopic parathyroid adenoma is an uncommon cause of primary hyperparathyroidism. Ectopic thymic tissue as well, is a rare condition frequently seen in pediatric patients but extremely rarely in adults, being found incidentally on cervical imaging. Here we report a rare case of a 2,5 cm large pericarotideal parathyroid adenoma associated with ectopic thymus.
Material and methods: A 64 years old woman with primary hyperparathyroidism was referred with a 11C-methionine PET reporting a left laterocervical lesion. Focal abnormal uptake in the laterocervical region along the left lateral aspect of the carotid was identified. During the surgery a parathyroid adenoma measuring 2.5 cm was confirmed and enucleated. The patient recovered uneventfully, with normalization of serum calcium, parathyroid hormone (PTH), and normal vocal cord function.
Results: macroscopically the lesion appeared as a solid, yellow and well circumscribed nodule surrounded by fatty tissue. The histopathological examination showed a capsulated proliferation of oncocytic cells admixed with a population of clear cells, adjacent to thymic tissue, in absence of mitosis or necrosis.
Conclusions: Parathyroid adenoma is the most common cause of primary hyperparathyroidism. Its prevalence is about 2-43% in anatomical series and up to 16% and 14% in patients with primary and secondary hyperparathyroidism, respectively. Ectopic parathyroid glands are detected occasionally, especially in cases of recurrent hyperparathyroidism after initial parathyroidectomy. Their ectopic locations usually result from faulty migration during embryogenesis. Indeed, due to the longer embryologic migration tract, inferior parathyroid glands are more likely to be found in ectopic locations or incompletely descended. Ectopic thymic tissue, as well, is a rare anomaly which could be associated with parathyroid glands due to their embryologic relationships. The combination of the two anomalies is seldom reported and it poses a diagnostic challenge and lack of successful identification may lead to lack of success in parathyroid surgery.

PATOLOGIA FETOPLACENTARE

ID 796
SUB-CENTIMETRIC CHORIOANGIOMAS: ASSOCIATION WITH PREGNANCY COMPLICATIONS

E. Nardi[1], V. Seravalli[2], N. Strambi[2], M. Di Tommaso[2], F. Castiglione[1]

[1] Unit of Anatomic Pathology, University of Florence, Italy; [2] Department of Health Science, Division of Obstetrics & Gynecology, University of Florence, Italy

Objectives: Chorioangiomas are benign angiomas arising from chorionic tissue and they are the most common non-trophoblastic tumors of the placenta, occurring in 1% of all examined placentas.
Most chorioangiomas are small, asymptomatic, and generally not considered of clinical significance. However, those measuring more than 4-5 cm in diameter may be associated with maternal or fetal complications including preeclampsia, maternal mirror syndrome, preterm delivery, nonimmune fetal hydrops, fetal growth restriction and fetal demise. Such large placental chorioangiomas can usually be detected prenatally by grayscale or Color Doppler sonography. The aim of this study was to correlate the presence of small chorioangiomas, often missed on prenatal ultrasound, with complications of pregnancy.
Materials and methods: 26 samples of formalin-fixed and paraffin-embedded placental tissue were retrospectively selected from those with a histopathological diagnosis of chorioangioma, after excluding multiple pregnancies, cases of major fetal malformations, and large placental chorioangiomas detected on prenatal ultrasound. All the chorioangiomas examined measured less than 1 cm. The incidence of pregnancy complications was retrospectively evaluated.
Results: Among the cases examined, we observed a 35% incidence of hypertensive disorders of pregnancy,a 31% incidence of fetal growth restriction, and a 10% incidence of preterm premature rupture of membranes (pPROM). Incidence of these complications was much higher than in the general pregnant population.
Conclusion: The present study showed that maternal hypertensive disorders, fetal growth restriction and pPROM were frequently associated with the presence of millimetric chorioangiomas not detected prenatally.

ID 907
INTRAVASCULAR MICROORGANISMS IN ACUTE VILLITIS. A CASE REPORT AND LITERATURE REVIEW

P. Srsen[1], R. Biancotti[1], K. Ludwig[1], A. Guerriero[1], L. Alessandrini[1], A.P. Dei Tos[1]

[1] Department of Medicine-DIMED, Section of Pathology, University of Padova, Padova, Italy

Introduction: Acute villitis, defined as presence of neutrophils in fetal villous capillaries and stroma, with intravascular microorganisms in fetal vessels, is a rare histological entity and only a limited number of cases has been reported in the literature.
Materials and methods: A 31-year-old women (PARA 1011) with no preexisting comorbidities presented at 25 weeks of gestation to the gynecology emergency department with malaise, chills, abdominal pain and fever. Laboratory test revealed an elevated white blood cell count with neutrophilia. An ultrasound scan revealed the absence of fetal movements and heartbeat and after prostaglandin induced abortion the placenta was sent without delay for histopathological examination.
Discussion and Results: The histological examination of the placenta revealed acute chorionitis/subchorionitis associated with vasculitis of chorionic plate vessels and diffuse acute villitis with numerous Gram-positive cocci within the fetal vessels. The microorganisms were shown to be *Streptococcus agalactiae*. The mother responded to antibiotics and was discharged after two days without symptoms.
Subsequently a systematic literature search in different databases of reviews, case series and case reports of placentas with acute villitis and intravascular microorganisms was carried out. Clinical data, presentation, and histomorphologicalplacental characteristics were obtained from eligible articles for which full texts were written in English. From the initially identified 25 articles, only 6, containing a total of 18 cases were included.
The literature testifies to an extremely varied spectrum of maternal symptoms, ranging from a fast resolution to severe conditions such as sepsis, the need for hysterectomy and in a single case the patient's death. Without exception cases of acute villitis with intravascular microorganisms are associated with fetal death.
Conclusions: Due to the extreme variability of symptoms and complications brought about by acute villitis with intravascular microorganisms, a detailed histological analysis of the placenta is warranted in order to better understand the causes and mechanisms underlying acute and severe intrauterine infection.

ID 955
THIRD TRIMESTER PLACENTAL ALTERATIONS OF SARS-COV-2 POSITIVE PREGNANT WOMEN

A.G. Giannone[1], G. De Lisi[1], E. Orlando[1], A.M. Florena[1], G. Morello[1], E. Maresi[1]

[1] Department of Promotion of Health, Mother and Child Care, Internal Medicine and Medical Specialties, University of Palermo, Italy

Objectives: Pregnancy is not a risk factor for maternal infection and SARS-CoV-2 does not seem to have an increased risk to trans-placentally infect the foetus. However, this doesn't completely exclude

vertical transmission that occurs in 3–6% of pregnancies, more frequently in the third trimester and rarely earlier.
The study reports the histological, immunohistochemical and molecular analyses of a series of placentas from SARS-CoV-2-positive mothers to highlight any alteration attributable to SARS-CoV-2.
Materials and methods: Twenty-one third trimester placentas from SARS-CoV-2-positive pregnant women were studied, with no clinical or serological evidence of SARS-CoV-2 vertical transmission. Twenty-one third-trimester placentas from SARS-CoV-2-negative women were selected as control group.
Results: In SARS-CoV-2 positive pregnant women were observed non-specific lesions: Maternal VascularMalperfusion (MVM) was present in 19 cases (90.4%); Fetal Vascular Malperfusion (FVM) occurred in 20 cases (95.2%); Maternal/Fetal Inflammatory Response (MFIR) was observed in 3 cases (14.2%). In 17 cases (80%) MVP and FVM were associated; in 2 cases (10%) FVM and MFIR were associated; in 1 case (5%) MVM, FVM and MFIR occurred together; in 1 case (5%) MVM occurred alone. Comparison of placental lesions between SARS-COV-2 positive pregnant women with the control group showed a statistically significant difference (*p value: 0,0089)*. In all the cases, biomolecular and immunohistochemical analyses (RNASCOPE with SARS CoV-2 probe; anti-spike protein) were negative for viral mRNA and/or spike protein. Preterm delivery was significantly more frequent (*p value: 0,0048)* in virus-positive women.
Conclusions: Placental tissues from positive women were negative for SARS-COV-2 and showed non-specific alterations, maybe linked to an indirect viral effect. Of note, a higher frequency of preterm deliveries was observed in the SARS-COV-2 positive group. Although wider studies are still needed for patients with infection in different stage of gestation, the data of the present study provides informations that canaid the clinical management of SARS-COV-2 positive pregnant women.

PATOLOGIA GINECOLOGICA

ID 745
MURAL NODULE OF ANAPLASTIC CARCINOMA IN MUCINOUS OVARIAN TUMOR: RARE ENTITY WITH ADVERSE PROGNOSIS

S. Franceschetto[1], A. Guerriero[1], A.P. Dei Tos[1,2]

[1] Department of Pathology, Azienda Ospedale-Università Padova, Padua, Italy; [2] Department of Medicine, University of Padua School of Medicine, Padua, Italy;

Objective: Mucinous cystic tumors of the ovary account for 15% of all ovarian tumors. Rarely they may include mural nodules which are discrete neoplastic proliferations of sarcoma-like mural nodules, true sarcomas and anaplastic carcinomas. The former do not impact in the prognosis, conversely the presence of sarcoma or anaplastic carcinoma is associated with a very aggressive behaviour, mostly in advanced stages.
Materials and methods: Pathologic second opinion was asked to the Department of Pathology of Azienda Ospedale-Università Padova. The patient was a 64-year-old previously healthy woman who underwent excision of a left adnexal mass involving the jejunum. Initial diagnosis was mucinous carcinoma with benign and borderline components, metastatic to the small intestine wall. Diagnostic revision was of a cystic mucinous tumor with mural nodules of anaplastic carcinoma associated with intestinal metastasis. The anaplastic component was positive for pancytokeratins, and negative for mesenchymal markers, hormones receptors and neuroendocrine markers. Patient died of disease with diffuse recurrence few weeks from diagnosis.
Conclusions: Mural nodules occurring in cystic ovarian mucinous tumors represent a rare finding. Anaplastic carcinoma in this setting was first described in 1982[1] and is subclassified into rhabdoid, spindle, and pleomorphic patterns[2] which have no prognostic significance but can be diagnostically challenging. Furthermore, mural nodules of different types may be present in the same neoplasm. Hence, careful histologic examination of this rare lesion is essential for correct patient management.

References

[1] Prat J, Young RH, Scully RE. Ovarian mucinous tumors with foci of anaplastic carcinoma. Cancer 1982; 50:300-304. https://doi.org/10.1002/1097-0142(19820715)50:2<300::aid-cncr2820500222 > 3.0.co;2-e

[2] Provenza C, Yong RH, Prat J. Anaplastic carcinoma in mucinous ovarian tumors: a clinicopathologic study of 34 cases emphasizing the crucial impact of stage on prognosis, their histologic spectrum, and overlap with sarcomalike mural nodules. Am J Surg Pathol 2008;32:383-389. https://doi.org/10.1097/PAS.0b013e3181451b93

ID 825
UTERINE LEIOMYOSARCOMA WITH PLEOMORPHIC LIPOSARCOMATOUS DIFFERENTIATION: A CASE REPORT AND REVIEW OF LITERATURE

A.Muccilli[1], L.Pugliese[2], F.Castiglione[2]

[1] Section of Pathology, Department of Health Sciences, University of Florence, Florence, Italy; [2] Histopathology and Molecular Diagnostics, Careggi Teaching Hospital, Florence, Italy

Objectives: Uterine leiomyosarcomas are uncommon aggressive tumours, accounting for 1-2% of

uterine malignancies. Heterologous differentiation has been recorded even more rarely. We describean extraordinary case of uterine leiomyosarcoma with a pleomorphic liposarcomatous component.
Materials and methods: A 73-year-old, gravida 2, para 2, postmenopausal woman underwent surgical curettage of a uterine mass detected at transvaginal ultrasonography. Following a diagnosis of high-grade sarcoma, hysterectomy with bilateral annessiectomy was performed.
Results: On gross examination, the uterus appeared enlarged by a 6-cm solid intramural mass,white-grey in colour with yellow areas. Histologically, the tumour consisted of aleiomyosarcomatous component (70%) intermingled with a liposarcomatous component (30%), without intervening lipoleiomyomatous areas. The liposarcomatous component had the morphology of pleomorphic liposarcoma with numerous lipoblasts immunoreactive for S100 protein. MDM2 staining was negative. A thorough sampling showed no evidence of a malignant epithelial component thus excluding carcinosarcoma. A final diagnosis of leiomyosarcoma with pleomorphicliposarcomatous differentiation was rendered.
Conclusions: To the best of our knowledge, only four similar cases have been reported in literatureso far. The lack of any benign component implies that sarcoma arose de novo and not as a result of malignant transformation from a benign precursor.

ID 936
IMMUNOHISTOCHEMICAL EXPRESSION OF MESOTHELIN IN A SERIES OF HIGH GRADE TUBAL-OVARIAN SEROUS CARCINOMA

E. Ferioli[1], A. Tafuni[1], S. Tagliaferri[2], N. Campanini[1], R. Berretta[3], G. Giordano[1]

[1] Department of Medicine and Surgery, Pathology Unit, University of Parma; [2] Department of Medicine and Surgery, University of Parma; [3] Department of Medicine and Surgery, Unit of Surgical Sciences, Obstetrics and Gynecology, University of Parma

Objectives: In the current study, immunohistochemical expression of Mesothelin (MSLN), in a series of high grade tubal-ovarian serous carcinoma (HGSC), was analyzed and correlated with BRCA mutation, the stage of development at diagnosis, recurrences and survival, in order to establish whether the expression of this marker could be useful for predicting mutational aspect and clinical outcome of this severe malignancy.
Material and methods: HGSCs were collected from 73 patients who had been surgically treated at Parma University, from 2001 and 2021. For immunohistochemical study the tissue sections of primary, metastatic and recurences were incubated with a mouse monoclonal antibody against mesothelin (clone SP74, ready to use, Ventana-Roche). The proportion of mesothelin expression was evaluated according to this percentage of positive cells: 0, 0%, +1 < 10%, +2, with positivity between 51 and 75%, +3, with positivity between 75% and 95%, +4 with positivity > 95%. The scores + 3 and +4 were considered as intense positivity, the scores +2 and +1 instead as weak positivity. Pattern of intense immunoreactivity mixed with weak positivity was defined as heterogeneous. Chi-square test and Fisher's exact test were performed to investigate the relationship between MSLN expression and clinico-pathological data. We compared disease free survival (DSF), overall survival (OS) and progression free survival (PFS) differences among groups of MSLN expression and disease stages by the Kaplan–Meier method and log-rank tests. $P < 0.05$ was taken as level of significance.
Results: We did not find any significant statistical differences in the MSLN expression between cases with and without BRCA mutation, as well as no differences were observed between low stages (stage I/II) and higher stages of development (stage III/IV) and metastatic tissue.
Some recurrences and post-chemotherapy tissues showed statistically differences, with reduction of expression of MSLN compared with primary neoplasm (respectively p Value: 0,023 and 0,0156).
On contrary, the follow-up revealed that the survival of HGSCs seems to be independent of the MSLN expression.
Conclusions: To summarize, the present study demonstrates that MSLN expression is an independent prognostic factor and particularly this could be used as potential target for targeting therapy in primary and metastatic neoplasm.

ID 939
MALIGNANT MELANOMA ARISING ON MATURE CYSTIC TERATOMA: FROM A CLINICAL CASE TO META-ANALYSIS

M. Paudice[1], V. Parrella[1], Z. Leoni[1], P. Bracco[2], R. Brizio[3], V.G. Vellone[4]

[1] Department of Integrated Surgical and Diagnostic Sciences (DISC), University of Genoa, Italy; [2] Gynecology and Obstetrics, Hospital of Imperia; [3] Pathology, S. Croce & Carle Teaching Hospital, Cuneo; [4] Anatomic Pathology University Unit, IRCCS Ospedale Policlinico San Martino, Genoa, Italy

Objectives: Mature cystic teratoma of the ovary (MCT) is one of the most frequent female tumors, whereas its malignant transformation is uncommon (2%); malignant melanoma (MM) is a rare histotype (0.2-0.8%) counting around 50 cases in literature. After evaluating a case in our practice, we present a metanalysis concerning clinical presentation, diagnosis, macroscopic and microscopic aspects, treatment and prognosis of MM in MCT.
Materials and methods: Research on Pubmed/

Google Scholar was performed in January 2022 to gather data on patients with MM in MCT using keywords, such as: "melanoma in mature cystic teratoma", "ovarian melanoma", "dermoid cyst melanoma". Inclusion criteria are: time of publication (> 2000); article types: case reports, case series, letters to editor, systematic reviews; full-text articles; English articles; presence of teratomatous elements by histological examination.
Clinical presentation, surgical treatment, histological characteristics, follow up and prognostic factors were considered.
Results: 33 articles met our inclusion criteria totaling 36 patients plus our case. The average age at diagnosis is 49.1 years old; 60% of the patients have localized disease (of which 30% have demonstrated recurrence of disease), whereas 40% have metastatic disease. MM in MCT presents as a solid-cystic mass with a mean diameter of 14.1 cm. By histological examination, a melanocytic proliferation is present, sometimes associated with epithelioid cells, spindled cells and necrosis. The average time from diagnosis to death is 10.3 months and it occurs in 58.6% of the patients.
Conclusions: Metastatic disease and mass diameter (> 10 cm) demonstrate to have an impact on patients' survival, probably the presence of spindled cells and necrosis too. Even though there is not a consensus on treatment, surgery remains the gold standard in association with complete surgical staging.

PATOLOGIA MAMMARIA

ID 753
CYSTIC NEUTROPHILIC GRANULOMATOUS MASTITIS (CNGM): AN UNUSUAL FORM OF GRANULOMATOUS MASTITIS

Coppola G[1], Pisano A[1], Pernazza A[2], d'Amati G[1], Cerbelli B[2].

[1] Department of Radiological, Oncological and Pathological Sciences, ‹Sapienza› University of Rome, Rome, Italy; [2] Department of Medical-Surgical Sciences and Biotechnologies, Sapienza University of Rome, Rome, Italy

Cystic neutrophilic granulomatous mastitis (CNGM) is an extremely rare type of mastitis, often associated with *Corynebacterium* species.
Objectives: our purpose is to analyze the clinical and pathological features of CNGM.
Materials and methods: we retrieved all cases of granulomatous mastitis (GM) diagnosed in our Institution from 2018 to 2022. We reviewed all the H&E slides and Gram stains and collected patients' relevant clinical and radiological features.
Results: in our series, CNGM accounted for 4 out of 9 cases of GM (44%). Main histological features were the presence of optically empty vacuoles rimmed by neutrophils, with an outer layer of epithelioid histiocytes surrounded by a lymphocytic infiltrate. In 2/4 cases some of the vacuoles contained ropey material with rod-shaped, Gram-positive bacilli. Interestingly, the diagnosis was always made on surgical samples from patients with invasive breast cancer; in 3 of 4 cases it was an incidental finding in the breast parenchyma surrounding the neoplasia while in the remaining case it was clinically misinterpreted as an additional focus of cancer.
Conclusion: CNGM is a specific type of mastitis which, in a subset of cases, can benefit from targeted antibiotic therapy. This recently described entity is probably still under recognized and must be differentiated on histology from other forms of granulomatous mastitis. According to our experience this entity can simulate malignancy on clinical examination and imaging.

ID 777
PHYLLODES BREAST TUMOR: DIAGNOSTIC CORRELATION BETWEEN PREOPERATIVE BIOPSY AND HISTOLOGICAL SAMPLE

[1]Lina Cardisciani, [1]Angela Santoro, [1]Damiano Arciuolo, [2]Francesca Addante, [1]Federica Cianfrini, [1]Federica Vegni, [1]Federica Policardo, [1]Giulia Scaglione, [1]Alessia Piermattei, [1]Antonino Maria Andrea Mulè

[1] Unit of General Pathological Anatomy, Department for Women's Health Sciences, Children and Public Health, Fondazione Policlinico Universitario "Agostino Gemelli" IRCCS, Rome, Italy. [2] Department of Emergency and Organ Transplantation, Section of Anatomic Pathology, School of Medicine, University "Aldo Moro", Bari, Italy

Objectives: Phyllodes tumors (PTs) are rare breast fibroepithelial neoplasms, that are classified into benign, borderline and malignant based on their stromal morphology. Our study aims to assess the degree of concordance between core needle biopsy (CNB) diagnoses and those assessed in the definitive operative specimens.
Materials and methods: We analyzed data from 42 female patients aged between 12 to 79 years by comparing the diagnoses of CNB examination and definitive histological report.
Results: Forty-two breast biopsies and relative surgical samples were analyzed. The concordance between preoperative and definitive diagnoses was 21/25 (84%) for benign PTs, 10/12 (83%) for borderline PTs, and 5/5 (100%) for malignant PTs. Specifically, 4/25 (16%) benign PTs in needle biopsies were evaluated as borderline PTs at the histological examination. 2/12 (16.6%) borderline PTs in needle biopsies was upgraded as malignant at surgery.
Conclusion: Our study showed that CNB can be considered the most useful investigation for the preoperative diagnosis of PTs. In our studies malignant PTs

had 100% concordant diagnoses between CNBs and definitive histologic examination. Moreover, all malignant/borderline PTs were not understimated on CNBs.

ID 783
GRANULAR CELL TUMOR OF THE BREAST: A SERIES OF FIVE CASES

F. Vegni, S. Sfregola, L. Cardisciani, F. Policardo, G.Scaglione, N, D'Alessandris, D. Arciuolo, A. Santoro, G.F. Zannoni, A.M.A. Mulè

Unit of General Pathology Anatomy, Department for Women's Healt Sciences, Children and Public Healt, Fondazione Policlinico Universitario "Agostino Gemelli" IRCCS, Largo A. Gemelli 8, 00168, Rome, Italy

Objectives: Granular cell tumors are uncommon soft tissue tumors derived from the nerve sheath, that are benign in 98-99.5% of cases. They can occur anywhere in the body, but head and neck are the most common sites of origin. GCT of the breast are rare (4-6% of all GCT) and they canclinically, radiologically and macroscopically mimic breast carcinoma.
Materials and Methods: We reviewed the histologic samples of five patients affected by GCT of the breast treated over the course of ten years (January 2011-January 2021) at the Fondazione Policlinico Universitario Agostino Gemelli IRCCS of Rome, Italy.
Results: All of the five cases presented as solitary, painless and firm lumps, palpable in three of five cases; the radiological findings were non-specific and heterogeneous. Gross examination showed a nodular appearance, greyish-white to pale yellow in colour and well circumscribedmargins in three of five cases. Histologically they were composed of large, polygonal cells with abundant, eosinophilic and granular cytoplasm and small central nuclei. These cells resulted positive at the immunohistochemistry for S100, Vimentina, CD56, while they resulted negative for HMB45, MelanA, AE1/AE3, EMA and Desmin in all the cases. Furtheremore three cases resulted positive for CD68 and MITF.
Conclusions: GCT is a rare and usually benign breast tumor that can mimic breast cancer. Treatment of choice consists in wide resection or lumpectomy with margin assessment. Histological and immunohistochemical examination are necessary to make a definitive diagnosis.

ID 858
TUMOR MICROENVIRONMENT AND RISK PROFILE ANALYSIS OF BREAST CANCER DURING PREGNANCY, A SPECIAL TYPE OF EARLY-ONSET BREAST CANCER

E. Sajjadi[1,2], K. Venetis[1,2], M. Noale[3], G. Bonizzi[1], S. Ferrero[4], S. Maggi[3], G. Viale[1,2], F.A. Peccatori[1], E. Guerini-Rocco[1,2], N. Fusco[1,2]

[1] IEO, European Institute of Oncology IRCCS, Milan, Italy; [2] University of Milan, Milan, Italy; [3] Neuroscience Institute Aging Branch, Padua, Italy; [4] Fondazione IRCCS Ca' Granda – Ospedale Maggiore Policlinico, Milan, Italy

Objectives: Breast cancer during pregnancy (PrBC) is a rare tumor with only a little information on its immune landscape. Here, we sought to characterize the cellular composition of the tumor microenvironment (TME) of PrBC and identify its differences from early-onset breast cancer (EOBC) in non-pregnant women.
Materials and Methods: A total of 83 PrBC and 89 EOBC were selected from our Institutional registry and subjected to tumor-infiltrating lymphocytes (TILs) profiling according to TILs Working Group recommendations. Immunohistochemistry for CD4, CD8, FOXP3, and PD-L1 (clone 22C3) was performed. Fisher's and Chi-squared tests, multinomial logistic regression models, ROC curve and survival analyses were performed.
Results: A significantly lower frequency of hormone receptor (HR)-positive tumors was observed in PrBC (ER+ n = 46, 55.4% vs. n = 65, 73.0%; PgR+ n = 43, 51.8% vs. n = 63, 70.8%; $p < 0.01$). The prevalence of low/null PD-L1 and CD8+TILs was higher in PrBC than in the controls, specifically in HR+/HER2- breast cancers (CD8: n = 68, 81.9% vs. n = 61, 68.5%, $p = 0.04$; PD-L1: n = 82, 98.8% vs. n = 76, 85.4%, $p = 0.001$). Overall, PrBC had higher risk of relapse (n = 36, 44.4%) and disease-related death (n = 16, 19.8%) than EOBC (n = 6, 10.7%, $p = 0.01$; and n = 2, 3.6%, $p = 0.01$; respectively) independent of tumor subtype and HR/HER2 status. The presence of TILs and each TIL subpopulation were significantly associated with disease relapse. Moreover, the death rate was higher in PrBC with CD8+ TILs (n = 13, 19.7% vs. n = 0, $p = 0.001$). Kaplan-Meier confirmed worse disease-free survival of PrBC ($p = 0.01$).
Conclusions: PrBC TIME is characterized by specific TIL subpopulation patterns with significant biological and prognostic roles. TILs assessment would be a valid addition to the pathology report that might help to identify clinically relevant PrBC subsets.

PATOLOGIA MOLECOLARE

ID 856
FREE CANCER CELLS DETECTION IN INTRAPERITONEAL LAVAGE OF PATIENTS WITH GASTROESOPHAGEAL JUNCTIONADENOCARCINOMA

I.M. Montagner[§,1], M.R. Biasin[§,1], A. Volpato[1], V. Mourmouras[1], C.M. Biatta[1], R. Alfieri[2], M.A. Piano[3], M. Curtarello[3], P. Pilati[2], and A. Scapinello[1]

[§]co-first authors; [1] Pathology Unit, Veneto Institute of Oncology IOV-IRCCS, Castelfranco V.to, Italy; [2] Oncological Sur-

gery Unit, Veneto Institute of Oncology IOV-IRCCS, Castelfranco V.to, Italy; [3] Immunology and Molecular Oncology, Veneto Institute of Oncology IOV-IRCCS, Padova, Italy

Objectives: The prognosis of patients with gastro-esophageal junction adenocarcinoma (GEJA) is dismal and surgical resection remains the cornerstone of treatment. Unfortunately, this curative option presents a high risk of dissemination of neoplastic cells in the abdominal cavity, correlating with the development of peritoneal recurrences and metastases. Extensive intraperitoneal lavage (EPIL) is a new strategy to reduce cancer cell dissemination. Conventional cytology is routinely utilized for free cancer cells detection in peritoneal lavage (PL), despite it is a method with limited sensitivity. Recently, molecular approaches, as reverse transcriptase-polymerase chain reaction (RT-PCR), have been regarded as a better tool to increasediagnostic accuracy. The aim is to improve the detection rate of free cancer cells from peritoneal cavity after surgery, combining cytological technique with RT-PCR.
Materials and methods: PL samples were collected in 8 preliminary GEJA patients at the end of three different time point: exploration of peritoneal cavity (PL1), demolition phase (PL2), and EIPL (PL3). For each sample, one half was use to extract RNA for evaluating the expression levels oftarget genes (CEA, CK20, and SYT13) by RT-PCR. Whereas, cytology and immunohistochemical(IHC) analysis of the target antigens were performed on the remaining aliquot.
Results: Assuming PL1 as basal level value in RT-PCR analysis, target genes expression increasedin PL2 and decreased in PL3 in detectable samples. The cytological analysis identified tumor cells in two cases, while target genes positivity in IHC was assessed in five patients with lymph node metastases. Three of them developed peritoneal recurrence at one-year follow-up.
Conclusions: CK20 seems to be the most promising marker to evaluate EPIL efficacy. The implementation of the clinical records will allow both the correlation between IHC and lymph node metastases, and the comparison between molecular and cytological analysis.

ID 882
REFLEX-TEST IN NON-SMALL CELL LUNG CANCER: AN EFFECTIVE TWO-STEP APPROACH FOR 10 BIOMARKERS ANALYSIS

L. Caggiari[1], M. Battiston[1], G. Pelizzari[2], A. Follador[2], S. Cernic[1], S. Ciani[1], G. Fasola[2], S. Pizzolitto[1], G. De Maglio[1]

[1] SOC Anatomia Patologica, Azienda Sanitaria Universitaria Friuli Centrale, Udine, Italia; [2] Dipartimento di Oncologia, Azienda Sanitaria Universitaria Friuli Centrale, Udine, Italia

Objectives: Recent evolution in targeted therapies emphasizes the need of fast-track screening of multiple molecular biomarkers for first-line treatment selection in Non-small Cell Lung Cancer (NSCLC).
At our institution, we implemented a two-step *Reflex*-test (Rt) at the time of pathological diagnosis of non-squamous NSCLC for EGFR, KRAS, BRAF, ERBB2, ALK, ROS1, PDL-1, RET, MET, and NTRK testing.
Aim of this study is to evaluate the clinical utility of two-step molecular Rt in NSCLC.
Materials and Methods:
In the period November 2020-January 2022, 251 specimens were submitted for molecular Rt: DNA genes mutations have been screened by a hotspot panel, ALK and PDL-1 expression was analyzed by immunohistochemistry, ROS1 rearrangements investigated by FISH.
All DNA-ALK-ROS1 negative cases were reflexed to second level RNA Rt by real-time PCR for MET exon 14 skipping, RET and NTRK. NTRK was also evaluated by immunohistochemistry.
The turnaround time (TAT) was defined as the number of consecutive days from the histopathologic report to the final molecular report.
Results: Genetic mutations rate detected in first level Rt were EGFR 13%, KRAS-G12C 17%, other KRAS 22%, BRAF V600E 3%, ERBB2 1%, ALK 4%, ROS1 < 1%; 94 patients were reflexed to second-level Rt with alterations incidence of METex14 4%, RET < 1%, NTRK 0%.
Failure rates were 1% and 8% for DNA and RNA Rt, respectively.
The overall medium TAT was of 9,6 days: mean TAT was 7,2 days for first-level Rt positive cases and 16,5 days for second-level Rt.
Conclusions: Rate and type of pathogenic variants identified were consistent with literature data. These findings showed that a standardized comprehensive strategy for molecular testing increases opportunities for personalized therapy in NSCLC patients. Rt is desirable to minimize the interval between histo/cytopathologic diagnosis and the initiation of first-line therapies. A two-step Rt approach is effective to minimize failure rate due to RNA analysis along with an optimizedclinically useful reporting time.

ID 911
LUNG ADENOCARCINOMA CELLS CULTURED AS 3D SPHEROIDS ARE RESISTANT TO FERROPTOTIC CELL DEATH

F. Ascenzi[1]*, A. Esposito[2]*, S. Bruschini[1], V. Salvati[3], C. De Vitis[1], S. Scarpino[1], G. Ciliberto*, R. Mancini[1]

*[1] Department of Clinical and Molecular Medicine, Sapienza University of Rome, Rome, Italy; [2] Department of Experimental and Clinical Medicine, Magna Graecia University of Catanzaro, Catanzaro, Italy; [3] Preclinical Model and New Therapeutic Agents Unit, IRCSS Regina Elena National Cancer Institute, Rome, Italy; [4] Scientific Directorate, IRCSS Regina Elena National Cancer Institute, Rome, Italy; *Equal contributors*

Objectives: Scientific literature supports the evidence that Cancer Stem Cells (CSCs) retain inside low Reactive Oxygen Species (ROS) levels and are therefore less susceptible to cell death, including ferroptosis, a type of cell death dependent on iron-driven lipid peroxidation. Our objective was to investigate the ferroptosis pathway in a three-dimensional (3D) model of CSCs in Lung Adenocarcinoma (LUAD).
Materials and methods: An established collection of LUAD cell lines derived from malignant pleural effusions of patients was used to obtain 3D spheroids enriched for stem-like properties. Viability assays were performed to evaluate the susceptibility to ferroptosis activators in 2D and 3D conditions. The lipid peroxidation of treated cells was evaluated by flow cytometry. Real Time PCR and western blot analyses were carried out to investigate the molecular mechanisms underlying ferroptosis resistance in LUAD-CSCs.
Results: We observed that the ferroptosis inducer RSL3 triggers lipid peroxidation and cell death in a panel of primary tumour cell lines isolated from LUAD patients. The sensitivity to RSL3 varies greatly across cancer cell lines; however, when grown in 3D condition, all cell lines exhibit substantial resistance to RSL3 and therefore protection against ferroptotic cell death. Molecular analyses suggest that this phenomenon correlates with an increased expression of antioxidant genes and high levels of proteins involved in iron storage and export, indicating protection against oxidative stress and lowavailability of iron for the initiation of ferroptosis.
Conclusions: We demonstrated that CSCs-enriched 3D spheroids are resistant to ferroptosis and could constitute a useful testbed for precision medicine strategies involving ferroptosis.

PATOLOGIA PEDIATRICA

ID 820
FATAL CEREBRAL HEMORRHAGE CAUSED BY SARS-COV-2 IN A 2 YEARS OLD FEMALE

D. Colombo[1], F. Del Nonno[1], L. Falasca[1], P. Francalanci[2], C. Russo[3], P. Bernaschi[3], CF Perno[3], AO Muda[4], F. Vaia[5], R. Alaggio[2]

[1] Pathology and Electron Microscopy Unit, National Institute for Infectious Diseases L. Spallanzani, IRCCS, Rome, Italy; [2] Pathology Unit, Bambino Gesù Children's Hospital, IRCCS, Rome, Italy; 3 Microbiology and Immunology Unit, Bambino Gesù Children's Hospital, IRCCS, Rome, Italy; 4 Dept. of Laboratories, Bambino Gesù Children's Hospital, Rome, Italy; [5] General Direction, National Institute for Infectious Diseases L. Spallanzani, IRCCS, Rome, Italy

Objectives: A 2 yrs old girl, SARS-CoV-2 positive, deceased at the emergency room, where she was admitted for a sudden neurological sign and loss of counsciousness associated to respiratory symptoms and high fever. The purpose of this study was to find out the possible correlation, in pediatric patients, between Sars-CoV-2 infection and lethal neurological manifestations through extensive and detailed postmortem analysis.
Materials and Methods: A complete postmortem examination, including brain, was performed at National Institute for Infectious Diseases Lazzaro Spallanzani. Autopsy was performed following biosafety practices. Organs were sampled and placed in formalin for 72 hours. Deparaffinized and rehydrated sections were used for H&E staining and for immunohistochemistry. Quantitative reverse transcriptase polymerase chain reaction (RT-PCR) testing for SARS-CoV-2 was performed on tissue samples from lung, heart, brain, kidney, intestine, liver.
Results: The autoptic examination revealed acute lobar Staphylococcal pneumonia associated with Sars-CoV-2 lung acute interstitial damage. Brain inspection showed herniation of the cerebellar tonsils, cerebral edema with no apparent underlying cause (meningitis, encephalitis), skull base subarachnoid hemorrhagic suffusion with hemorrhagic infarction of the brainstem and necrotic hemorrhagic area of the occipital lobes bilaterally. A small incidental neurocytoma (III ventricle) was found. Sars-CoV-2 PCR was found positive in cerebral tissue as well as in all examined organs, nevertheless no other relevant alterations were found except for lung and brain.
Conclusions: This was one of the rare documented cases of pediatric death due to severe Sars-CoV-2 infection with acute lung damage and viral dissemination leading to cerebral hemorrhage.

PATOLOGIA PLEUROPOLMONARE

ID 788
MICRONODULAR THYMOMA WITH LYMPHOID STROMA: FOUR CASES WITH CLINICO-PATHOLOGICAL FEATURES AND REVIEW OF MAGGIORE HOSPITAL SERIES

Sofia Melotti[1-2], Federico Chiarucci[1-2], Nazarena Nannini[1], Stefania Damiani[3-4], Costantino Ricci[1-4], Pietro Bertoglio[5], Piergiorgio Solli[5], Francesca Ambrosi[1] and Michelangelo Fiorentino[1-4]

[1] Pathology Unit, Maggiore Hospital, AUSL Bologna, Bologna, Italy; [2] Department of Biomedical and Neuromotor Sciences, Unit of Anatomic Pathology, University of Bologna, Bellaria Hospital, Bologna, Italy; [3] Pathology Unit, IRCCS Policlinico Sant'Orsola-Malpighi, University of Bologna, Bologna, Italy; [4] Department of Experimental, Diagnostic and Specialty Medicine (DIMES), University of Bologna, Bologna, Italy; [5] Division of Thoracic Surgery, IRCCS Azienda Ospedaliero-Universitaria di Bologna, Bologna, Italy

Objectives: Micronodular Thymoma with Lymphoid

Stroma (MNTLS) is a rare subtype of thymic neoplasmfirst reported by Suster and Moran in 1999. It accounts for about 1-5% of all thymomas and only a few cases have been described to date, especially where the long-term prognosis is concerned. MNTLS presents typical histopathologic features, namely multiple diffuse or fused epithelial nodules in an abundant lymphocyte stroma with prominent germinal centers.
From a clinical perspective MNTLS is typically asymptomatic and often an incidental finding. It grows slowly and is usually either encapsulated or minimally invasive so, according to ITMIG statistics, 96% of MNTLS is low MM stages I and II. Myasthenia gravis or paraneoplastic syndrome are not more frequently associated than other subtypes of thymoma. Sometimes, on physical and radiological examination, patients present thymic cyst, cardiac myxoma, and salivary gland damage.
Our aim is to describe four cases of MNTLS, considering clinico-pathological features anddifferential diagnosis.
Materials and methods: All cases of thymoma which underwent surgery in Maggiore Hospital from January 2015 to May 2022 have been retrieved from the pathology archives and reviwed; 60 cases of thymoma (surgical specimens only) were found, and only the MNTLS were taken into account. For each case all clinicopathological, histological and immunohistochemical features have been collected according to the present WHO classification and staged according to AJCC 8th edition and Masaoka-Koga (MM) classification.
Results: 60 cases of thymus surgical specimens have been collected. There were 30 females and 30 malespatients (male-to-female ratio 1:1) with a mean age of 61 (min. age 32, max. age 82). All cases have been classified as following: 1 type A thymoma (1.6%), 10 type B1 thymoma (10%), 11 type B2 thymoma (18.3%), 4 type B3 thymoma (6.6%), 23 type AB thymoma (38.3%), 1 type B1 plus B2 thymoma and 3 cases of type B2 plus B3 thymoma (6.6%); considering the special histotypes 3 cases of microthymoma (5%) and 4 cases of micronodular thymoma with lymphoid stroma (6.6%). All cases have been staged according to AJCC 8th ed.: 25 cases (41.6%) resulted pT1a, 3 cases (5%) pT1b, 17 cases (28.3%) pT2 and 2 cases (3.3%) pT3.
Focusing specifically on MNTLS, two females and two males patients presented it (male-to-female-ratio 1:1) with a mean age at diagnosis of 74 years (min. age 67, max. age 80). From a clinical standpoint patients were asymptomatic and presented a mediastinic mass, while only in one case (1/3) there was a clinical association with myasthenia gravis. Grossly, the lesions were white-greyish mass whose maximum dimension was comprised between 3 cm and 7 cm (mean diameter 5 cm).
When subjected to microscopic analysis, the masses consisted of a nodular epithelial component interspersed in a lymphoid stroma with some lymphoid follicles which showed prominent germinal centers. With regards to nodular structures, the epithelial cells had a spindle to round appearance, with bland euchromatic nuclei, inconspicuous nucleoli and plump cytoplasm. There was no evidence of hemorrhage, necrosis or cytologic atypia. In all four cases the neoplasia infiltrated the fibrous capsule and extended to the anterior and posterior mediastinal adipose tissue. Only one case showed pleura infiltration. In all cases the excisional margins were free from neoplasia. One case showed adipose involution and microcystic changes with uni and multilocular cysts lined by flattened epithelium in the remaining parenchyma. All examined lymph nodes were free from neoplasia. In one case the MNTLS have been mixed with a nodule with type A thymoma component. The staging was pT1a N0 /stage II MM in three patients while there was only one pT1b N0 /stage III MM.
Immunohistochemistry highlights the epithelial cells, which resulted positive for pan cythocheratin and p63; negative for CD3, CD5, CD20, TdT and GATA3. Epithelial cells were constantly negative for CD117, CD5, chromogranin and synaptophysin.
Conclusions: In our Series of 60 thymoma cases 4 (6.6%) qualified as MNTLS with no gender predilection and a mean age at diagnosis of 74. All cases belonged to a low staging group comprised between pT1a and pT1b N0 or stage II and III MM. All patients are in follow-up (month mean 19) and none of them have developed recurrences or metastasis.
The general consensus is that MNTLS is a particularly rare thymic tumor and presents a typical morphology. It mostly occurs in middle-aged and elderly people and often develops in the anterior mediastinum, although there are also reports of cervical ectopic MNTLS.
The principal differential diagnosis of MNTLS are: type A thymoma characterized by large patches of mild fusiform or oval-shaped epithelial cells with no or few lymphocytes in the interstitium; type B thymoma, mainly composed of immature T lymphocytes and perivascular spaces; type AB thymoma composed of a combination of type A thymoma with fewer lymphocytes and type B; micronodular thymic carcinoma with lymphoid stroma, a rare variant of thymic carcinoma whosemorphological features such as invasive growth pattern, abnormally shaped epithelioid cells and,often, capsule invasion and necrosis, are crucial in differential diagnosis with the thymoma counterpart.
At present MNTLS is regarded as a borderline tumor with good prognosis and rare recurrences and metastasis, whose treatment predominantly involves surgical resection. Patients with cystic solid tumors have a better prognosis, while those with solid pattern and capsule invasion have a higher risk index. Based on our findings, MNTLS could represent a not so rare histotype andmorphological key features, supported by an immunohistochemical panel, could be useful

in differential diagnosis for better classifying these tumors.

ID 942
OPTIMIZATION OF STANDARDIZED AUTOPSY "VIRCHOW TECHNIQUE" FOR PATIENTS WITH CERTAIN OR SUSPECTED SEVERE ACUTE RESPIRATORY SYNDROME CORONAVIRUS(SARS-COV-2) INFECTION

E. Stigliano[1], M. Dell'Aquila [1], G. Amadoro [2], G. Rindi [1], Lavitrano M.,[3] U. Di Tondo[1], V. Arena[1], A. Capelli[1]

[1] Catholic University of Sacred Heart, Largo Francesco Vito, 1, 00168-Rome (Italy); [2] National Research Council, Institute of Translational Pharmacology (IFT), Via Fosso del Cavaliere 100,00133-Rome (Italy)[3] University Milano Bicocca, Department of Medicine and Surgery, Director BBMRI.it

Objectives: Following our experience in the preparation and execution of 200 autopsies of patients who died from Sars-CoV-2 infection (COVID-19) syndrome, here we provide an optimization of the classical autopsy protocol of "Virchow technique" that may help the autopsy staff in reducing the risks encountered by exposure to Hazard Group 3 organisms (HG3).
Materials and methods: In a negative pressure Biosafety Level 3 (BSL-3) sector room, complete postmortem examinations were performed by a trained team equipped wih Biosafety equipment (PPE). An innovative, minimally invasive thoracoscopic transdiaphragmatic autopsy approach was devised in order to (i) limit the time of exposure to the potentially infectious lung and upper airway organs and biological fluids, (ii) allow for a simpler re-composition of the body.
Results: This autopsy technique allowed us to study the COVID-19 disease with histological (Hematoxylin and Eosin) and molecular (real-time Reverse Transcriptase Polymerase Chain Reaction; rRT-PCR) investigations. Formalin fixed paraffin embedded material (FFPE) and frozen samples were also taken in the course of the autopsy. The presence of SARS-CoV-2 was widely documented especially at the level of the lung parenchyma and upper respiratory tract secretions in accordance with the clinical data in more than 95% of the autopsy cases.
Conclusions: A variant of the classic "Virchow technique", consisting of a transdiaphragmatic autopsy approach was successfully tested to collect pathological, high infectivity specimens from Covid19-deceased subjects. The large number of clinical data and high quality biological samples collected represent an important contribution to future BBMRI. it COVID-19 "biobanking" activities. We express our immense gratitude to Prof. A. Capelli who recently passed away for his long-lasting and exceptional professional expertise, compassion and empathy.

ID 961
SMALL CELL NEUROENDOCRINE THYMIC CARCINOMA COMBINED WITH TYPE B3 THYMOMA: A CASE REPORT

V. Pasini[1], T. Tatti[1], L. Voltolini[2], C.E. Comin[3]

[1] Section of Anatomic Pathology, Department of Health Sciences, University of Florence, Italy; [2] Thoracic Surgery Unit, Careggi University Hospital, Florence, Italy; [3] Department of Experimental and Clinical Medicine, University of Florence, Italy

Introduction. Small cell carcinoma is a high-grade thymic tumor that account for approximately 10% of all thymic neuroendocrine tumors, with an incidence of one case per 50 million individuals. We report a case of small cell thymic carcinoma combined with invasive type B3 thymoma.
Case. During the follow-up, a 73-year-old man with a previous thymectomy and right superior lobectomy for invasive type B3 thymoma combined with a type B2 thymoma (Masaoka-Koga stage IVa, TNM pT4 N0), performed a chest CT that showed pleural nodularities on the left. After chemotherapy and multidisciplinary discussion, indication to surgical intervention of pleural implantation was given. On macroscopic evaluation pleural specimens showed three solid-cystic neoformations, of 5, 2.5 and 3 cm maximum diameter respectively. Microscopically, the lesions were characterized by tumor lobules separated by fibrous septa with prominent perivascular spaces. Two admixed epithelial cellular components were observed: the first one consisting of polygonal cells with eosinophilic cytoplasm, round and elongated nuclei mixed with a small component of immature T lymphocytes; the second one consisting of small cells, characterized by scantcytoplasm, round nuclei with "salt and pepper" chromatin, with high proliferative activity performed by ki-67 (> 80%). The cellular component referable to type B3 thymoma showed the following immunohistochemical (IC) profile: CAM 5.2+, CK7+, P63+, PAX-8+, Synaptophysin-, CD56-, CD117-, CD5-, TTF-1-, with TdT-positive lymphocytes. The small cell component ICprofile was: CAM 5.2+, CK7+, Synaptophysin+, CD56+, P63-, PAX-8-, CD117-, CD5-, TTF-1-. The final diagnosis was pleural localization of small cell carcinoma combined with type B3 thymoma.
Conclusions: **.** Small cell neuroendocrine carcinoma is a rare type of thymic epithelial tumor. Literature data reported thymic neuroendocrine carcinoma with an additional component of other thymic epitelial tumors. We report a rare case of dedifferentiation of small cell carcinoma combined to type B3 thymoma after chemotherapy.

PATOLOGIA TESTA-COLLO

ID 785
NUT-CARCINOMA OF THE PAROTID: A RARE TUMOR IN A VERY UNUSUAL LOCATION.

M. Vinci[1], L. Alessandrini[1], M. Sbaraglia[1], A.L.C.Carobbio[2], M. Fassan[1], A.P. Dei Tos [1], L.Alessandrini1, M. Ferrari2, E. Savietto2, D. Cazzador2, M. Sbaraglia1

[1] Department of Medicine- DIMED, Section of Pathology, University of Padova, Padova, Italy; [2] Department of Neuroscience DNS, Otolaryngology Section, University of Padova, Padova, Italy

Objectives. NUT carcinoma is a rare and aggressive tumor that usually arises from midline structures and harbors a characteristic rearrangement of the *NUT* gene. To date only 11 cases occurring in the parotid of adults have been reported[1].
Materials and methods. A 42-year-old man underwent incomplete right parotidectomy with neck dissection and adjuvant radiotherapy in 2016 in another Hospital, due to an original diagnosis of primary SCC. In 2022, a relapse in right parapharyngeal space extended up to skull base was biopsied in our Hospital. Besides routine assessment, Next Generation Sequencing (NGS) analysis using Archer FusionPlex Sarcoma v2 Panel was performed on an Illumina platform (NextSeq). Sequence analysis was carried out by Archer Analysis v6.0.
Results. Histological examination showed infiltrative solid nests composed of large, discohesive mitotically active, cells with eosinophilic cytoplasm, hyperchromatic nuclei, prominent nucleoli and isolated foci of abrupt keratinization, in a fibrous stroma. Neoplastic cells were strongly immunopositive for p63, p40, CK5/6 and NUT, displayed a focal positivity for CK7, p16 and were negative for SOX10, S100, CD99 and neuroendocrine markers. In situ hybridization for EBER was negative as well. NGS analysis identified the *BRD4-NUTM1* fusion, confirming the diagnosis of NUT carcinoma.
Conclusions: Primary NUT carcinoma is a diagnostic challenge that should be considered in the differentials of primary and metastatic high-grade carcinomas in the parotid (mainly mucoepidermoid/ myoepithelial/squamous cell/ neuroendocrine/sinonasal undifferentiated carcinomas), to provide important prognostic information and to guide emerging therapeutic strategies targeting the underlying gene fusion. NUT expression and *NUTM1* rearrangement are helpful to avoid misdiagnosis.

References

1. Chen M, Zhao S, Liang Z, et al. NUT carcinoma of the parotid gland: report of two cases, one with a rare ZNF532-NUTM1 fusion. Virchows Arch. 2022;480(4):887-897.

ID 806
INTESTINAL-TYPE ADENOCARCINOMA OF THE TONGUE: DETAILED IMMUNOHISTOCHEMICAL AND NGS ANALYSIS

G. Veronesi[1], L. Alessandrini[1], BM. Del Forno[1], R. Salmaso[1], M. Ferrari[2],M. Sbaraglia[1], M. Fassan[1], A.P. Dei Tos[1], L.Alessandrini1, M. Ferrari2, E. Savietto2, D. Cazzador2, M. Sbaraglia1

[1] Department of Medicine- DIMED, Section of Pathology, University of Padova, Padova, Italy; [2] Department of Neuroscience DNS, Otolaryngology Section, University of Padova, Padova, Italy

Objectives: Intestinal-type adenocarcinomas (ITACs) are aggressive tumors, mainly occurring in the sinonasal tract. Primary ITAC of the tongue are extremely rare, with only 8 cases reported.
Materials and methods: A 30-year-old man presented in November 2021 with an ulcerated tongue mass and lymphadenopathy. Extensive imaging examination failed to identify any other primary tumor. After ITAC was diagnosed on biopsy, the patient underwent total glossectomy with later neck dissection. The tumor was characterized by immunohistochemistry, MSI analysis and targeted NGS (*ALK, BRAF, EGFR, ERBB2, FGFR3, HRAS, IDH1, IDH2, KIT, KRAS, MET, NRAS, PDGFRA, PIK3CA, RET, ROS1*).
Results: Pathologic examination confirmed a submucosal, G2, pT3N2b adenocarcinoma forming tubuloglandular structures with central necrosis. Diffuse perineural and lymphovascular invasion and a positive excision margin were detected. Neoplastic cells showed positive immunostaining for CK20, CDX2, and were negative for CK7, HER-2, ALK, ROS1, PanTRK; PD-L1 CPS was 7 and p53 had a wild-type expression pattern. The expression of mismatch repair proteins (MMR) was retained and the tumor was MSS at MSI analysis.
No alteration was observed by NGS, as were previously reported cases analyzed by traditional sequencing techniques for *KRAS, NRAS, BRAF, PIK3CA* genes.
Postoperative chemotherapy/radiotherapy were administered. Six months later, the patient is alive, with radiological evidence of lung metastases.
Conclusions: The diagnosis and therapy of lingual ITACs are very challenging. Further studies on larger series could identify potential therapeutic targets to improve patients' poor prognosis.

ID 912
HISTOGENETIC HYPOTHESIS ABOUT SIALOLIPOMA OF THE PAROTID GLAND: A TRULY REPRESENTATIVE BENIGN NEOPLASM OR AN HAMARTOMATOUS LESION

Santarelli M, Pepi L, Paparesta F, Rubini C, Filosa A, Goteri G,

Pathology Unit, Department of Biomedical Sciences and Public Health, United Hospitals, Polytechnic University of Marche, Ancona, Italy

Objective: Sialolipoma is a rare histological variant of lipoma composed of a proliferation of mature adipocytes with secondary entrapment of normal salivary gland tissue. In the 5th Edition of the World Health Organization Classification of Head and Neck Tumors, sialolipoma is the only entity included in the chapter of mesenchymal tumours. The clinical and radiological characteristic (i.e pushing borders, slowly growing encapsulated mass) are often confused with other benign salivary gland tumour. When the epithelial component predominate also cytological features overlap with those of oncocytic lesion of the parotid gland. Histogenesis of sialolipoma is uncertain, although some theories have been proposed. Sialolipoma may represent an adenoma mainly composed of mature adipose tissue, a benign lipoma with entrapped salivary gland elements, or it could be the result of a a hamartomatous process.
Material and methods: We report a case of man, 50 years old, with an asymptomatic slowly growing parotid lesion 2 cm in diameter.
Result: Histologically the lesion showed a well circumscribed capsule with salivary gland lobules and with interspersed small lobules of mature adipose tissue. The lesion was well circumscribed and the lipomatous component is well structured. Histological examination of the residual gland did not show any particular alteration in term of fibrosis, inflammation and lobular atrophy.
Conclusion: The reported case allow us to extensively describe the histological aspect of a such rare lesion of the parotid gland. Moreover the histological feature we observed in our case could suggest us the possibility that sialolipoma histogenetically truly represent a benign lipoma with entrapped salivary gland elements rather than an hamartomatous lesion or a reactive process affecting all the salivary gland parenchyma. The pathologists should describe the morphological variation observed in cases similar ours, in order to support the histogenetic hypothesis and to correlate histogenesis with clinical behavior of the lesion itself.

ID 925
SCARY FEATURES OF PLEOMORPHIC ADENOMA

R. M. Di Crescenzo[1], G. Borriello [2], S. Ardone[1], D. Loffredo[1], S. Simonelli[1], F.Merolla[3], S. Staibano[1]

[1] Department of Advanced Biomedical Sciences, Pathology Unit, University of Naples Federico II; [2] Maxillofacial Surgery Operative Unit, Department of Neurosciences, Reproductive and Odontostomatological Sciences, University of Naples Federico II; [3] Department of Medicine and Health Sciences "V.Tiberio", University of Molise, Campobasso, Italy

Background: The most frequent tumor of the salivary gland is pleomorphic adenoma (PA). Pleomorphic Adenoma encompasses a wide range of histological appearances of this common tumor.
Aside from the well-known histological findings of PA (variable epithelial, myoepithelial, and stromal components in a mixed pattern)[1], this tumor might have unique characteristics that can be mistaken as indicative of malignancy[2].
We present a case of PA with unmistakable foci of tumor deposits in the blood arteries.
Case presentation: In October 2021, a 69-year-old female patient appeared with a swelling of the left parotid area. The lesion was found benign at FNC (category IV sec. Milan system). The patientunderwent a superficial parotidectomy. The tumor had all of the hallmarks of PA on histological evaluation. It was made up of a chondromyxoid stroma with regions of hypercellularity and a combination of epithelial and myoepithelial cells. A fibrous capsule of varied thickness around the tumor, with no indications of branching.
Neoplastic deposits were seen in vascular channels inside the tumor, which were validated by CD31 positivity.
Conclusions: PA has been observed to have vascular invasion in 9% of cases, particularly in small salivary glands.
This finding's clinical importance will be discussed.

References

1. El-Naggar AK, Chan JKC, Grandis JR, Takata T, S. P. WHO Classification of Head and Neck Tumours. WHO Classification of Tumours (2017), 4th Edition, Volume 9. (2017).
2. Hernandez-Prera, J. C. et al. Pleomorphic adenoma: the great mimicker of malignancy. Histopathology **79**, 279–290 (2021).

ID 926
MUCORMYCOSIS IN POST-COVID 19 PATIENTS

D. Del Guercio[1], R. M. Di Crescenzo[1], F. Calabria[2], A. Autore[1], F. Stumpo[1], F.Merolla[3], S. Staibano[1]

[1] Unit of Anatomic Pathology, Department of Advanced Biomedical Sciences, University of Naples Federico II; [2] Maxillofacial Surgery Operative Unit, Department of Neurosciences, Reproductive and Odontostomatological Sciences, University of Naples Federico II; [3] Department of Medicine and Health Sciences "V.Tiberio", University of Molise, Campobasso, Italy

Background: COVID-19 illness is caused by the coronavirus 2 that causes severe acute respiratory syndrome (SARS-CoV-2). It has a wide range of severity and is linked to a number ofbacterial and fungal superinfections, which can make the condition more difficult to treat.
The COVID-19 outbreak has recently resulted in a rise in Mucormycosis infections, mainly in India, where

over 28.000 cases have been documented. Only a few instances have been documented outside of India1.
Case presentation: We present a case of a 74-year-old male patient with a recent history of COVID-19 infection who presented with maxillary osteonecrosis and sinusitis in December 2022.
An inflammatory condition was suspected after a maxillofacial CT with contrast revealed aberrant soft tissue thickening in both maxillary sinuses, as well as obliteration of the left sinus and ethmoidal cells. FESS (functional endoscopic sinus surgery) was carried out. In both sinuses, histological examination revealed a significant lymphoplasmacytic infiltration, bone necrosis, and wide aseptate hyphae, which were also emphasized by Grocott's stain. After six months of follow-up, there was no sign of illness recurrence in the patient who had received postoperative antibiotic treatment.
Conclusions: Mucormycosis is a rare and lethal infection caused by the Mucormycetes mold family. Mucormycosis has become more common as a result of the current COVID-19 outbreak, particularly in India1. The diagnosis of this illness requires histology2. Early detection of indications of this illness in COVID-19 patients provides a more accurate histology diagnosis and more effective treatment.

References

1 .Danion, F. *et al.* Coronavirus Disease 2019-Associated Mucormycosis in France: A Rare but Deadly Complication. *Open Forum Infect. Dis.* **9**, (2022).

2. Dannaoui, E. Recent Developments in the Diagnosis of Mucormycosis. *J. fungi (Basel, Switzerland)* **8**, (2022).

ID 958
GHOST CELL ODONTOGENIC TUMORS: A RARE CASE REPORT

N. Scibetta[1], E.Unti[1]
[1] Pathology Unit,ARNAS Civico Palermo, Italy

Objectives: We report a case of Ghost Cell Odontogenic Carcinoma (**GCOC**) -less than 50 casesreported in literature- transformed from a dentinogenic ghost cell tumor (**DGCT**) -only few cases of trasformation reported.
Materials and methods: A 81-year-old woman underwent subtotal mandibulectomy with histological diagnosis of ameloblastoma on the right side. Twenty years after, a mass (cm 10x9x4.5)was removed from right hemi-maxilla of the same patient, with diagnosis of DGCT, that recurred five years later in the right orbito-maxillary region (cm 4.5x5.5). Two years later a right hemimaxilectomy rivealed a mass of cm 6 in her right zygomatic bone and the diagnosis was GCOC in DGCT.
Results: No histological material from the first surgery was available to us for verification (reported ameloblastoma). There was no difference between the second and the third mass examined by us(DGCT): dysplastic atubular dentine (dentinoid) forming lobulated deposits (eosinophilic, fibrillarand red with van Gieson's stain) in intimate relation with odontogenic epithelial islands, nests of epithelial cells that resembled ameloblastoma and areas of anucleate cells characterized by pale, eosinophilic cytoplasm (so called "ghost cells"), sometime calcified. Odontogenic and amelobastic-like cells were positive for keratins and negative for CEA, vimentin, S100; ghost cells were not stained with cytokeratins. Mitoses- and Ki67-indexes were low.
The fourth surgery (GCOC) showed pleomorphic epithelial cells characterized by atipical nuclei, high number of mitoses and necrotic foci, along with keratinization and some features of DGCT.
Conclusions: In our specimens, two mass revealed typical DGCT, that sometime recurs locally, without metastasis, even when it infiltrates the bone, like our case. The last recurrence showed cytologic criteria of malignancy indicative of GCOC, that can occur *de novo* or after DGCT orCalcifying Odontogenic Cyst (COC). Therefore, it is very important to examine the tumorsufficiently and to review anamnesis and former tissue when possible, differentiatingameloblastoma or ameloblastic carcinoma.

References

[1]G.Vijayakumar et al. Ghost cell odontogenic carcinoma of anterior mandible. J Oral Maxillofac Pathol doi 10.4103/jomfp.JOMFP_195_20

[2]J Kim et al. Odontogenic ghost cell carcinoma. Oral Surg Oral Med Oral Pathol Oral Radiol Endoddoi 10.1067/moe.2000.109016

PATOLOGIA TESSUTI MOLLI E DELL'OSSO

ID 786
RETROPERITONEAL LIPOSARCOMA WITH HIBERNOMA-LIKE FEATURES, REPORT OF A CASE

F.A. Tucci[1], G. Dagrada[2], A. Tosoni[3], A. Pellegrinelli[3], A. Gronchi[4], G. Pruneri[2], P. Collini [2,5], & M. Barisella[2]
[1] University of Milan, Milan, Italy; [2] Pathology Unit, IRCCS Istituto Nazionale dei Tumori, Milan, Italy; [3] Pathology Unit 2, L. Sacco Hospital, Milan, Italy; [4] Sarcoma Service, IRCCS Istituto Nazionale dei Tumori, Milan, Italy; [5] Soft Tissue and Bone Pathology and PediatricPathology Unit, IRCCS Istituto Nazionale dei Tumori, Milan, Italy

Objectives: Well-Differentiated Liposarcoma (WDLPS), in its most common lipoma-like subtype, closely resembles mature white adipose tissue. It is characterized by high-level amplification of the MDM2

gene. A diagnosis of WDLPS is relevant in retroperitoneum, implying multivisceralsurgery +/- chemoradiotherapy. Usually, a diagnostic needle biopsy is performed. Here we describe a case of LPS with multiple foci of hibernoma-like features. Hibernoma is a rare benign adipocytic retroperitoneal neoplasm featuring mature brown fat, with peculiar ultrastructural characteristics, usually bearing small deletions in MEN1 and AIP genes, without MDM2 amplifications.
Materials and methods: Immunohistochemistry and FISH analysis for MDM2, together with electron microscopy analyses were performed.
Results: A 67-year-old female presented with a 23-cm retroperitoneal mass. The biopsy revealed a WDLPS, with MDM2 amplification confirmed by FISH. The patient underwent neoadjuvant radiotherapy followed by en-bloc multivisceral resection. The mass showed residual viable tumor represented by WDLPS, lipoma-like, sclerosing, myxoid-like, and cellular types. In addition, a plurifocal component made of multivacuolated, granular lipoblasts with prevalent central round nuclei without nuclear indentations, resembling brown fat, occurred. FISH analysis on this component, performed after MDM2 immunohistochemistry, showed MDM2 amplification.
Conclusions: We presented a case of retroperitoneal LPS with hibernoma-like features. This is relevant to differential diagnosis in tru-cut needle biopsies performed before preoperative therapy and/or multivisceral resection. MDM2 amplification should be applied in all needle biopsies of retroperitoneal lipogenic tumors to avoid an erroneous diagnosis of benign lipogenic tumor.

ID 839
SCLEROSING MESENTERITIS, A CASE REPORT

E. Piombino[2], S.Poidomani[1], C. Colarossi[2], G. Magro[1], L. Memeo[2]

[1] Department of Medical and SurgicalSciences and Advanced Technologies, G.F. Ingrassia, University of Catania, Catania, Italy;[2] Pathology Unit, Department of Experimental Oncology, MediterraneanInstitute of Oncology, Catania, Italy

Objectives: Sclerosing mesenteritis (SM) is an idiopatic inflammatory fibrosing process of the mesenteric adipose tissue. It is associated with previus abdominal surgery and some neoplasms. The most common site of involvement is the small bowel mesentery.It could be a diagnostic pitfall, inparticular in patient with a diagnosis of genital tract neoplasia.
Materials and methods:We report a case of a 54-year-old female admitted in our institutionpresenting with a histologic diagnosis, on biopsy, of endometrial tromal sarcoma. A CT scan wasperformed, and underwent then a laparoscopic hysteroctomy. Two nodular and solid tumor of 2 and 7 cm pertaining to the omentum, were identified and were performed the intraoperative consultationon suspicion of metastases. After ruling out the metastatic nature of the lesions, a hysterectomy wasperformed and the two tumors removed.
Results: Histologically, the final diagnosis of uterine neoplasia was carcinosarcoma and the twonodule were composed of a proliferation of bland-looking spindle cells without nuclear atypia, haphazardly set in a myxoid stroma, partially sclerosing, which is an inusual morphology for thispathology; fat necrosis and infiammatory cells were also evident. Immunohistochemical analysesshowed a focal staining for desmin; smoothmascleactin, S100 protein,SOX-10, CD34, CD31, ERG, MDM2,pancytokeratins, and EMA were negative.
Conclusions: This case remark how important histology is for diagnosis of SM, in ct preoperativeradiological-based diagnosis could have led to considering the two mesenteric tumor metastatic and excluding the surgical lapproch to the uterine neoplasia. SM should be included in the differentialdiagnosis, in consideration of its benign clinical behavior and the different management, compared to the most common neoplasms arising in the same site.

UROPATOLOGIA

ID 747
DNA METHYLATHION ANALYSIS STRATIFIES THE RISK OF HIGH GRADE UROTHELIAL CARCINOMA IN URINARY SAMPLES

V. Fiorentino[1], P. Tralongo[1], E.D. Rossi[1], M.Martini[2], G. Fadda[2,] F. Pierconti[1]

[1] Fondazione Policlinico Universitario A. Gemelli IRCCS, Largo A. Gemelli 8, 00168, Rome, Italy; [2] Diagnostic Department of Anatomic Pathology, University of Messina, 98100, Messina, Italy

Objectives: We aimed to stratify the high risk for HGUC as indicated in Bladder EpiChek test in non-muscle invasive bladder carcinoma (NMIBC) patients during the follow-up, correlating the EpiScore with incidence of HGUC recurrences confirmed by clinical evidence or histological biopsies.
Materials and methods: 290 patients with a diagnosis of NMIBC were treated and followed for 1 year: the diagnosis was formulated according to the Paris System for Reporting Urinary Cytology (TPS)[1]. 175 cases received a cytological diagnosis of HGUC (High Grade Urothelial Carcinoma) or Suspicious for High Grade Carcinoma (SHGUC), 115 patients received a cytological diagnosis of negative for urothelial carcinoma (NHGUC) or atypical urothelial cells (AUC).
Results: Patients with a cytology consistent with HGUC and an Episcore ≥ 60 showed histological positivity in 72,6% of cases, while in 37,4% the his-

tology was negative. Patients with an EpiScore between 81 and 90 showed a percentage of histological positivity for HGUC double compared to those with an Episcore < 80, while an EpiScore > 90 received a diagnosis of HGUC in an amount four times higher than that seen with an EpiScore < 70. Moreover, all patients with an EpiScore > 90 tested positive at 6 months during follow-up, while no patients with an EpiScore between 60-69 showed a HGUC recurrence during follow-up.
Conclusions: We stratified the high risk for HGUC as indicated by Bladder EpiCheck test in NMIBC patients in follow-up. Moreover, we validated a combined approach of cytology and methylation test not only in terms of diagnosis of HGUC but also as a predictive method for clinical and histological recurrences.

References

1. Rosenthal DL, Wojcik EM, Kurtycz DFI, eds. The Paris System for Reporting Urinary Cytology. Cham, Switzerland: Springer International Publishing AG; 2016

ID 845
A PRELIMINARY STUDY INVESTIGATING THE IMPACT ON INTER-OBSERVER AGREEMENT OF DOUBLE STAIN "OCT4/CD34" FOR THE DETECTION OF LYMPHOVASCULAR INVASION IN GERM CELL TUMORS OF THE TESTIS

OCT4/CD34 in Germ Cell Tumors of the testis

Authors and M.E. Maracci[1], M. Sirolli[1], T. Franceschini[2], F. Ambrosi[2], M. Fiorentino[1,2], M. Colecchia[3], C. Ricci[2,4]

[1] Pathology Unit, IRCCS Azienda Ospedaliero-Universitaria di Bologna, Bologna; [2] Pathology Unit, Maggiore Hospital, AUSL Bologna, Bologna; [3] Department of Pathology, IRCCS San Raffaele Scientific Institute, Milano;[4] Department of Experimental, Diagnostic and Specialty Medicine (DIMES), University of Bologna, Bologna;

Objectives: Lymphovascular invasion (LVI) is a relevant prognostic factor in germ cell tumors of the testis (GCTT) but several studies showed discordant results and low inter-observer agreement(IrOA) with H&E, also among uropathologists (UP). We tested double stain (DS) for OCT4/CD34 in a cohort of GCTT and analyzed its impact on IrOA compared to H&E.
Materials and methods: Four authors [(2 uropathologists (UP) and 2 non-UP)] independently evaluated 34 consecutive and retrospectively enrolled cases of GCTT (29 seminomas, 3 embryonal carcinomas and 2 mixed GCTT) diagnosed between January 2019-April 2022 at our Institution. We assessed and compared the IrOA (Fleiss's Kappa/FK and intraclass correlation coefficient/ICC) with both H&E and OCT4/CD34 for all four authors and in the subgroups of UP and non-UP.
Results: For the four authors, the IrOA was superimposable with H&E (KF = 0.269; ICC = 0.602) and OCT4/CD34 (KF = 0.361; ICC = 0.698), and it ranges from fair to moderate. Also, in the subgroup of UP, the IrOA was superimposable with H&E (KF = 0.433; ICC = 0.628) and OCT4/CD34 (KF = 0.462; ICC = 0.638), and it is moderate. By contrast, in the subgroup of non-UP, the IrOA was improved by OCT4/CD34 (KF = 0.304; ICC = 0.479) compared to H&E (KF = 0.197; ICC = 0.330), moving from slight/poor to fair.
Conclusions: Our study showed that the IrOA was superior for UP rather than non-UP for both the stain, so confirming that the LVI is a histological parameter that requires dedicated experience. Besides, OCT4/CD34 improved the IrOA for non-UP but not for UP and if they are matched together. Future studies are needed to investigate the potential impact of OCT4/CD34 on the detection of LVI in GCTT if applied to a routine scenario.

ID 914
MICROSATELLITE INSTABILITY IN PEDIATRIC TESTICULAR GERM CELL TUMORS: ANALYSIS THROUGH IHC, PCR AND NGS

G. Zannini[1], M. Montella[1], I. Tedesco[1], G. Savarese[2], R. Sirica[2], M. E. Errico[3], V. Donofrio[3], F. Pagliuca[1], F. Zito Marino[1], R. Franco[1]

[1] Pathology Unit, Department of Mental and Physical Health and Preventive Medicine, University of Campania "L. Vanvitelli", 80138 Naples, Italy; [2] AMES, Centro Polidiagnostico Strumentale srl, 80013 Casalnuovo, Naples, Italy; [3] Pathology Unit Department of Pathology, Santobono-Pausilipon Children's Hospital, Via Posillipo, 80123 Naples, Italy

Objectives: Testicular germ cell tumors (TGCT) are the most common solid malignancies in young men. Innovative therapies for TGCT patients are currently needed. Immunotherapy represents a successful therapeutic approach for the treatment of different tumors carrying the deficit of mismatch repair (MMR) proteins and the microsatellite instability (MSI)[1]. To date, no studies have been performed regard the potential role of MSI in pediatric TGCTs. The main goal was to determine the frequency and the clinical impact of MSI in this neoplasm.
Materials and methods: A series of 17 pediatric TGCTs was examined through IHC, PCR and NGS analysis. IHC for MLH-1, PMS-2, MSH-2 and MSH-6 was evaluated as follows: proficient MMR (pMMR) with all MMR proteins positive, and deficient MMR (dMMR) with the loss of one heterodimer. Cases with instability in at least two markers by PCR were MSI-high (MSI-H) and with instability in one marker, MSI-low (MSI-L). Cases without instability were evaluated as microsatellite-stable (MSS). All cases were analyzed by NGS for confirmation of microsatellite status.
Results: Only 1 out of 17 cases was dMMR by IHC,

showing the loss of MLH1/PMS2 heterodimer. However, PCR and NGS results didn't corroborate with dMMR status. All cases pMMR confirmed MSS status by PCR and NGS.
Conclusions: Our findings suggest that MSI is not a frequent event in pediatric TGCT.Unfortunately, MSI could not have a predictive role for the treatment with immune checkpoint inhibitors. MMR-IHC could be used as a screening test for MSI detection and only cases with loss MMR-IHC could be needed additional molecular tests.

References

1) Zito Marino F, Amato M, Ronchi A, et al. Microsatellite Status Detection in Gastrointestinal Cancers: PCR/NGS Is Mandatory in Negative/Patchy MMR Immunohistochemistry. Cancers (Basel). 2022 Apr 28;14(9):2204. https://doi.org/10.3390/cancers14092204

ID 948
EXPRESSION AND SIGNIFICANCE OF PD-L1 IN SEMINOMA

A. Bordonali[1], T. Ientile[1], G. De Lisi[1], A.M.Florena[1], A.G. Giannone[1]

[1] *Anatomic Pathology, Department of Promotion of Health and Mother and Child Care, Internal Medicine and Medical Specialties, University of Palermo, Italy*

Tumor microenvironment is a specialized biosystem of cells, which, influencing angiogenesis, migration, drug resistance and immunological tolerance, have effects on initiation, development and tumor progression. The components of the tumor microenvironment assume a diagnostic, prognostic, predictive or therapeutic role. Testicular seminomas are abundantly infiltrated by immune cells, mainly represented by CD3+ cells and memory T cells. The immune system is involved in the biology of seminomas and immune checkpoints may represent promising targets for immune therapy. Testicular tissue is an "immunologically privileged" site where immune responses are naturally suppressed by PD-1/PD-L1 interaction. PD-L1 could assume the role of treatment target, especially in seminomas refractory to conventional chemotherapy.
Objectives: Our purpose was to identify associations between PD-L1 expression and characteristics of the seminoma, attributing to PD-L1 expression a possible prognostic and/or predictive role.
Materials and methods: We examined 11 cases of seminomas admitted to our Hospital from March 2020 to December 2021. The hematoxylin-eosin stained slides were recovered to re-evaluate the characteristics of seminomas and the immune infiltrate. The immunohistochemical stains were performed using the antiPD-L1 monoclonal antibody. PD-L1 expression was assessed using the Combined Positive Score (CPS).
Results: pT1 cases had a mean CPS value lower than the average CPS of all the cases examined and lower than the mean CPS value of the cases with pT2 stage. Cases with necrosis showed an average CPS value higher than the average CPS value of the cases without necrosis. Cases with Ki67 < 50% had a mean CPS value lower than the mean value of the cases with Ki67 > 50%.
Conclusions: Despite the study was unable to find strong correlation between CPS and prognostic characteristics due to the paucity of the sample, it still suggested the existence of an association between the values of CPS and the characteristics related to the aggressiveness of the disease (proliferation index, necrosis, pT stage), making it necessary further investigations in this direction.

SIAPeC Case Report Award 2022

Breast Edition

SIAPEC - IAP

AI HELPS DETECT INVASIVE BREAST CARCINOMA: A CHALLENGING CASE

G. Mallel[1], M. Vecsler[1], A. Nudelman[2], J. Sandbank[1,2]

[1] Ibex Medical Analytics, Tel Aviv, Israel; [2] Institute of Pathology, Maccabi Healthcare Services, Rehovot, Israel

Artificial intelligence (AI) is quickly becoming a key factor in support of diagnosis and quality control in pathology. Its role is especially impactful in virtually eliminating misdiagnosis, thus allowing the best predictive ability of the histological examination. This is particularly true for breast biopsies where the tissue material is often relatively abundant and fragmented and complex lesions are frequent.
In October 2021, a 30-year-old woman underwent an MRI guided left breast biopsy of the upper quadrant due to suspicious findings at imaging. She was then diagnosed with ductal carcinoma *in situ* (DCIS). Immunohistochemistry (IHC) for p63 supported the diagnosis. The neoplastic cells were negative for estrogen receptor (ER) and progesterone receptor (PR). As the imaging was very suggestive of an invasive malignancy, a second biopsy was performed in November 2021. This time, multiple foci of high grade solid DCIS with comedo necrosis and extensive lobular cancerization were observed. Microinvasion could be neither ruled out nor confirmed. Again IHC for p63 supported the diagnosis and the neoplastic cells were negative for ER and PR.

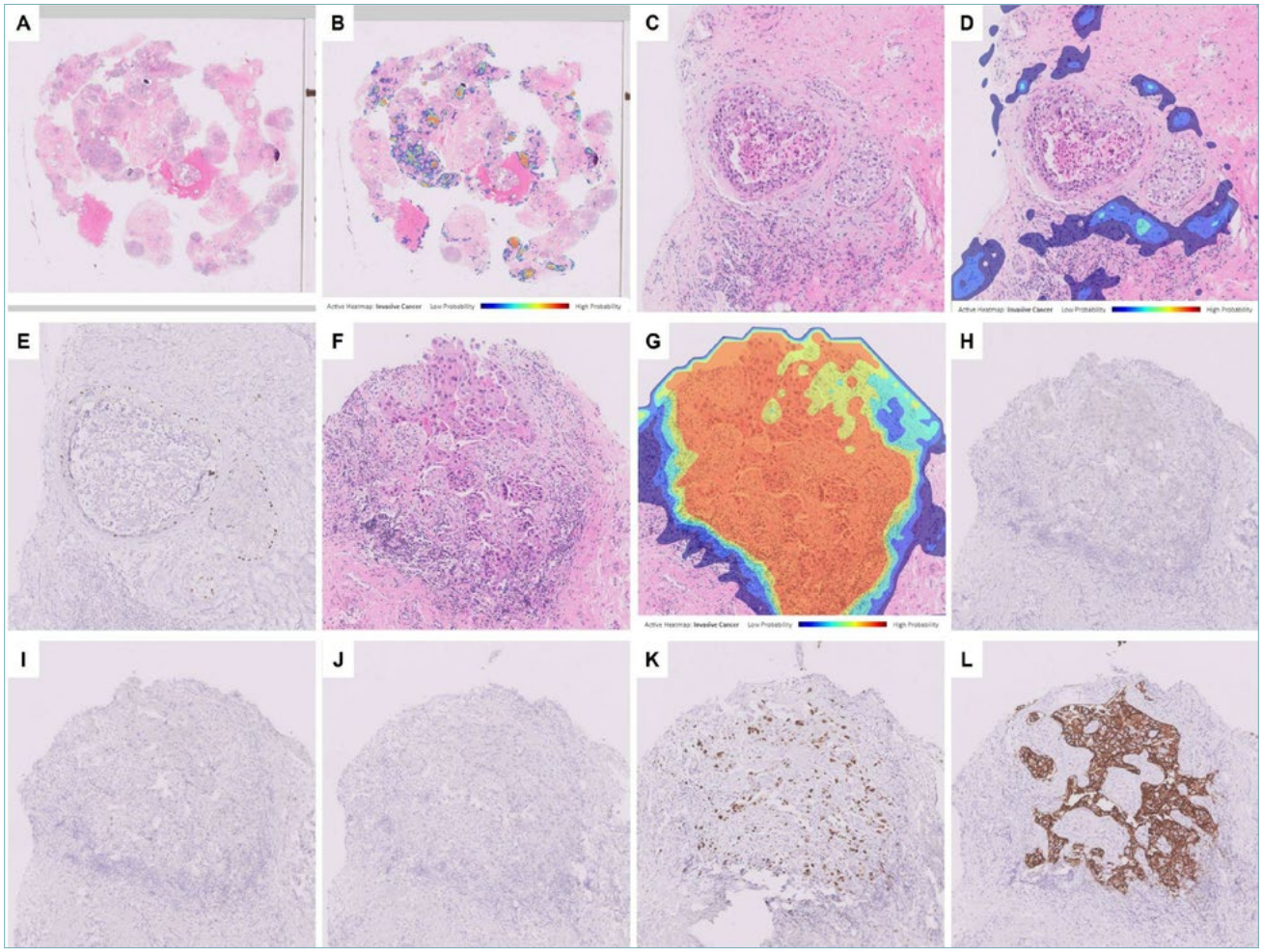


Figure 1. (A-B) Low magnification overview; in B the "invasive cancer" heatmap* is shown (magnification 4X). (C-E) DCIS area; in D the "invasive cancer" heatmap* is shown; the area displays p63 positive basal cells (E) (magnification 100X). (F-L) Invasive carcinoma area; in G the "invasive cancer" heatmap is shown; the area is devoid of basal cells as highlighted by a negative p63 stain (H); the neoplastic cells are negative for ER (I) and PR (J), they have a proliferative index of 50% as shown by Ki67 stain (K) and are scored as 3+ in the HER2 stain (L) (magnification 100X). *"Invasive cancer" heatmap: low probability is displayed in blue, high probability is displayed in red.

Within the framework of AI-based quality control routinely performed in our lab, the whole slide images of the histological slides were analyzed by the Galen™ Second Read Breast application (Ibex Medical Analytics). The application raised an alert for "invasive cancer" and highlighted the areas of maximum likelihood for it to be found. Upon review, the regions were devoid of basal cells (p63 negative) and their morphology was highly suggestive of invasion. Thus the diagnosis was revised and the presence of multiple foci of *bona fide* poorly differentiated invasive breast carcinoma with apocrine features was indeed confirmed. Importantly, testing of the prognostic factors on the invasive areas showed that the neoplastic cells were negative for ER and PR but had a proliferative index (Ki67) of 50%. Furthermore, Her-2 staining was scored as 3+ (Fig. 1).
Thus the neoplasia could be correctly classified as belonging to the Her-2 enriched subtype. These findings allowed for the proper therapeutic strategy to be pursued.
The present case shows how AI based systems can play a crucial role in the management of breast samples, especially in morphologically challenging cases.

"LUMINAL A - TRIPLE NEGATIVE" BREAST CANCER

S.M. Rossi, G. Sabarese, D. Righi, G. Perrone
Pathology Dept, Policlinico Universitario Campus Biomedico di Roma

BACKGROUND

Triple-negative breast carcinomas (TNBCs) are defined by the absence of hormone receptors (ER, PGR) and HER2 overexpression, and account for 10-17% of all breast cancers [1]. They are usually Basal-like intrinsic molecular subtype and associated with poor outcomes. TNBCs are not eligible to breast cancer genomic testing because of their negativity for ER and PGR. Here we present two cases of invasive lobular TNBCs (TNBC-ILCs) with discordant result at Prosigna/PAM50 test.

CASE RESENTATION

Case 1: A 61-year-old female with a 2 cm ILC, G3, node negative. ER (Dako Clone EP1) and PGR (Dako Clone PgR 1294) were negative, HER2 (HercepTest Dako) absent (score test 0) and proliferating fraction (Ki67) 10%. Prosigna/PAM50 test results Luminal A (ROR score 27 and a 4% probability of distant recurrence).
Case 2: A 76-year-old female patient presented with a 2,1 cm ILC, G3, node negative. ER (Dako Clone EP1) and PGR (Dako Clone PgR 1294) were negative, HER2 (4B5 Ventana) absent (score test 0) and proliferating fraction (Ki67) 5%. Prosigna/PAM50 test results Luminal A (ROR score 23 and a 4% probability of distant recurrence). Based on molecular results, we perform in both cases AR immunostaining that show strongly positivity (> 90%) providing evidence that AR could be the driver of the luminal phenotype observed in these two cases of TNBC-ILCs [2].

CONCLUSIONS

The prognosis of TNBC was significantly impacted by the intrinsic molecular subtype's classification because luminal BC seemed to have a very low risk of distant recurrence. Even if breast cancer genomic test are not currently refundable in TNBCs, here we show the key role of PAM50 test in AR positive lobular histotype BCs in order to define the exact molecular intrinsic subtype for therapeutic and clinical approach.

References

[1] Bergeron A, MacGrogan G, Bertaut A, et al. Triple-negative breast lobular carcinoma: a luminal androgen receptor carcinoma with specific ESRRA mutations. Mod Pathol 2021;34:1282-1296. https://doi.org/10.1038/s41379-021-00742-9.

[2] Conforti F, Pala L, Pagan E, et al. Biological and clinical features of triple negative Invasive Lobular Carcinomas of the breast. Clinical outcome and actionable molecular alterations. Breast 2021;59:94-101. https://doi.org/10.1016/j.breast.2021.06.011

HER2 HETEROGENEITY IN BREAST CANCER: A CASE REPORT

R. Accarino[1], S. Varricchio[1], L. Stasio[1], A. Caltavituro[2], P. De Placido[2], L. Insabato[1], D. Russo[1]

[1] *Pathology Unit, Department of Advanced Biomedical Sciences, University of Naples "Federico II",Naples, Italy;*
[2] *Department of Clinical Medicine and Surgery, University of Naples "Federico II",Naples, Italy*

BACKGROUND

The human epidermal growth factor receptor 2 (HER2) is an oncogene that stimulates cancer cell growth and division associated with increased disease recurrence and poor prognosis.
According to current guidelines, HER2 status is routinely assessed by immunohistochemistry (IHC) to stratify breast cancers (BCs) in positive (score 2+ with fluorescence in situ hybridization/FISH confirmation, or score 3+), negative (score 0), and in recently so-called "HER2-low" ones (score 1+ or 2+ with negative FISH). Unfortunately, many cases show a heterogeneous expression of HER2 in which HER2-directed therapies have been ineffective.

CASE PRESENTATION

In November 2021 a 39 years-old woman with multiple lesions in the right breast and lymph nodes involvement was referred to Ospedale del Mare of Naples. At diagnostic core needle biopsy (CNB), histology revealed an invasive high grade No Special Type (NST) BC HER2 3+ with reported areas of HER2 0. Since the advanced stage of the disease, histology and age, it was decided to start therapy with paclitaxel, trastuzumab and pertuzumab. However the patient underwent clinical disease progression, so another CNB was made and sent to our department: the diagnosis of NST BC with HER2 heterogeneity was confirmed, showing areas with score 2+ and with score 1+. Hence, the patient was considered eligible for therapy with Trastuzumab-Deruxtecan (T-DXd), with an appreciable clinical reduction of the tumoral masses after just one month.

DISCUSSION

HER2 is overexpressed in ~20-25% of BCs mainly as a result of gene amplification [1]. However, there are cases with intratumoral heterogeneity in which not all neoplastic cells overexpress HER2. Recently, clinical trials [2-3] have shown favorable results for T-DXd in BCs with heterogeneous HER2 expression because of its bystander effect with death of HER2 positive cells and also of neighboring HER2-negative cells.
In this case, we identified a heterogeneity pattern of HER2 expression which allowed starting therapy with T-DXd with good clinical results.

CONCLUSION

HER2 IHC evaluation is an important predictive marker of therapeutic response, also for patients without evident HER2 positivity. It must be kept in mind that HER2 heterogeneity might be present in some BCs that can benefit from new therapies with a better prognosis.

References

1 King CR, Kraus MH, Aaronson SA. Amplification of a novel v-erbB-related gene in a human mammary carcinoma. Science 1985;229:974-6. https://doi.org/10.1126/science.2992089

2 Horisawa N, Adachi Y, Takatsuka D, et al. The frequency of low HER2 expression in breast cancer and a comparison of prognosis between patients with HER2-low and HER2-negative breast cancer by HR status. Breast Cancer 2022;29:234-241. https://doi.org/10.1007/s12282-021-01303-3

3 Mosele MF, Lusque A, Dieras V, et al. Unraveling the mechanism of action and resistance to trastuzumab deruxtecan (T-DXd): biomarker analyses from patients from DAISY trial. Annals of Oncology 2022;33:S123-S147. https://doi.org/10.1016/j.annonc.2022.03.277

KEEPING AN EYE OUT FOR THE UNPREDICTABLE: ESTROGEN RECEPTOR CONVERSION AFTER ADJUVANT TREATMENT

C. Rossi[1], A. Vanoli[1], E. Ferraris[2], L. Pozzi[3], M. Paulli[1], M. Lucioni[1]

[1] Unit of Anatomic Pathology, Department of Molecular Medicine, University of Pavia, and IRCCS San Matteo Hospital Foundation, Pavia, Italy; [2] Unit of Medical Oncology, IRCCS San matteo Hospital Foundation, Pavia, Italy; [3] Unit of Endoscopy, IRCCS San Matteo Hospital Foundation, Pavia, Italy

Hormone receptor (HR) conversion in breast cancer is a well-known phenomenon in the metastastic setting. Here we describe the case of a woman with an unusual presentation of a triple negative metastasis of a HR-positive primary breast cancer.

A 55-year-old woman presented to our Oncology department with obstructive jaundice and a radiological suspicion of advanced cholangiocarcinoma. She had a history of invasive lobular carcinoma treated at another hospital when she was 36-year-old, with a late recurrence with bone metastases in 2017, treated with Fulvestrant. Laparoscopic exploration showed peritoneal carcinomatosis and a nodule on the serosal surface of the gastric lesser curvature. Gastroscopy with sampling of the gastric mucosa reported findings compatible with gastric linitis plastica. At histology, the gastric mucosa showed infiltration by a discohesive neoplasia, with medium-sized cells with a round nucleus, inconspicuous nucleoli and large eosinophilic cytoplasm, growing as single cells or files, with marked desmoplastic response. The neoplastic cells were positive for broad-spectrum keratins only, and negative for estrogen receptor (ER) and other lineage markers, both breast- and non-breast-specific. The results were inconclusive in assessing whether the neoplasia should be classified as a primary gastric or recurrent breast cancer. Immunostaining against GATA3 was performed and showed a strong nuclear positivity in the neoplastic cells. Breast primitivity was suggested and the patient started therapy accordingly. Unfortunately, the patient progressed and died one year later from her disease.

Albeit late metastases are a well-known occurrence in the natural history of lobular carcinoma of the breast, and may occur at unusual sites, triple-negative lobular carcinoma is rare. Fulvestrant is a selective estrogen receptor

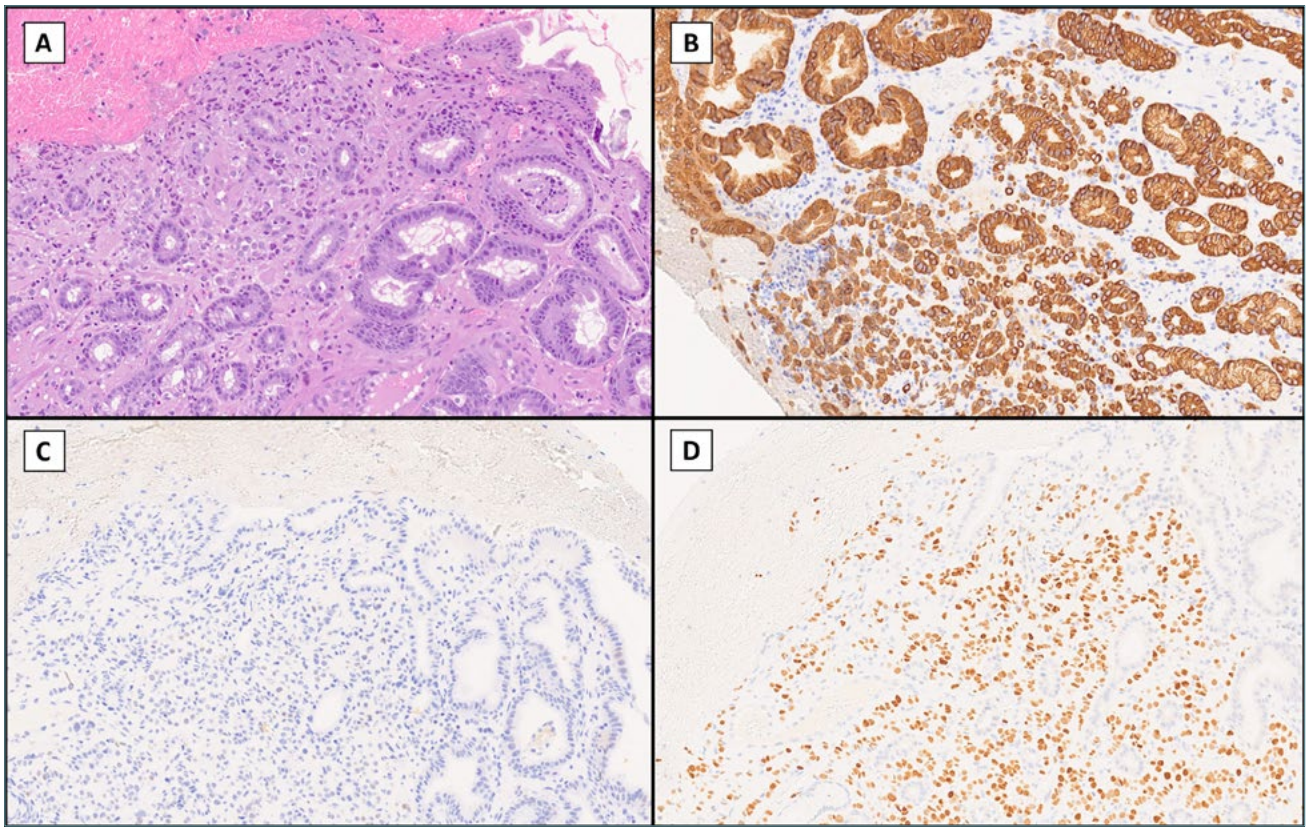


Figure 1. Histology of the gastric lesion stained with hematoxylin and eosin (A, 10x), immunohistochemistry for broad-spectrum keratins (B, 10x), estrogen receptor (C, 10x) and GATA3 (D, 10x).

degrader that specifically targets ERα by inducing its polyubiquitination-mediated degradation, and it has been demonstrated to reduce the IHC expression of ER. The introduction of novel target drugs causing a reduction in the expression of ER may complicate the diagnosis in similar instances, and careful review of the history of the patient, with particular attention to the pharmacological history, may help render a final diagnosis is such cases.

(N)TRACKING THE CLINICAL AND PATHOLOGIC FEATURES OF AN ANOMALOUS CASE OF SECRETORY BREAST CARCINOMA

A. Ascione[1], C. Chiappetta[2], S. Scagnoli[1], B. Cerbelli[1]

[1] Department of Radiological, Oncological and Pathological Sciences, Sapienza University of Rome;[2] Department of Translational and Precision Medicine; Sapienza University of Rome

Secretory carcinoma (SC) is a very rare type of invasive breast carcinoma (IBC) composed of cells with cytoplasmic vacuoles and extracellular secretions. Its hallmark genetic alteration is a translocation causing ETV6-NTRK3 gene fusion and production of a chimeric tyrosine kinase with transforming activity. Most SCs are triple-negative with low proliferation index (Ki-67). Among IBCs, SC carries a favourable prognosis [1].
We report the case of a 60-year-old woman seeking medical attention for a lump in the lower-inner quadrant of her left breast. Mammography and ultrasonography confirmed the presence of a mass, which was diagnosed on core biopsy as IBC of no special type (NST) with cribriform morphology. Oestrogen receptor (ER) was positive in 35% of cells, progesteron receptor (PR) was negative and HER2 was not overexpressed; Ki-67 was 30%. Pre-operative staging was negative. At surgery, the tumour measured 3 cm on gross examination, and was classified as SC with microcystic growth pattern. ER was weakly positive in 5% of cells (ER-low), PR and HER2 were negative, Ki-67 was 23%. The patient received adjuvant chemotherapy with administration of epirubicin and cyclophosphamide followed by paclitaxel and subsequent radiotherapy and hormonal therapy with anastrazole. Two years later, a CT scan identified two lung metastases, which were treated with metastasectomy and confirmed to be SC (Fig. 1A). ER was positive in 25% of cells (Fig. 1B), PR and HER2 were negative, Ki-67 was 30%. Real Time PCR successfully detected NTRK3 fusion (Fig. 1C) and, in accordance with the current guidelines, therapy with entrectinib, an NTRK inhibitor, was initiated.
This case reflects how challenging it can be to deal with rare entities, especially when the presentation is atypical. In our case, SC showed an unusual “luminal B-like” phenotype with positive ER, high proliferation index and an aggressive clinical behaviour, with early lung metastases. A correct histological diagnosis is fundamental considering the therapeutic option in the recurrent/metastatic setting of SC with one of the TRK inhibitors, which have been shown to result in profound and long-lasting responses [1]. In addition, the Pathologist is in charge of confirming the presence of NTRK rearrangements using appropriate and validated molecular techniques.

Reference

[1] Pareja F, Weigelt B, Reis-Filho JS. Problematic breast tumors reassessed in light of novel molecular data. Mod Pathol 2021;34(Suppl 1):38-47. https://doi.org/10.1038/s41379-020-00693-7

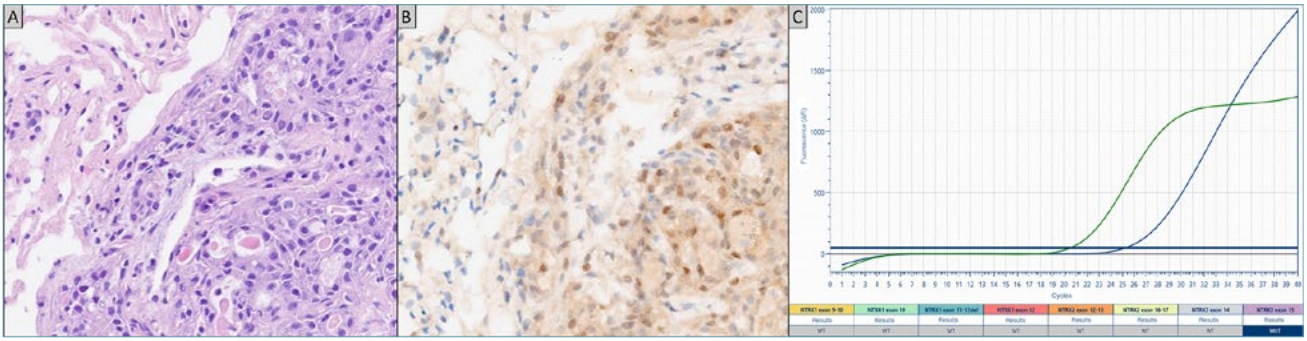


Figure 1. (A). Secretory breast carcinoma, metastatic to the lung. The tumour presents a prevalently microcystic growth pattern, with evidence of extracellular secretions. The tumour cells have eosinophilic, granular cytoplasm and mild to moderate nuclear atypia (H&E; 20X). (B). Immunohistochemistry shows focal nuclear stain for ER (20X). (C). Qualitative detection by One-Step Real Time-PCR of NTRK3 exon 15 fusion in RNA isolated from tumour tissue. The blue and green sigmoidal fluorescence curves represent the amplification reactions of the sample and of the endogenous control respectively.

UN UNCOMMON AGNOSTIC BREAST CARCINOMA. THE UTILITY OF ETV6-NTRK3 GENE FUSION FROM DIAGNOSIS TO TREATMENT

G. Mastrosimini

Department of Diagnostic and Public Health, Section of Pathology, University Hospital Trust of Verona, Verona, Italy; Ospedale Sacro Cuore Don Calabria, Negrar, Verona, Italy.

A 57-year-old woman presented with a left breast nodule. Sonographic examination revealed a 7 mm ill-defined hypoechoic lesion. Core needle biopsy was performed and the microscopic examination showed a proliferation of relatively uniform small glands, with focal microcystic growth pattern and intraluminal eosinophilic material (Alcian-PAS positive). The cells had pink-to-clear often vacuolated cytoplasm, nuclei with mild atypia (Fig. 1A), and displayed negative immunostaining for CK5 and weak positive expression of estrogen receptor (ER). Pathological diagnosis was "atypical microglandular adenosis" (B3 category), despite S100 protein expression was weak and patchy (Fig. 1B). Differential diagnosis between benign and malignant lesion containing secretory features was challenging considering age, morphology, and immunohistochemical characteristics.
The subsequent excisional biopsy showed a tubular-microcystic proliferation with intraluminal secretions lined by cells with mild to moderate nuclear pleomorphism and low mitotic activity. The periphery of the tumor was characterized by infiltrative margins and microcystic *in situ* component (Fig. 1C). The tumor cells showed weak expression of ER, negative immunostaining for progesterone receptor and HER2, and Ki-67 < 5%. The expres-

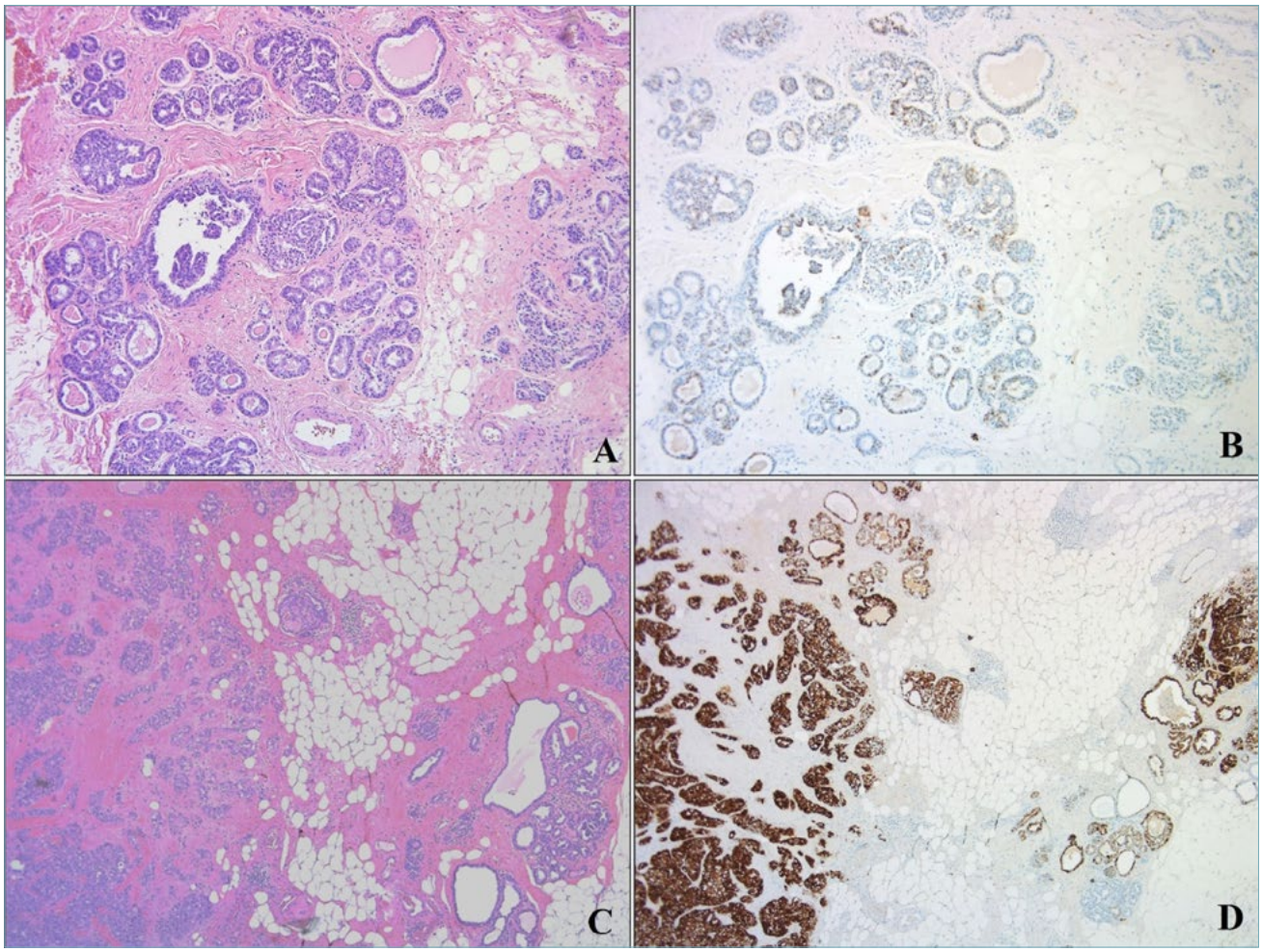


Figure 1. Core biopsy: (A) H&E 10x; (B) S100, 10x. Surgical specimen. (C) H&E 4x; (D) S100, 4x.

sion of S100 protein was strong and diffuse in the invasive component but patchy in the *in situ* component (Fig. 1D). The latter was the only component present in the core needle biopsy. The diagnosis of Secretory Breast Carcinoma (SBC) was supported by the detection of ETV6-NTRK3 gene fusion by next generation sequencing. This gene fusion is due to a balance translocation t(12;15) [1] and is pathognomonic of SBC and mammary analogue secretory carcinoma of salivary gland, whereas it rarely concerns other solid tumors. NTRK alteration represents an agnostic tumor marker, after microsatellite instability, for the target therapy of any solid tumor. Although patients with SBC usually present a local disease and are surgically managed, TRK inhibitors are approved by the FDA for patients with metastatic or unresectable disease [2].

References

1 Li L, Wu N, Li F, et al. Clinicopathologic and molecular characteristics of 44 patients with pure secretory breast carcinoma. Cancer Biol Med 2019;16:139-146.

2 Cima L, Kaya H, Marchiò C, et al. Triple-negative breast carcinomas of low malignant potential: review on diagnostic criteria and differential diagnoses Virchows Arch 2022;480:109-126.

NOT ALL THAT GLITTERS IS GOLD – HER2 IN A CASE OF METAPLASTIC BREAST CARCINOMA WITH HETEROLOGOUS MESENCHYMAL DIFFERENTIATION

C. Taverna

Pathology Unit, ASL AL, P.O. Novi Ligure, Italy

Summary

Human epidermal growth factor receptor 2 (HER2) is an important prognostic and predictive tool in the treatment of breast carcinoma, as the positive (HER2 3+) or the equivocal stain (HER2 2+) associated to amplification of the *HER2* gene using in situ hybridisation techniques (ISH), on one side is related to more aggressive neoplasms, while on the other one is a predictor of anti-HER2 therapies response, with good results.
Metaplastic breast carcinomas (MTBC) comprehend a wide spectrum of rare neoplasms, with different histological subtypes and variable prognosis and clinical course. Most of them are very aggressive diseases, usually with a triple negative molecular phenotype (TBNC) and high proliferation index. For these reasons, although surgery is widely used as first line treatment, some patients undergo neoadjuvant chemotherapy (NACT), with general poor response.
We present an exceeding rare case of metaplastic breast carcinoma with heterologous mesenchymal differentiation (chondromyxoid), diagnosed as no special type (NST) breast carcinoma on core needle biopsy. Ki-67 index was high (60% of neoplastic cells) and HER2 was assessed as equivocal (2+) on immunohistochemical (IHC) stain, with *HER2* being amplified on FISH, so the patient underwent NACT associated to anti-HER2 drugs, with almost no response.
This challenging case suggests that, despite HER2 status is a strong and well know predictive biomarker in breast carcinomas, recognising the correct histotype is mandatory, especially in preoperatory setting, as despite of the routinary predictive factors, it can drastically change the management of the patient.

INTRODUCTION

The human epidermal growth factor receptor 2 (HER2) is a tyrosine kinase receptor which belong to the Human Epidermal Receptor family. It is a well study prognostic and predictive factor, with a dual role, as it is usually expressed in clinically aggressive tumours and it is related to poor prognosis, but, on the other hand, it is targetable by anti HER2 therapies (trastuzumab, pertuzumab, lapatinib, tucatinib, and neratinib) in adjuvant, neoadjuvant and metastatic settings [1].
The overall proportion of breast carcinomas with *HER2* amplification is supposed to be around 15% [2], most of them being no special type breast carcinomas (NST). Regarding special types of carcinomas, HER2 can be found amplified in a small proportion of pleomorphic lobular carcinoma, mucinous carcinoma and carcinoma with apocrine differentiation.
In some recent studies, the theory that non only molecular expression, but also levels of HER2 protein can influence the amount of response to target therapy is proposed [3-4]. Indeed, some authors detect a higher amount

of pathological complete response in HER2 3+ cases (55-70%) compared to HER2 2+, amplified tumors (17-20%). The latter results are quite comparable to those found in breast cancers that do not express HER2 (15-25%), so in the last few years an increasing use of anti-HER2 antibody-drug conjugates in these neoplasms is seen [3-4]. Consequently, anti-HER2 therapies are often proposed in combination with chemotherapy in cases of high-grade neoplasms, even if HER2 status is not 3+.
Metaplastic breast carcinomas were first described by Huvos and colleagues in 1973 [5] and account for less than 5% of all breast tumors [6]. They comprehend a wide spectrum of lesions, characterized by the total or partial replacement of the typical glandular component of breast carcinomas with other cellular types, as squamous and sarcomatoid, including heterologous elements. The 5th edition of World Health Organisation (WHO) classification of breast tumors divided metaplastic breast carcinomas in low-grade adenosquamous carcinoma, fibromatosis-like, pure squamous cell carcinoma, pure spindle cell carcinoma, metaplastic carcinoma with heterologous mesenchymal differentiation and mixed metaplastic carcinoma [7]. Most of them are triple negative neoplasms, with variable proliferation index depending on the subtypes. Usually, the first line treatment is surgical excision, followed by radiotherapy and/or systemic therapy based on the final pathological report and the type of surgery.
We present an exceeding rare case of metaplastic carcinoma with heterologous mesenchymal differentiation (chondromyxoid), misdiagnosed in the preoperative core needle biopsy and reported as NST breast carcinoma. For the high proliferation index and HER2 equivocal score on IHC, with FISH being positive for *HER2* amplification, patient underwent chemotherapy associated to anti-HER2 drugs. The tumor demonstrated poor clinical, radiological and pathological response, at it was recognised as metaplastic carcinoma only after the systemic treatments.
We conclude that even though HER2 is a good predictive marker of response to target therapy, it is mandatory to correlate the prognostic and predictive markers to the tumor histotype to optimise the patient management.

CASE REPORT

In August 2021, a 43 years-old woman, without previous clinical history, came to our attention after eight months of NACT associated to trastuzumab for a 25 mm lesion in the upper-inner quadrant of the breast, with clinical and radiological positive axillary lymph nodes. The diagnosis was made in another hospital and the pathological report described a high grade, NST invasive carcinoma. The immunohistochemistry showed negative stains for estrogenic and progesterone receptors (ER and PR), the Ki-67 index was highly (60% of neoplastic cells) and HER2 showed weak to moderate, complete membranous stain, in 70% of neoplastic cells (2+). FISH test demonstrated *HER2* amplification.
After chemotherapy, imaging analyses revelated the persistence of the disease both in the breast and in the lymph nodes and the patient underwent surgery, consisting in quadrantectomy (for the favourable ratio between tumor size and residual breast tissue) followed by axillary dissection.
Adjuvant treatment consisted in adjuvant radiotherapy both on breast and on axillary region and anti-HER2 drugs. The patient is free of disease at the time of writing.

PATHOLOGICAL AND IMMUNOISTOCHEMICAL FEATURES

Macroscopically the lesion had enhanced consistency, was firm and with a myxoid appearance, 22x20 mm wide. Fifteen lymph nodes were isolated from the axillary adipose tissue.
Microscopically the tumour consisted in a mixture of mesenchymal and epithelioid elements, strictly intermixed, with moderate to high grade pleomorphism, arranged in sheets, lobules and nodules, extensively infiltrating in breast parenchyma. The mesenchymal component was composed by ovoidal to spindle cells, with eosinophilic cytoplasm and moderate to high grade atypia, arranged in small nests, chords, with sparse single elements, in a myxoid and chondroid background. The epithelioid cells had large size, abundant cytoplasm, prominent nucleoli and high-grade atypia. Mitotic count in the epithelioid component was high, with mitosis up to 27 in 0,65 mm field diameter. The lesion was extensively sampled, for a correct evaluation of the amount of residual tumor and only scattered areas of oedema, fibrosis and fibrosclerosis were found (*Figure1*).
Immunohistochemistry was performed, confirming the dual cell population, with metaplastic component showing positivity to S100 and Cytokeratin K903 (34BE12), while epithelioid elements staining focally positive to CK AE1/AE3, and CAM 5.2 (Fig. 1). Regarding the breast prognostic and predictive factors, estrogen (ER) and progesteron (PR) receptors were not expressed in the nuclei of neoplastic cells. Ki-67 was present in about 60% of cancerous elements. HER2 presented a complete, weak to moderate membranous stain, so score 2+ was assess according to the current ASCO/CAP guidelines, and FISH was performed, showing *HER2* amplifi-

cation. The same molecular phenotype was found on the initial core biopsy.
According to the 5th edition of WHO classification of breast tumors, the diagnosis of metaplastic breast carcinoma with heterologous mesenchymal differentiation (chondroid and myxoid) was made. Nottingham Bloom Richardson grading system was used in the assessment of histological grade, and tumor had score 9 (tumor tubule formation 3; tumor pleomorphism 3; mitotic count 3). Lymphatic and vascular invasion were present, while infiltration of perineural spaces was not found. Excisional margins were microscopically negative. Out of 15 lymph nodes isolated from the adipose tissue of the axillary region, 2 were positive for macro metastasis and 1 for micro metastasis. The case was studied by two breast pathologists.
Tumoral response to therapy was evaluated using two methods. Pinder score, which evaluates the ratio between the primary tumour bed and the residual one in breast tissue, and the presence of residual metastasis associate to the possible presence of chemotherapy effects, demonstrated the absence of significative tumoral regression in breast tissue and the presence of residual disease, in the absence of neoadjuvant therapy effects in lymph nodes. MD Anderson Residual Cancer Burden (RBC) assess different variables of both the primary tumours and lymph nodes (extension of the primary bed area, cancer cellularity, percentage of *in situ* disease in the breast tissue, numbers of positive lymph nodes and maximum diameter of lymph node metastasis). Our case presented a score III, which means the presence of extensive residual tumoral component.

DISCUSSION

The case herein described presents different challenging aspects. Indeed, in clinical practice HER2 positivity is one of the best predictive factors for target therapy, as high Ki-67 index predict response to NACT. Here, despite the tumour presents both *HER2* amplification and a high Ki-67 index, the histological subtype, correctly diagnosed in the post-NACT pathological report, demonstrates that other factors have an important role in predict the amount of pathological response as well.
For the high Ki-67 index, associated to the absence of ER and PR expression, patients underwent NACT. This

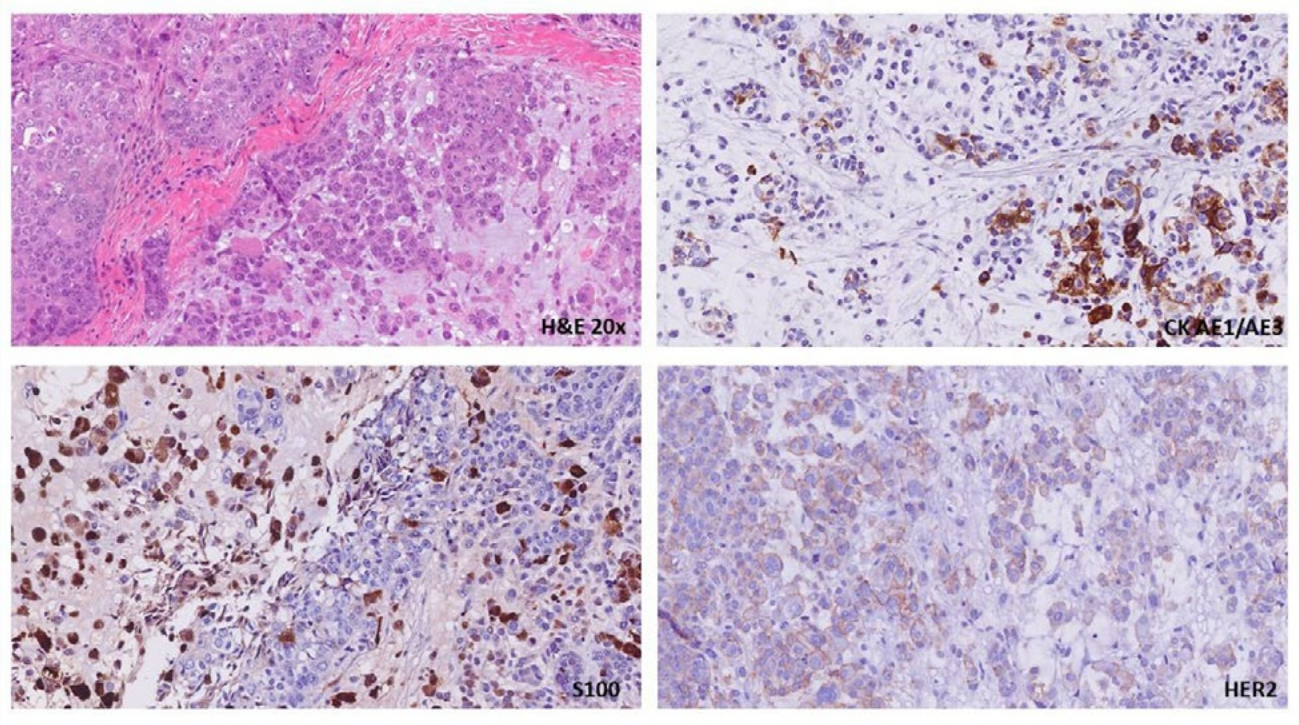


Figure 1. The tumor shows two cells types, with a mesenchymal, ovoidal to spindle cell component, dispersed in a chondroid and myxoid background, mixed to epithelioid, basaloid elements. Cells are arranged in nests, chords and dischoesive single elements with moderate to high grade cytological atypia. Epithelioid population demonstrates scattered positive stain to CK AE1/AE3, while metaplastic cells strong react to S100. HER2 shows a complete, weak to moderate, membranous expression, with equivocal result (2+).

decision is commonly made for TNBC, as in most cases patients experience significant amount of pathological response. Regarding metaplastic breast carcinomas, however, results are poor as, although occasionally complete pathological response (pCR) is described in literature [8-10], most of the studies reveals low rates of pCR, ranging from 10% to 17% compared to about 75% in TNBC [8]. Moreover, some cases of disease progression during NACT are reported, ranging from 27% to 62,5% of cases [8,12,14], leading to a delay of the surgical excision of the neoplasm and worse survival rates.
Our case was treated with anti-HER2 drugs, for the equivocal IHC score and *HER* amplified FISH results. HER2 is not usually used as a strong predictive factor in the neoadjuvant treatment of MTBC, with only occasion cases described [8-13]. Results are limited, as Wong [8] and colleagues describe only 2 cases of *HER2* amplified MTBC which did not respond to target treatment and Han [13] reports that in his cohort out of 5 patients experiencing pCR, only one of them has *HER2* amplified.
Moreover, there is some evidence that the tumor subtype of MTBC can represent a predictive factor of good pathological response to therapy more than the immunophenotype. Indeed, some cases of matrix-producing MTBC show good pathological response, as one case reported by Wong [8] and 3 out of 5 cases in Han study [13], while other authors describe good results with target therapies in sarcomatous (ifosfamide and anthracycline), epidermoid (platinum based) and squamous cell (EGFR target-based therapy) MTBC [14].
Our case demonstrates that, even though HER2 is one of the most useful, reproducible and simple predictive markers of response to neoadjuvant systemic treatment in breast carcinomas, it has always to be related to the histological type of carcinoma, which can also widely influence the response to treatments.

References

1 Hayes DF. HER2 and Breast Cancer - A Phenomenal Success Story. N Engl J Med 2019;381:1284-1286.

2 Sapino A, Goia M, Recupero D, et al. Current Challenges for HER2 Testing in Diagnostic Pathology: State of the Art and Controversial Issues. Front Oncol 2013;3:129.

3 Krystel-Whittemore M, Xu J, Brogi E, et al. Pathologic complete response rate according to HER2 detection methods in HER2-positive breast cancer treated with neoadjuvant systemic therapy. Breast Cancer Res Treat 2019;177:61-6.

4 Katayama A, Miligy IM, Shiino S, et al. Predictors of pathological complete response to neoadjuvant treatment and changes to post neoadjuvant HER2 status in HER2-positive invasive breast cancer. Mod Pathol. 2021;34:1271-81.

5 Huvos AG, Lucas JC Jr, Foote FW Jr. Metaplastic breast carcinoma. Rare form of mammary cancer. N Y State J Med 1973;73:1078-82.

6 Salimoğlu S, Sert İ, Emiroğlu M, et al. Metaplastic Breast Carcinoma: Analysis of Clinical and Pathologic Characteristics – A Case Series. J Breast Health 2016;12:63-66.

7 Reis-Filho J, Gobbi H, McCart Reed A, et al. Metaplastic Breast Cancer. Board WCoTE, editor. Breast Cancer. Lyon, France: IARC Press 2019, pp. 135-138.

8 Wong W, Brogi E, Reis-Filho et al. Poor response to neoadjuvant chemotherapy in metaplastic breast carcinoma. NPJ Breast Cancer 2021;7:96.

9 Takuwa H, Ueno T, Ishiguro H, et al. A case of metaplastic breast cancer that showed a good response to platinum based preopera tive chemotherapy. Breast Cancer 2014;21:504-50710.

10 Schroeder MC, Rastogi P, Geyer CE, et al. Early and Locally Advanced Metaplastic Breast Cancer: presentation and Survival by Receptor Status in Surveillance, Epidemiology, and End Results (SEER) 2010–2014. Oncologist 2018;23:481-488.

11 Chen IC, Lin CH, Huang CS, et al. Lack of efficacy to systemic che- motherapy for treatment of metaplastic carcinoma of the breast in the modern era. Breast Cancer Res Treat 2011;130:345-351.

12 Esbah O, Turkoz FP, Turker I, et al. Metaplastic breast carcinoma: case series and review of the literature. Asian Pac J Cancer Prev 2012;13:4645-4649.

13 Han M, Salamat A, Zhu L, et al. Metaplastic breast carcinoma: a clinical-pathologic study of 97 cases with subset analysis of response to neoadjuvant chemotherapy. Mod Pathol 2019;32:807-816.

14 Aydiner A, Sen F, Tambas M, et al. Metaplastic Breast Carcinoma Versus Triple-Negative Breast Cancer: Survival and Response to Treatment. Medicine (Baltimore). 2015;94:e2341.

A TRICKY BREAST BIOPSY: ALWAYS LOOK BEYOND YOUR NOSE

A. Pernazza[1], B. Cerbelli[1], A. Pisano[2], A. Botticelli[2], A. Cirillo[2], G. d'Amati[2]

[1] Department of Medico-Surgical Sciences and Biotechnology, Sapienza University of Rome; [2] Department of Radiological, Oncological and Pathological Sciences, Sapienza, University of Rome

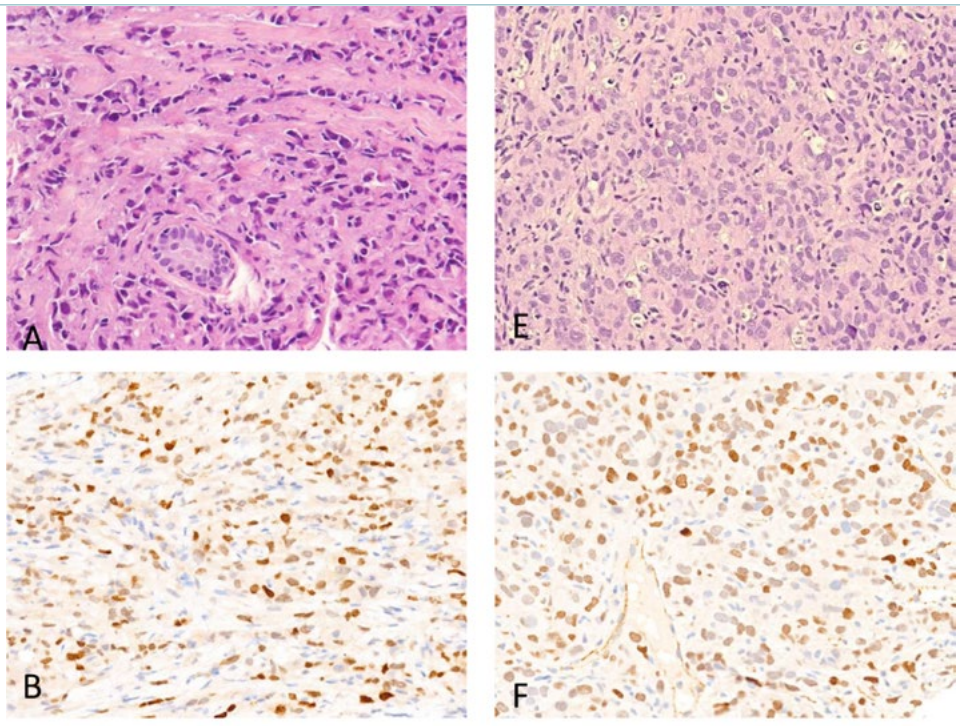
Thoracic SMARCA4-deficient undifferentiated tumors (SMARCA4-UT) are rare and aggressive neoplasms, occurring mostly in the mediastinum of male smokers (1). They are characterized by an inactivating mutation of *SMARCA4,* resulting in loss of expression of brahma-related gene 1 (BRG1) [1,2]. Most patients have metastatic disease at diagnosis, with a median survival of six months [2]. We report the case of an 80-year-old female smoker seeking medical attention for a lump in her right breast.

Mammography and ultrasonography confirmed the presence of the mass, which was diagnosed on core biopsy as an invasive pleomorphic lobular (E-cadherin -/+) breast carcinoma, triple negative, with a proliferation index (Ki-67) of 80%. Staging with CT scan revealed a large mediastinal solid mass and additional lesions ranging from 3 to 6 cm in the left lung and right scapula. The patient underwent an additional biopsy of the scapular mass. Histology revealed a malignant lesion made up by loosely arranged pleomorphic cells, resembling the tumor in her breast (Fig. 1).

However, neoplastic cells showed a more marked degree of pleomorphism with some rhabdoid features, which, in light of CT findings, raised the suspicion of a primary thoracic tumor.

Immunohistochemical stains on both the scapular and breast lesion revealed overlapping findings. Both tumors were positive for CD30 SALL-4, synaptophysin, CKAE1/AE3 (weak and focal), CD138 (focal) and negative for TTF-1, p63, CK5/6, desmin, NUT, S100, WT-1, p40, CD3, CD20, CD45 (Fig. 1). The proliferative index was 90%.

Given the clinical presentation of an aggressive, metastatic tumor, together with the focal rhabdoid morphology and the immunophenotype, a SMARCA4-UT was considered. Accordingly, immunohistochemistry showed loss of BRG1 expression with an intact positive internal control; INI-1 expression was preserved. Based on this immunoprofile the final diagnosis was SMARCA4-UT with multiple metastatic lesions, including the breast. This is the first reported case of SMARCA4-UT metastatic to the breast, highlighting the potential diagnostic pitfalls in the differential diagnosis between primary and metastatic breast lesions on biopsy and the crucial role of clinical-pathological correlations.



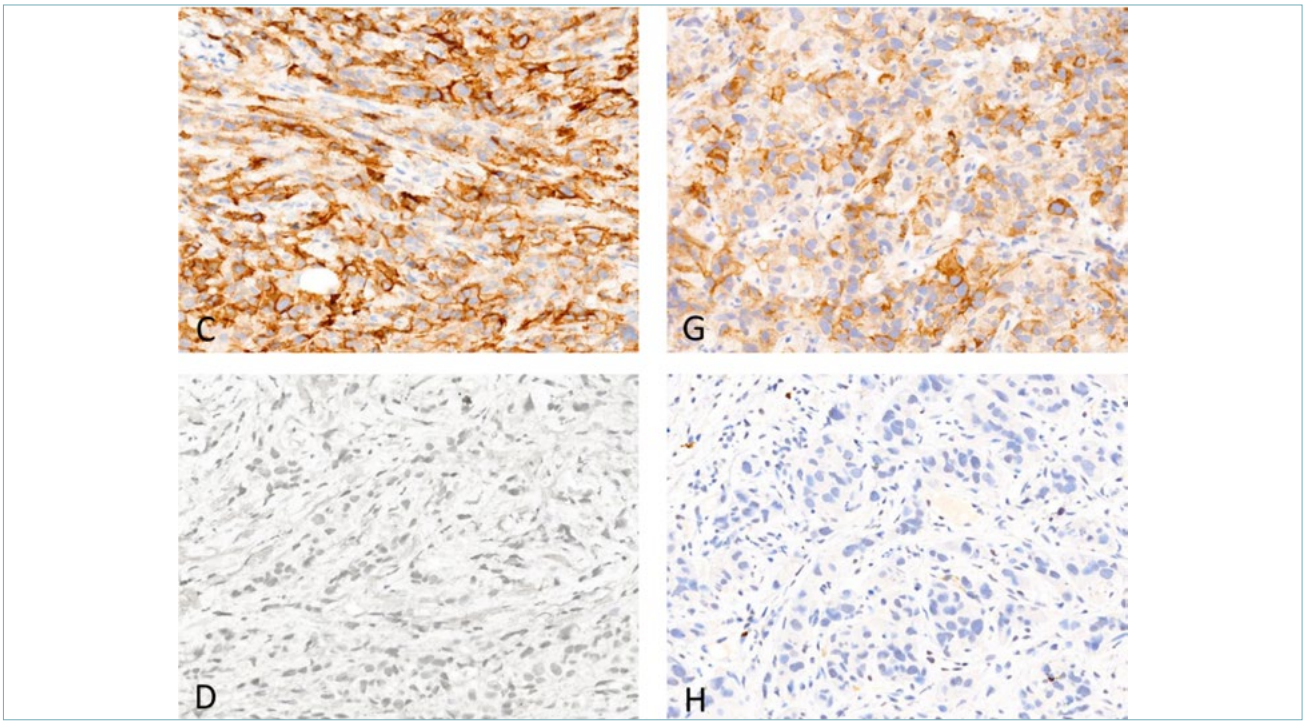


Figure 1. (A-D): breast lesion; (E-H): scapular mass (A): breast biopsy showed neoplastic lesion made up by high grade loosely cohesive epithelioid cells (hematoxylin ad eosin, original magnification). (E): scapular mass biopsy revealed a malignant lesion composed by loosely arranged pleomorphic cells with some rhabdoid features (hematoxylin ad eosin, original magnification X20). Immunohistochemistry showed a positivity of neoplastic cells of both lesions for SALL 4 (B, F), CD30 (C, G) and negativity for BRG1 (D, H), with internal positive control staining in inflammatory and stromal cells.

References

[1] Nambirajan A, Jain D. Recent updates in thoracic SMARCA4-deficient undifferentiated tumor. Semin Diagn Pathol 2021;38:83-89. https://doi.org/10.1053/j.semdp.2021.06.001

[2] Chatzopoulos K, Boland JM. Update on genetically defined lung neoplasms: NUT carcinoma and thoracic SMARCA4-deficient undifferentiated tumors. Review Virchows Arch 2021;478:21-30. https://doi.org/10.1007/s00428-020-03011-3