DRUG-INDUCED LIVER FAILURE SECONDARY TO PSORALEA CORYLIFOLIA: A CASE REPORT AND LITERATURE REVIEW

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Background: We present a case of a 56-year-old male presenting with acute liver failure requiring transplantation after intake of Psoralea corylifolia for five weeks. Acute liver injury secondary to herbal and dietary supplements (HDS) should be suspected in patients presenting with acute liver failure. Awareness of HDS's associated risk is important in the setting of their increasing use, with at least 23% of adults in the United States reporting using alternative therapies. One such implicated remedy is the plant Psoralea corylifolia, used in Ayurvedic medicine as an alternative treatment for conditions including psoriasis and vitiligo.

Aims and methods: A literature review using Ovid-Medline was undertaken to identify similar cases. Other potential causes were excluded clinically, radiologically and serologically to establish causality. Sections of the recipient liver were formalin-fixed and paraffin-embedded for staining with haematoxylin and eosin and other routine special stains.

Results and Conclusions: Sections of the liver showed extensive necrosis varying from submassive to massive. No other cause for the liver injury was identified on clinical correlation. Thus histological findings were compatible with a clinical history of Drug Induced Liver Injury.

This report highlights a case of Psoralea corylifolia-induced acute liver failure requiring transplantation, compared to seven other cases in the literature, with one reported subsequent death. The popularity of Psoralea corylifolia as an Ayurvedic remedy highlights the clinical significance of its potential hepatotoxicity. A clinician should have a high index of suspicion for HDS use with patients presenting with acute liver failure.

ENDOMETRIAL CARCINOMA SENTINEL LYMPH NODE ULTRASTAGING: BALANCING COST AND EFFICACY TO DEVELOP AN OPTIMAL PROTOCOL

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Background: Sentinel lymph node (SLN) sampling is a relatively novel, increasingly used method of staging endometrial carcinomas, but a standardised accepted methodology does not exist. After introduction of SLN sampling at our institution, we developed an in-house protocol for SLN examination, which specifies three “up-front” serial haematoxylin and eosin (H&E) levels with an accompanying broad-spectrum cytokeratin (MNF116).

Aims: We have audited Mater’s up-front approach, and compared it to the published MD Anderson Cancer Center (MDACC) and the Memorial Sloan Kettering Cancer Center (MSKCC) step-wise, algorithmic protocols, to compare cost-efficiency and efficacy.

Methods: Reports from all endometrial carcinoma cases examined over the last twenty-nine months were reviewed to identify positive (metastases and isolated tumour cells; ITCs) SLNs. The corresponding slides were re-examined to determine which level the malignant cells were detected. This data was then used to simulate the MDACC and the MSKCC protocols.

Direct evaluation of the sensitivity of each protocol was considered to be outside of the scope of this retrospective study.

Results and Conclusions: One-hundred-and-eighty-nine cases evaluated detected eight cases with metastases, discovered on the first H&E level. Twelve of the thirteen cases with only ITCs were discovered on the first MNF116 level, with the thirteenth on the second MNF116 level. The third paired level failed to detect further positive nodes in any case.

We show the Mater protocol is comparable to the MDACC and MSKCC protocols in terms of efficacy. The step-wise algorithms reduce cost, but potentially result in prolonged turnaround times; whereas the Mater protocol generates more slides up-front to reduce turnaround time, but results in a slightly higher average cost of per case.

MATURE TERATOMA IN AN UNDESCENDED TESTIS

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Background: Cryptorchidism occurs in 2-5% of boys born at term and is the most common congenital urogenital abnormality. Germ cell tumours are more likely to occur in cryptorchid testes, however pre-pubertal teratomas are unrelated to germ cell neoplasia in situ and are one of the most common tumours in the testes of boys. In the literature, there are only approximately 30 cases of mature teratoma occuring in a cryptorchid testis. In our case, laparoscopic orchipexy was performed on a 12-month-old boy with a left undescended testis.

Aims: To exclude malignancy in the undescended testis.

Methods: Photos were taken of macroscopic features as well as routine H&E staining.

Results &Conclusions: We describe a mature teratoma in an undescended testis, with mature cartilage, bone, and fibrous tissue as the predominant components. Negative staining for AFP, bHCG and PLAP excluded other common germ cell differentials. Prepubertal teratomas are benign, however given limited experience in the cryptorchid setting, continued observation is recommended.

PULMONARY NODULAR LYMPHOID HYPERPLASIA PRESENTING AS MULTIFOCAL LUNG OPACITIES

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Background: Pulmonary nodular lymphoid hyperplasia (PNLH) is a rarely encountered reactive lymphoproliferative lesion [1]. Clinically PNLH presents as a solitary area and in some cases multiple nodules with an important differential diagnosis of the lesion including extranodal marginal zone B-cell lymphomas of mucosa associated lymphoid tissue (MALT) type [2].

Aims: We present the case of a 47-year-old female who was found to have multifocal pulmonary nodular opacities on imaging. Initial core biopsy samples raised a differential diagnosis of MALT lymphoma or inflammatory myofibroblastic tumour (IMFT). Further biopsies were suggested for full evaluation of the lesion and the patient proceeded to wedge resection of the areas of concern. We aim to compare and contrast the core biopsy and wedge resection findings to highlight the salient histological and immunohistochemical features of PNLH.

Methods: A review of the histological features and immunophenotype of PNLH and the suggested differential diagnoses will be presented along with literature relevant to the case discussion.

Results & Conclusions: Follow up wedge biopsy of the lesions revealed a polymorphous interstitial lymphoid population with the architectural features and immunophenotype of PNLH. The findings of this case confirm that whilst PNLH is a rare entity it remains an important differential diagnosis in lung processes of lymphoid origin.

References:

1. Yell M, Rosado F. Pulmonary nodular lymphoid hyperplasia. *Arch Pathol Lab Med* 2019:143:1149-1153
2. Abbondanzo SL, Rush W, Bijwaard KE, Koss MN. Nodular lymphoid hyperplasia of the lung: a clinicopathologic study of 14 cases. *Am J Surg Pathol.* 2000;24(4):587–597

PINEAL REGION GERMINOMA – A CASE REPORT

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Intracranial germinomas are rare primary CNS tumours of children and young adults. They resemble seminomas from testes and dysgerminomas from ovaries. Histologically, germinomas exhibit characteristic two cell populations consisting of large neoplastic cells and small lymphoid cells, occasionally with associated granulomatous inflammation.

We present a case of germinoma of pineal region diagnosed intraoperatively on frozen section and smears. A 22-year-old male presented with a history of headache and visual disturbance for a month. MRI showed a 1.8cm mass in the pineal region with obstructive hydrocephalus. The patient underwent endoscopic third ventriculostomy (ETV). Cerebral spinal fluid (CSF) biochemistry showed no increase of Alpha-Fetoprotein (AFP) and Human chorionic gonadotropin (HCG). Six months post initial presentation, an MRI showed the mass increased to 2.8cm in size.

The patient underwent a biopsy of the pineal tumour. Intraoperative assessment (smears and frozen sections) of small biopsy fragments revealed mixture of small lymphoid cells, granulomatous aggregates and occasional larger atypical epithelioid cells with vesicular chromatin and prominent nucleoli. These atypical cells were best appreciated on the smears. The permanent sections and immunostains confirmed the intraoperative diagnosis of germinoma with the atypical cells displaying strong positivity for OCT3/4 and CD117.

Tumours of the pineal region represent a surgical challenge due to difficulties with surgical access and achieving complete resections. This case highlights the utility of cytology smears when dealing with minute amounts of tissue, and is an illustrative example of the diagnostic clue granulomatous inflammation serves as a clue to this diagnosis in this region. Because germinomas are sensitive to radiation therapy, unnecessary and potentially harmful radical resection can be avoided with correct intraoperative diagnosis.

A SOMATOTROPH PITUITARY ADENOMA, WITH SIGNET-RING LIKE CELLS. A CASE REPORT AND REVIEW OF THE LITERATURE

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Background: Pituitary adenomas are neoplasms affecting the adenohypophysis and are derived from either early progenitor or fully differentiated hormone secreting cells. Histologically signet-ring like cells in pituitary adenomas are considered a normal morphological variant. This is an extremely rare entity, with 4 cases reported in the literature to date.

Aims: We report a case of a 36-year-old man with symptoms of acromegaly and MRI findings of a 6mm pituitary adenoma.

Methods: A transphenoidal resection of the pituitary tumour was undertaken and submitted for light microscopic, immunohistochemical and electron microscopic assessment

Results &Conclusions: On histology the adenoma consisted of a monotonous population of neuroendocrine cells diffusely positive for synaptophysin, cytokeratin AE1-3 and Pit-1, with patchy positive growth hormone staining. There were admixed signet-ring like cells in which the cytoplasmic vacuoles are positive for synaptophysin and PAS- diastase and negative for the CD68 macrophages stain.
This case describes the rare morphological signet-ring like cell variant in pituitary adenomas as well as highlighting differences between this benign neoplasm and metastatic signet ring cell carcinoma.

Reference:
Trabzonlu L, Trabzonlu TA, Girbuz Y et al. ACTH-Cell Pituitary Adenoma With Signet Ring Cells: A Rare Case Report and Review of The Literature. *Appl Immunohistochem Mol Morphol.* 2018;28(2):e13-e16

ECTOPIC LIVER TISSUE IN THE GALLBLADDER WALL

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Background: Ectopic liver tissue is a rare finding that has been documented at multiple different sites in the body including the gallbladder [1]. The ectopic tissue, having no hepatic connection, is usually incidentally detected at the time of surgery or histological assessment [2]. Evidence suggests in some cases there is an increased risk of malignant progression [3].

Aims: We present the case of a 61-year-old man with a macroscopically identified intact nodule on the surface of the gallbladder after a routine laparoscopic cholecystectomy. The patient had a clinical history of micronodular cirrhosis of the liver secondary to non-alcoholic steatohepatitis (NASH).

Methods: A brief outline of the histological findings and immunophenotype of the ectopic liver tissue will be presented, with a review of literature relevant to the case discussion.

Results & Conclusions: The nodule was entirely submitted for histological assessment and showed liver parenchyma with features of micronodular cirrhosis. Ectopic liver tissue despite being a rare and often incidental finding can have important clinical implications including the potential risk of neoplastic transformation.

References

1. Zonca P, Martinek L, Ihnat P, Fleege J. Ectopic liver: Different manifestations, one solution. *World J Gastroenterol* 2013;19(38):6485-6489
2. Burke E, Harkins P, Arih I, O'Donoghue G. A case of ectopic liver tissue adherent to the gallbladder. *Journal of Surgical Case Reports* 2018;6:1-3
3. Arakawa M, Kimura Y, Sakata K, Kubo Y, Fukushima T, Okuda K. Propensity of ectopic liver to hepatocarcinogenesis: case reports and a review of the literature. *Hepatology* 1999;29:57-61

A CASE OF SQUAMOUS CELL CARCINOMA ARISING IN POROKERATOSIS OF MIBELLI

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Background: Porokeratosis is an uncommon disorder of epidermal keratinization. Characteristic features include hyperkeratotic lesions with an atrophic centre and raised wall-like border, and the pathognomonic histological finding of a coronoid lamella.[1] Malignant transformation into squamous, bowenoid or basal cell carcinoma rarely has been reported in all major variants.[2]

Aims: To describe a case of squamous cell carcinoma arising in porokeratosis of Mibelli

Methods: A 73-year-old man presented for routine skin malignancy surveillance with a new, asymptomatic lesion within a porokeratosis of Mibelli on his right calf, clinically suspicious for squamous cell carcinoma. His history included multiple non-melanoma skin cancers and one previous melanoma in situ. No other lesions typical for porokeratosis were found on examination. Histopathological examination of the lesion confirmed moderately differentiated squamous cell carcinoma arising in background changes of porokeratosis. The lesion was completely excised within a week of biopsy.

Results & Conclusions: Porokeratosis is a premalignant condition, however malignant transformation is rare[1] It is hypothesized that chromosomal instability and reduced immune surveillance with overexpression of p53 in contribute to the development of cutaneous malignancy within porokeratoses.[2] Risk factors for malignancy include ionizing radiation, sun exposure, immunosuppression, long history of the lesion and large size of the porokeratosis.[3]
This case adds to the existing literature surrounding malignant transformation of porokeratoses and highlights the importance of regular skin surveillance in affected patients.

References:
1. Sertznig P, von Felbert V, Megahed M. Porokeratosis: present concepts. J Eur Acad Dermatol Venereol. 2012;26(4):404
2. Sasson M, Krain AD Porokeratosis and Malignancy. Dermatol Surg. 1996 Apr; 22(4):339-42.
3. Weidner T, Illing T, Miguel D. Elsner P. Treatment of porokeratosis: a systemic review. Am J Clin Dermatol. 2017;18:435-49

CONSTITUTIONAL MISMATCH REPAIR DEFICIENCY IS A CHILDHOOD CANCER PREDISPOSITION SYNDROME ASSOCIATED WITH AN INCREASED RISK OF HIGH GRADE PAEDIATRIC BRAIN TUMOURS: REPORT OF 3 CASES AND REVIEW OF THE LITERATURE

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Background: Constitutional mismatch repair deficiency syndrome (CMMRD) represents a biallelic (homozygous or compound heterozygous) mutation in one of the mismatch repair genes (MLH1, MSH2, MSH6 or PMS2), resulting in a recessively inherited childhood cancer predisposition syndrome. A broad range of tumours in infants and children have been described in the CMMRD spectrum, most commonly brain tumours, intestinal tumours, and haematological malignancies. This contrasts with the monoallelic mismatch repair defects (MMR) of Lynch Syndrome, in which there is an increased risk of development of malignancies in particular involving the gastrointestinal and genitourinary tract, with onset in adulthood. Phenotypic features of CMMRD overlap with Neurofibromatosis, with café-au-lait macules a common finding in this cohort.

Aims: To highlight an increasingly recognized childhood cancer predisposition syndrome, and the importance of identification of these cases given significant implications for family counselling. There is an increased cancer risk in siblings, as well as for the parents, who may not yet be aware of their own associated monoallelic MMR defect (Lynch Syndrome).

Methods: We present 3 cases of childhood primary brain tumours that were subsequently confirmed in the setting of CMMRD. The histological, immunohistochemical and genetic findings of these cases are discussed, with focus on diagnostic pathologic features that suggest an underlying CMMRD.

Results & Conclusions: Three high grade brain tumours described include two high grade gliomas (a glioblastoma multiforme and a high grade diffuse intrinsic pontine glioma, H3K27M negative), and a medulloblastoma. These examples all showed complete absence of PMS2 staining on immunohistochemistry in both tumour cells and normal "internal control" tissues of the patient, correlating with biallelic loss of gene function. Review of the literature shows brain tumours as the most frequent presentation in children with CMMRD, with an increased risk of gastrointestinal tumours (including colonic carcinoma) and haematological malignancies also reported.

KI67/BCL2 INDEX IN INVASIVE BREAST CARCINOMA(NST)

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Background: B-cell lymphoma-2(BCL2) is an anti-apoptotic protein that has an anti-proliferative effect and is a favourable prognostic marker in breast carcinoma. Ki67/BCL2 ratio is emerging as a superior prognostic marker than either marker alone.

Aim: This study investigated the association of Ki67/Bcl2 index with hormonal receptor status and other clinicopathological parameters in premenopausal and postmenopausal primary invasive breast carcinomas(NST)

Method: Clinicopathological data were obtained from 63(42%) premenopausal and 87(58%) postmenopausal carcinomas from a prospective study. Immunohistochemical assessment of ER, PR(Allred score), HER2(ASCO/CAP 2013), Ki67 and BCL2 in the tumours were performed using routine protocols. BCL2 and Ki67 scores were obtained based on the percentage of positively stained cells (0=0-10%, 1=11-32% and 2=>33%). BCL2 staining score was subtracted from the Ki67 score to produce five categories (−2, −1, 0, 1, 2) and further categorized into two groups, low-index (-2, -1 & 0) and high-index (1 and 2). Association of Ki67/Bcl2 index with, tumor grade, tumor size, T & N stage, nodal status, node ratio (<0.15), ER, PR and HER2 status and molecular subtype, were evaluated using the Chi-square test(SPSS-20) Associations were considered significant when $p \leq 0.05$.

Results & conclusion: High Ki67/BCL2 index was significantly associated with negative ER (premenopausal $\chi2=4.701_{(1)}$; $p=0.030$: postmenopausal $\chi2=23.104_{(1)}$; $p=<0.001$) and negative PR (Premenopausal $\chi2=12.106_{(1)}$; $p=0.001$: Postmenopausal $\chi2=12.956_{(1)}$; $p=<0.001$) expression in both groups. It was also significantly associated with premenopausal grade III tumours ($\chi2=8.168_{(2)}$; $p=0.017$) and postmenopausal triple negative breast cancers ($\chi2=19.510_{(4)}$; $p=0.001$). It is evident that high Ki67/BCL2 index, indicating a high proliferation rate is associated with poor prognosticators in breast carcinoma and is suggestive of a poor clinical outcome.

EPITHELIAL-MESENCHYMAL PLASTICITY AND LYMPH NODE METASTASES IN INVASIVE BREAST CARCINOMA OF NO SPECIAL TYPE

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Background: Breast cancer is the commonest cancer among women with distant metastases being the leading cause of mortality. Epithelial-mesenchymal plasticity (EMP) is a proposed mechanism involved in establishment of metastasis. E-cadherin and Vimentin can be used as epithelial and mesenchymal markers, respectively, to identify EMP.

Aims: To compare E-cadherin and vimentin expressions between node-negative and node-positive invasive breast carcinoma-no special type-(IBC-NST) and their lymph node metastases.

Methods: Forty node negative tumours and 44 axillary node positive tumours of IBC-NST with their metastatic lymph nodes were retrieved from patients who had undergone mastectomy at the Teaching hospital, Peradeniya. Samples were subjected to dual immunohistochemical staining with anti-E cadherin and anti-vimentin primary antibodies. Any cytoplasmic staining for Vimentin and 70% or more of membranous staining for E-cadherin were taken as positive and cells counted in 10 high power fields. Intensity of staining was graded as high-3, moderate-2, low-1 and absent-0 by comparing with internal controls. Percentage of positive cells was multiplied by the intensity to obtain the immuno-score-(IS) for both protein expressions. IS was compared using independent and paired t-test.

Results & Conclusions: A lower E-cadherin ($p=0.001$) and higher vimentin ($p=0.008$) expression was seen in node positive group compared to node negative group. A higher E-cadherin ($p<0.001$) and lower vimentin ($p<0.001$) expression was seen in lymph nodes compared to the corresponding primary tumours.
A significant loss of epithelial properties and acquisition of mesenchymal properties in the node positive group indicates epithelial to mesenchymal transition of tumour cells, contributing to metastasis. In the lymph nodes, higher E-cadherin and lower vimentin expression suggests a reverse mechanism i.e. Mesenchymal to epithelial transition. These results are suggestive of epithelial-mesenchymal plasticity of tumour cells in establishment of metastasis in IBC-NST.

COLORECTAL NEUROENDOCRINE CARCINOMA WITH SQUAMOUS CELL DIFFERENTIATION: A RARE AND UNUSUAL TUMOUR

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Background: Neuroendocrine carcinoma (NEC) of the colorectum is rare, accounting for 0.4% of all colorectal neoplasms[1]. Squamous cell carcinoma (SCC) of the gastrointestinal tract is also a rare entity and accounts for 0.1-0.25% per 1000 cases of colorectal carcinoma [2].

World Health Organization (WHO) classification defines mixed adeno-neuro-endocrine carcinoma as a neoplasm with both exocrine and neuroendocrine components, with each component exceeding 30%.

Aim: We present a rare case of a patient with colorectal neuroendocrine carcinoma with squamous differentiation.

Method: An 86-year-old gentleman presented with right sided abdominal pain. A colonic mass was noted on CT scan with multiple liver lesions. Subsequent right hemicolectomy revealed an obstructive hepatic flexure tumour.

Results & Conclusions: Histopathological examination of the mass revealed a high grade neuroendocrine carcinoma (NEC) with diffuse sheets and cords of epithelial cells with crowded nuclei, scant cytoplasm and frequent mitotic figures (23 per 10 high-power-field).

Amongst the NEC component, small areas (less than 30% of the tumour volume) comprising larger epithelial cells with abundant eosinophilic cytoplasm and areas suggestive of keratinisation were noted.
The NEC component was positive for synaptophysin and CD56 immunohistochemistry with a Ki67 proliferation index of 80-90% and negative for P40 and CK5/6 immunohistochemistry. In contrast, the areas with large eosinophilic cells were positive for P40 and CK5/6 immunohistochemistry, whilst negative for synaptophysin and CD56 immunohistochemistry.

The tumour exhibited aggressive behaviour with lymphovascular, perineural and venous invasion and lymph node metastases.

Neuroendocrine carcinomas and squamous cell carcinomas of the colorectum are rare and extremely aggressive neoplasms[3] on their own merits. This phenomenon of mixed neuroendocrine carcinoma with squamous cell differentiation is an unusual and rare pathology which lacks evidence-based guidelines for therapy. It is worth documenting as the optimal therapeutic strategy remains the main challenge.

References:

1. FT Bosman, F Carneiro, RH Hruban et al. eds. WHO classification of tumours of the digestive system, vol. 3. 4th ed. Lyon: International Agency for Research on Cancer, 2010.
2. Shinya Munakata, et al. Mixed neuroendocrine carcinoma and squamous cell carcinoma of the colon: case report and literature review. Case Rep Gastroenterol 2018; 12:240-246.
3. Konstantinos Vardas, et al. Mixed large cell neuroendocrine carcinoma with squamous cell carcinoma of the rectum: report of a rare case and review of the literature. International Journal of Surgery Case Reports 4 (2013) 1076-1079.
4. Adel Elkbuli, et al. Mixed neuroendocrine and squamous cell carcinoma of the colon: A case report and literature review. International Journal of Surgery Case Reports 60 (2019) 309-313.

IDENTIFICATION OF POTENTIALLY MISLEADING TERMINOLOGY IN THE FIFTH EDITION WHO CLASSIFICATION OF TUMOURS DIGESTIVE AND BREAST TUMOURS BLUE BOOKS

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Background: Terminology with misleading potential is a problem with recognised impact in all medical specialties, including training and research. The WHO Classification of Tumours (WCT) is not excluded.

Aims: To identify and describe the frequency of use of a set of potentially misleading terms in the 5th edition WCT Digestive System and Breast Tumours Blue Books.

Methods: The project had two parts. Firstly, four independent pathologists of the WCT group identified a set of 26 potentially misleading pathology-related terms, including prefixes and suffixes through consensus. The main investigator reviewed 15 relevant information sources, including dictionaries, for definitions of these terms and compared these with each other. Secondly, a descriptive analysis was performed on the frequency of use of potentially misleading terms in the two recently published 5th edition WCT.

Results & Conclusions: Twenty-six terms were identified. No sources provided definitions for all terms. Eight of 15 sources used the terms or described them without providing a true definition.

The Digestive System Tumours book used misleading terms 1477 times. The six terms most frequently observed were “dysplasia”, “high-grade dysplasia”, “epithelioid”, “pseudo”, “oid” and “-like”.

The Breast Tumours book used misleading terms 574 times. The six terms most frequently observed were “microinvasion and microinvasive”, “Paget disease”, “epithelioid”, “-pseudo”, “oid” and “-like”.

The Digestive System Tumours book showed a more even distribution of potentially misleading terms across chapters than the Breast Tumours book, which had more than half (57%) confined to one chapter. The suffix “-oid” was the most frequently used term in both books (38% in Digestive System Tumours and 46% in Breast Tumours).

In conclusion, terminologies were defined inconsistently and use of selected terms was frequent. Uniform definitions for commonly misused terms are required to enhance clarity of communication in medical writing and improve medical education.

A RETROSPECTIVE ANALYSIS OF THE EFFECT OF TWO DECALCIFYING AGENTS ON THE QUALITY AND QUANTITY OF DNA FROM BONE TISSUE

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Background: Formic acid is currently the decalcifying agent in our laboratory. However, it is known to cause damage to DNA, thus limiting the availability of high-quality DNA for downstream molecular application. The use of milder decalcifying agent, such as ethylenediaminetetraacetic acid (EDTA), has been advocated as it has been shown to yield improved DNA from bone marrow tissues.

Aims: To determine and compare the effects of formic acid and EDTA on the quantity and quality of DNA from bone tissue and to compare the duration of the decalcification process.

Methods: Femoral heads of 19 patients were retrospectively selected for study. A 4-mm slice of bone had previously been sectioned and decalcified using 15% formic acid. Two 4-mm bone slices were sectioned from the left-over material, fixed in 10% neutral buffered formalin, and decalcified using EDTA. The endpoint detection was determined based on 'physical' and 'chemical' methods.
DNA from EDTA-decalcified bone was extracted and compared to those extracted from formic acid-decalcified bone.
DNA quantity was determined by UV-spectrophotometer and DNA quality by DNA amplification of different lengths (incl. 135bp, 315bp, and 480bp).

Results & Conclusions: DNA quality from EDTA-decalcified bone was better. The percentage of EDTA-decalcified bone, yielding 135bp-, 315bp-, and 480bp-long DNA fragments were 69% (N=36), 75% (N=30), and 73% (N=19), respectively. By contrast, 31% (N=16), 25% (N=10), and 27% (N=7) of formic acid-decalcified bone generated 135bp-, 315bp-, and 480bp-long DNA fragments, respectively.
Endpoint detection by 'physical' or 'chemical' methods resulted in no difference in the quality and quantity of DNA.
On average, decalcification by EDTA was 3.2 days vs. 1.5 days for formic acid.
In conclusion, the study showed that bone tissue decalcified by EDTA resulted in improved DNA quality. While turn-around time for EDTA-decalcified bone will be prolonged, the improved DNA may prove invaluable as molecular diagnostics and therapeutics play an increasingly important role in patient care.

ATYPICAL SPINDLE CELL LIPOMATOUS TUMOR – A CASE REPORT

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The atypical spindle cell lipomatous tumor (ASLT) is a relatively new entity proposed by Mentzel et al in 2010. The clinicopathological characteristics were further detailed in a large series of 232 cases by Marino-Enriquez et al in 2017. These lesions presented as a persistent or enlarging mass with a medium size of 5 cm at a wide anatomic distribution. A spectrum of cytological atypia present in these lesions could be alarming. The cellular cases with marked nuclear atypia and frequent lipoblasts could be mistaken as atypical lipomatous tumour/ dedifferentiated liposarcoma. Lack of MDM2 amplification is an important biological characteristic different from its morphology mimic. Despite the concerning histomorphology, none of those 232 patients in the 2017 series developed metastasis or died of disease. The local recurrence rate was 13%.

We hereby present such an interesting case in which an 81-year-old male presented with an 8 cm mass at the right shoulder. The cut surfaces of the mass showed fatty tissue with variegated and gelatinous areas. Microscopic examination revealed an admixture of variably sized adipocytes, univacuolated or multivacuolated lipoblasts and spindle cells. Many spindle cells were bland while the remainders were irregular, pleomorphic and hyperchromatic. Multinucleated cells reminiscent of floret giant cells were also present. The stroma was variable ranging from myxoid to collagenous nature. CD34 was diffusely positive among the spindle cells and pleomorphic cells. S100 was positive among the adipocytic cells and negative among the spindle cells. MDM2 and CDK4 immunostains were negative. FISH analysis showed no evidence of MDM2 amplification.

The poster will demonstrate the morphological spectra of ASLT and the awareness of this entity will avoid a malignant misdiagnosis.

TUMOR-TO-TUMOR METASTASIS: METASTATIC RECTAL ADENOCARCINOMA WITHIN ADENOCARCINOMA OF THE LUNG, AND A REVIEW OF THE LITERATURE

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Background: Tumor-to-tumor metastasis (TTM) is a rare but well-recognized phenomenon. When presented with a case of synchronous colon and pulmonary adenocarcinoma in the lung, one must consider the differential diagnosis of TTM and exclude a tumor with divergent phenotype because of difference in clinical management.

Aims: Herein, we present a rare case of TTM, in which a rectal adenocarcinoma has metastasized to a pulmonary adenocarcinoma. We illustrate how morphologic, immunohistochemical, and molecular analyses can help in the pathological diagnosis of TTM.

Methods: A case of TTM with a review of the literature is presented.

Results & Conclusions: A 67-year old male with a history of metastatic rectal adenocarcinoma to the liver has been in regular follow-up since 2012. Post-operative colonoscopy was normal and a computer tomography scan in 2015 revealed a 0.6 cm subpleural nodule in the left upper lobe of the lung. For the next 3 years, the nodule only showed mild interval enlargement, until a follow-up CT scan in 2019 revealed sudden increase in size to 1.5 cm. Suspicious of lung metastasis, the nodule was resected and histopathological analysis unexpectedly revealed a malignant tumor composed of a pulmonary and metastatic rectal adenocarcinoma. IHC for Napsin A, TTF-1, and CK7 was positive in the pulmonary adenocarcinoma, while CK20 and CDX-2 were positive in the metastatic rectal adenocarcinoma. KRAS mutation was identified in the pulmonary but not in the metastatic carcinoma. The latter was wild-type for KRAS and had TP53 mutation similar to the original rectal tumor. A review of the literature identified some 165 cases, of which only one previous case of colon adenocarcinoma metastasizing to lung adenocarcinoma has been reported.

Diagnosis of TTM requires histomorphologic assessment and is aided by immunohistochemical and molecular analysis. Awareness of TTM is important because of the significant difference in clinical management.

THE CORRELATION OF PROGRAMMED DEATH LIGAND 1 IMMUNOEXPRESSION WITH NEUTROPHIL-TO-LYMPHOCYTE RATIO IN DIFFUSE LARGE B-CELL LYMPHOMA

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Background: Diffuse large B-cell lymphoma (DLBCL) is the most common subtype of Non-Hodgkin Lymphoma. Host immune response in the tumor microenvironment (TME) plays a critical role in DLBCL tumor progression. Programmed Death Ligand 1 (PD-L1), an immune check-point creating local immunosuppressive condition in TME. The Neutrophil-to-Lymphocyte Ratio (NLR) has been reported to be associated with poor prognosis in several malignancies. However, the role of neutrophil in TME is controversial, whether as protumor or antitumor immunity.

Aims: This study aimed to investigate the correlation of PD-L1 expression with NLR in DLBCL.

Methods: This cross-sectional study included 40 cases of newly diagnosed DLBCL selected from Hasan Sadikin General Hospital, Bandung, Indonesia, from November 2015- July 2019. Demographic, clinical data and NLR of white blood cell count were retrieved from the first hematological profile on medical records by summing up total neutrophil segments and bands dividing by total lymphocyte count. PD-L1 immunoexpression in tumor tissues was detected using immunohistochemistry. Data were analyzed with the ROC curve & Pearson Correlation Test statistically using SPSS 20.00 and Medcalc 19.1.

Results & Conclusions: High PD-L1 immunoexpression was detected in 6 of 40 cases (15%) and the result showed that there was a significant moderate correlation between PD-L1 expression and NLR (r = 0.380, p=0,016, cut off NLR 2,2). These results support a concordance between the role of PD-L1 and neutrophil as protumor immunity that leads to DLBCL tumor progression.
Higher NLR also has higher immunoexpression of PD-L1. Higher NLR reflects low host immune response and progress of tumor growth. Higher circulating neutrophils in peripheral blood also reflect higher recruiting neutrophils into the tumor site in the tissue. Higher IFNγ stimulated neutrophils can upregulate PD-L1. Evaluation of NLR may be used as consideration for anti-PD-L1 immunotherapy eligibility. Further investigation is needed to support this research.

References:

1. WHO. GLOBOCAN. Cancer Today. International Agency for Research on Cancer. 2018;(cited September 30).
2. Song M, Park B, Uhm J. Understanding Immune Evasion and Therapeutic Targeting Associated with PD-1 and PD-L1 Pathway in Diffuse Large B-Cell Lymphoma. Int J Mol Sci. 2019;20(1326):1-15.
3. Shaul M, Fridlender Z. Cancer-related Circulating and Tumor-associated Neutrophils-Subtypes, Sources, and Function. The FEBS Journal. 2018;285(18):4316-42.
4. Ren F, Zhao T, Liu B, Pan L. Neutrophil-Lymphocyte Ratio (NLR) Predicted Prognosis for Advanced Non-Small Cell Lung Cancer (NSCLC) Patients Who Received Immune Checkpoint Blockade (ICB). Onco Targets Ther. 2019;19(12):4235-44.
5. Sangkhamanon S, Jongpainat A, Sookpraset A, Wirason K, Titapun A, Pugkhem A. Programmed Death-Ligand 1 (PD-L1) Expression Associated with a High Neutrophil/Lymphocyte Ratio in Cholangiocarcinoma. Asian Pac J Cancer Prev. 2017;18(6):1671-4.

UTILITY OF FOXL2 MUTATION TESTING IN DIFFERENTIAL DIAGNOSIS OF ADULT GRANULOSA CELL TUMOUR

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Background: Adult granulosa cell tumour (AGCT) is a low-grade malignant sex cord-stromal tumour accounting for approximately 5% of malignant ovarian tumours, with a peak incidence occurring at 50-55 years. The majority of cases demonstrate distinctive clinical and morphologic features, and up to 97% of AGCT cases carry a recurrent, somatic missense mutation in codon 134 of the transcription factor, FOXL2 (C134W). Whilst FOXL2 mutation testing may not be required in cases with classic morphology and immunophenotype (inhibin, calretinin, CD99 positive), FOXL2 mutation testing aids diagnosis in problematic cases. The differential diagnosis may include other sex cord-stromal tumours, such as cellular fibroma, thecoma, and juvenile granulosa cell tumour, all of which have a very low rate of FOXL2 mutation. Whilst immunohistochemistry is useful in confirming sex cord-stromal tumour type, immunophenotype does not distinguish between tumours within this category.

Aims: We explore the histologic spectrum of AGCT and other sex cord-stromal tumours; and correlate the findings with FOXL2 mutation testing.

Methods: FOXL2 mutation testing was performed initially using targeted PCR and Sanger sequencing (Hudson Institute) or subsequently by Next-Generation Sequencing using the Illumina Trusight Tumor 26 kit (Austin Health).

Results: Cases discussed comprise 3 AGCTs confirmed with FOXL2 mutation: 2 with atypical pathological features (1 luteinised, 1 extensively necrotic), and one with atypical clinical behaviour; and 2 cases of sex cord-stromal tumour where absence of FOXL2 mutation supported diagnosis (cellular fibromas).

KRAS AND *BRAF* MUTATION RATES AND SURVIVAL OUTCOMES IN COLORECTAL CANCER IN AN ETHNICALLY-DIVERSE COHORT

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Background: *KRAS*-mutant colorectal carcinoma (CRC) is resistant to anti-EGFR antibodies. Several studies compare the *KRAS* mutation rates between Caucasians and African Americans, but few studies have compared other ethnicities.

Aims: To compare the *KRAS* and *BRAF* mutation rates, as well as overall survival rates, in CRC between ethnic groups. Additionally, we report on correlations between clinicopathological variables relevant to CRC.

Methods: Clinicopathological data from 284 ethnically-diverse patients with CRC were retrospectively analysed at Liverpool hospital (2014-2016). *KRAS* and *BRAF* mutations were detected using real-time PCR kits (from Qiagen). Ethnicity was determined using both country-of-birth and surname. Survival outcomes were available for 191 patients. Overall survival was defined as date-of-diagnosis to death from any cause. Patients still alive were censored at 50-months post-diagnosis.

Results & Conclusions: *KRAS* mutation frequency differed significantly between Caucasians and Asians (36.1% vs. 56.4%, p=0.018), and Asians and South-Americans (56.4% vs. 16.7%, p=0.016), but not between other groups (Middle-Easterners:45.7%). *BRAF*-V600E mutation frequency differed significantly between Caucasians and Middle-Easterners (16% vs. 0%, p=0.029), but not between other groups (Asian:8.6%, South-American:16.7%). The 50-month survival rate did not differ between ethnicities (Caucasians:44%, Middle-Easterners:52%, Asians:58%, South-Americans:50%). Of patients who died, the median survival time was 28 months. Overall survival did not significantly differ between patients with wild-type and mutant *KRAS*. Tumours with *BRAF*-V600E mutations were more likely to display microsatellite instability (MSI) than tumours with wild-type *BRAF* (45.5% vs. 8.3%, $p<0.001$). Tumours displaying MSI occurred more frequently in the right colon ($p=0.007$). *KRAS* mutations were more frequent in circumferential tumours ($p=0.04$), and tumours without vascular invasion (p=0.004).

ANALYSIS OF CLINICAL AND MOLECULAR PROFILES OF PATIENTS WITH INNATE RESISTANCE TO ANTI-PD-1 +/- ANTI-CTLA-4 IMMUNOTHERAPY IN METASTATIC MELANOMA

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Background: Immune checkpoint inhibitors targeting the CTLA-4 and PD-1 receptors have significantly improved the outcomes of many patients with metastatic melanoma. However, approximately 50% of patients do not respond to immunotherapies. Understanding the underlying clinical, pathologic and genetic factors associated with failed response to immunotherapies is key to identifying therapeutic strategies to overcome resistance.

Aims: This study sought to characterise patients with innate resistance to anti-PD-1-based immunotherapies by investigating their baseline tumour characteristics.

Methods: Targeted RNA-sequencing using a custom panel of 500 genes was performed on pre-treatment formalin-fixed paraffin-embedded (FFPE) specimens from 37 non-responders (SD≤6 mo/PD) to anti-PD-1+/-anti-CTLA-4 therapy with metastatic melanoma. The levels of tumour-infiltrating lymphocytes (TILs) were assessed on hematoxylin and eosin stained FFPE sections. Clinical characteristics were compared between groups of non-responders based on their time to progression, level of TILs, and gene expression data.

Results & Conclusions: Patients clustered into two groups based on their gene expression profiles, with one group (*n*=19) expressing genes associated with interferon signalling and exhaustion (*STAT1, TIGIT, TBX21, HAVCR2,* and *FASLG*), and the second group (*n*=18) expressing genes associated with antigen presentation and angiogenesis (*HLA-A, TAPBP, ANGPT1,* and *WARS*). A significantly higher proportion of patients in the second cluster presented with brain metastases compared to those patients in the first cluster (*P*=0.03). There were no other significant differences in clinical characteristics between the two groups (*P*>0.05). Patients with rapid progression (PFS<2 mo) had significantly lower TILs compared to other non-responders (*P*=0.04), and this subset of patients also displayed an overall lower immune-related gene expression profile. Furthermore, a trend towards a higher tumour burden was observed in rapid progressors (*P*=0.06). These findings demonstrate the inter-patient heterogeneity within non-responders, and highlight the need for a personalised and multilayer (clinical and molecular) approach for identifying the most accurate treatments for such patients.

THE UTILITY OF TUMOUR INFILTRATING LYMPHOCYTES AS A RISK FACTOR FOR LOCAL RECURRENCES OF ABDOMINAL WALL FIBROMATOSIS

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Background: Desmoid-type fibromatosis (DF) is a locally aggressive tumour of fibroblastic/myofibroblastic origin that usually occurs in young adults. Extra-abdominal tumours (including abdominal wall tumours) is a biologically different type with special etiology, behavior, and the morbidity associated. Abdominal wall fibromatosis comprises a major sector of fibromatosis cases. Until now few risk factors are being documented as predictors of tumour recurrences in this type.

Aim: In the current study we proposed that the nature of the lymphocytic infiltrate in fibromatosis may provide a novel risk factor for predicting recurrences and may provide a novel approach for immunotherapy.

Methods: Forty retrospective cases of abdominal wall desmoid tumours are collected from two hospitals from cases from 2011 to 2014. Cases are divided into two groups according to occurrence of recurrences in the period of 5 years (until 2019). Sections from the tumour were stained for immunohistochemical markers for CD4 and CD8. The number of positive cells for each marker was examined in 5 high power fields, both in tumour invading margin and tumour core using image analysis system.

Results: Higher density of CD4 and CD8 T lymphocytes were found in cases of non- recurrent cases group ($P < 0.05$)The tumour margin CD4 and CD8 cells are more associated with non -recurrent cases that intra-tumoral (core) cells ($P<0.05$).

Conclusion: The study concluded that high CD4 and CD8 positive T lymphocytes mainly in the tumour margins are associated with favorable recurrence - free survival in abdominal wall fibromatosis and may present a portal for future immunotherapy for these cases.

MIXED EPIDERMAL CYST AND MUCINOUS NEOPLASM, A RARE COMBINATION

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Background: Epidermal cyst is a rare neoplasm of the ovary, comprises only <1% of the benign ovarian neoplasm. On the other hand, mucinous cystic neoplasm is one of the commonest ovarian tumor. Although it's frequent coexistent with other tumors, for example mature cystic teratoma, squamous cell carcinoma, clear cell adenocarcinoma, Brenner's tumor, serous cystadenoma etc. are well revealed, a combination of benign epidermal cyst and mucinous cystic neoplasm is rarely documented in the literatures.

Aims: To present this rare combination of tumours.

Methods: The cases were reviewed and discussed in details. Microscopic pictures are taken.

Results &Conclusions: Here we discuss two cases of ovarian epidermal cyst recently encountered in our department, one associated with a mucinous cystadenoma and the second one with a borderline mucinous tumor.

PULMONARY CRYPTOCOCCUS IN A 62-YEAR-OLD PATIENT DIAGNOSED BY FINE NEEDLE ASPIRATION CYTOLOGY: A CASE REPORT

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Background of the Study: Pulmonary cryptococcosis is a disease occurring worldwide. Patient's immune status is the main factor determining the pathogenicity of the lung infection. Majority of normal hosts with cryptococcal infections are asymptomatic and only a small proportion has pulmonary symptoms.

Aims: To present a pulmonary infection, clinical presentation, histopathologic findings and course of the disease

Results: Patient is a 62-year-old male, diabetic with CKD who experienced non-productive cough, fever and difficulty of breathing. Few hours prior to admission, patient had episode of fever and dyspnea warranting admission. CT scan of the chest revealed pulmonary artery aneurysm in the right lower lung measuring 1.6x1.1 cm with an impression considering mycotic aneurysm in the right pulmonary artery. CT Scan-guided biopsy was done. Patient was treated as a case of cryptoccocosis.

The specimen consists of 2 ml mucoid to bloody right lung aspirate and smears show numerous yeast cells with thick mucoid capsular halos in monolayers admixed with macrophages and neutrophils. Vague granuloma formations are noted in a background of red cells. Cell block shows same yeast cells with macrophages and lymphocytes. PAS stain shows bright red yeasts and pale capsule with spiney formation.
Final diagnosis was chronic inflammation with granuloma formation; numerous fungal microorganisms morphologically consistent with *Cryptococcus neoformans*.

Conclusion: Pulmonary cryptococcosis is a relatively rare disease in the Philippines. This disease is commonly seen among immunocompromised individuals including patients with AIDS. Proper approach in early detection and diagnosis should be followed. This case implies that incidence of a lung lesion in an immunocompromised should never be underestimated regardless of HIV status.

INCREASED ROS1 EXPRESSION IN NSCLC ASSOCIATED WITH SOMATIC *EGFR* MUTATIONS

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Background: ROS1 immunohistochemistry has become routine to triage NSCLC cases lacking *EGFR* and *ALK* alterations for subsequent FISH testing to determine patient eligibility for crizotinib therapy. We observed that, in the large majority of cases showing increased ROS1 expression, *ROS1* rearrangements were not detected.

Aims: To determine whether somatic mutations in clinically significant genes in NSCLC were correlated with increased ROS1 expression detected by IHC.

Methods: 1,278 NSCLC specimens were assessed for somatic *EGFR, KRAS* and *BRAF* mutations in addition to ROS1 screening IHC testing (with D4D6 antibody). Confirmatory FISH testing was performed on IHC-positive or equivocal cases lacking *EGFR* and *ALK* alterations. The cohort comprised 1,261 patients (671 female, 590 male) with a mean age of 71 ± 10 yr (range 19 – 99 yr). SCCs and tumours of uncertain origin were excluded from analysis.

Results & Conclusions: *EGFR* mutated tumours were more likely to be ROS1 equivocal/positive than wildtype (46.0% vs 21.9% respectively, $p<0.001$) whereas the inverse was the case for *KRAS* mutated cancers (21.8% vs 35.4%, $p<0.001$). No association was observed with *BRAF* status.

Associations between ROS1 expression and *EGFR*, *KRAS* and *BRAF* were was determined (x^2 tests).

ROS1 rearrangements were detected in 9/84 (10.7%) of ROS1 IHC equivocal/positive tumours. Interestingly, ROS1 overexpressing tumours were more significantly more likely to harbor *EGFR* mutations, whereas an inverse correlation was shown between ROS1 and *KRAS* alterations. ROS1 immunohistochemistry is sensitive but not specific for predicting *ROS1* gene rearrangements, and expression is commonly seen in tumours with somatic *EGFR* mutations. The significance of this observation remains unclear.