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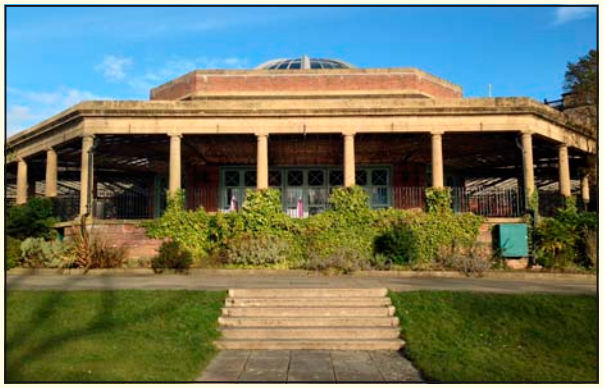

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P1

Contemporary Coronial Autopsy Practice in the NHS: An Audit

Ⓟ SS Chowdhury; SM McGrath

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An audit has been undertaken to review the coronial autopsy practice of a single histopathology consultant against the standards laid down by the Royal College of Pathologists 2002, National Confidential Enquiry into Patient Outcome and Death 2006, and Coroners Statistics Annual 2017 to identify areas of variance against national benchmarks. Outcomes from coronial autopsies facilitate trust morbidity and mortality review processes. The standard of post mortem practice and the accuracy of causes of death are therefore important factors to consider. The audit was approved by HM Coroner and registered with the Trust's audit department. Data were retrospectively collected from all post mortem examinations performed by a single consultant in 2017. The corresponding patient records were also consulted. The diagnostic value of histology and toxicology samples was explored. Discrepancies between suggested clinically and pathologically diagnosed causes of death were scrutinised. No attempt was made to reach objective judgements about the overall quality of the autopsy reports. 108 cases were audited of which 59% cases were community deaths and 41% of cases were hospital deaths. Compliance against most key standards was high, e.g. inclusion of supporting documentation, mandatory demographic details, external and internal examination. 52% of cases involved the retention of tissue samples; substantially higher than the national average for 2017 (23%). Histology confirmed the cause of death suspected during the post mortem examination (36%) or was essential in determining the cause of death (43%) in cases where tissue blocks were retained. Toxicology samples were retained in 24% of cases, compared to the national average (20%). It played a key role in determining the cause of death in 31% of cases. 60% of hospital deaths did not contain any clinical cause of death in medical notes. The clinician and pathologist given causes of death were similar in 33% of cases and significantly discrepant in 44% of cases. Histology remains a valuable component of autopsy practice. The autopsy sill has much to add when the cause of death cannot be determined clinically. Toxicology is very important in a small number of community deaths.

P2

This abstract has been withdrawn

P3

Enhancing Histopathology Laboratory Access Through Virtual Reality: Demonstrating Knowledge Gain and Potential Recruitment Benefits in Undergraduate Pathology Teaching

Ⓟ GGA Hutchins[1]; SR Bickerdike[2]; EV Verghese[1]; NP West[2]; R Bishop[3]; GS Frith[2]

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Use of Virtual Reality (VR) technology is increasing, both in postgraduate medical training and in undergraduate medical education. Development of VR resources, previously restricted to games developers, is now accessible to educators as a result of decreasing hardware/software costs. Because of the challenges of exposing undergraduate medical students to histopathology working practices (and therefore enhancing their interest in the subject), we evaluated the use of VR, and 360 degree video, as an adjunct to pathology teaching on a medical undergraduate degree course at a UK-based University. Using 360 degree video, enhanced with a full narration audio, interactive hotspots and high-definition video pop-outs, we produced a fully immersive walk-through 'tour' of the histopathology laboratories within a large UK-based teaching hospital. The resource was produced with the intention of being deliverable using two methodologies: a traditional laptop/tablet device or using VR headsets for a fully immersive tour experience. As a trial, we delivered the 360 VR resource to separate cohorts of year 2 medical students. The value of the resource as a whole was assessed consecutively over 2 years, initially using entire year cohorts and in smaller 'focus groups'. An emphasis was made, by application of pre- and post-intervention questionnaires, on students' knowledge gain and their perceptions of 'immersiveness' and the relative values of non-VR versus VR-based delivery platforms. The relative value of high cost VR headsets vs low cost (cardboard) VR viewers was also assessed. Knowledge gain was slightly greater using ipads (0.65 vs 0.47), student's feelings of immersiveness were greater using VR (73% vs 27%), particularly when using high-end VR viewers.Notably 50% of students indicated that they would be more likely to consider pathology as a career post intervention. We thus demonstrate the delivery of an immersive histopathology experience to large cohorts at low cost.

P4

Tips for Academic Pathology Trainees (APT): A Website offering Advice for Academic Pathology Trainees and Medical Students/Doctors Considering Histopathology as a Career

Ⓟ C Young; A Wright; M Waterhouse; P Quirke; D Treanor

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Histopathology and academic pathology suffer from low recruitment and high attrition. Resources which promote histopathology have been produced by different organisations, but this makes them difficult to locate by medical students, junior doctors, or doctors from other specialties who are interested in histopathology. Resources offering advice and support to academic pathology trainees are usually delivered as face-to-face meetings; not all trainees are able to attend and the information may not be relevant to trainees until later in their careers.

We have created a website, "Tips for Academic Pathology Trainees (APT): a website offering tips for academic pathology trainees, doctors considering histopathology and medical students" (http://www.apt.virtualpathology.leeds.ac.uk). This serves as a single-site, permanent, universally-accessible, comprehensive set of resources for both medical students/doctors interested in histopathology and academic pathology trainees.

Content is divided into 12 main sections: Histopathology; Pre-PhD Fellowships; PhD Fellowships; Clinical Lectureships; Teaching; Networking; Literature-based skills; Finances; Professional Relationships; Patient and Public Engagement; General Research Skills; and Inspiration.

Users can submit suggestions, comments or questions via the email address: aptwebsite@pathsoc.org.

The website was officially launched in January 2019 and has been promoted via Twitter and email distribution lists. Two months post-launch, the website has had 809 users and 5131 page views, with most users viewing 6–7 pages per session. Users are from the UK, USA, Canada, India, Ireland and the Netherlands. Promotion is ongoing and website content will be regularly reviewed and updated. The high website-usage figures indicate that the website addresses a previously unmet need. We encourage you to use, promote and engage with the website.

This work was generously funded by an Open Scheme PathSoc Grant.

P5

Taking Another Look: Using New Simulation Techniques to Evaluate Old Antibiotics

Ⓟ JWS Cattrall[1]; E Asín-Prieto[2]; J Freeman[1]; IF Trocóniz[2]; A Kirby[1]

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Purpose of the study: Resistance to oral antibiotics recommended for common infections is increasing. Modern modelling/simulation techniques can assist in discovering new indications and dosing regimens for older antibiotics.
Objective: To design a framework to assess alternative treatments/novel dosing regimens for the treatment of common infections using simulation.
Methods: The framework development considered modern, robust model development techniques. Several simulation software packages were considered. Decisions were made using both microbiological and pharmacological expertise.
Summary of results: A framework was developed to systematically evaluate pharmacokinetic models of suitable quality from the literature for a selection of antibiotics. Once identified, models are evaluated for robust development methodology using quantitative and qualitative methods: 1) a numerical score: confidence in quality check based on key components (goodness-of-fit, NONMEM relative standard error, alternative software equivalent precision estimate, bootstrap analysis and simulation-based model diagnostics (SBMD)). 2) reviewer assessment of quality and relevance (raw data fit and potential for extrapolation to other populations). Highest quality models are then selected. This information can be combined with locally collected MIC data. Pharmacokinetic-pharmacodynamic simulations (R package mxlR), using the Monte Carlo method, can calculate the area under the curve of concentration/time graphs. This enables the generation of cumulative fraction response (CFR) values for sub-populations of the bacterial population at standard and non-standard doses, indicative of clinical success.
Conclusions: It is possible to conduct feasibility assessments for a number of commonly used oral antibiotics for varying indications and dosage regimens using limited resources. These assessments can indicate new potential in older antibiotics.
This research was supported by a Pathsoc Travel Grant.

P6

Raising the profile of Histopathology among Medical Students: A District General Hospital Perspective (The Mid-Yorkshire Hospitals NHS Trust, An Associate Teaching Hospital Trust)

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Fewer medical students choose histopathology today as a career due to lack of awareness and poor perception. An ever-expanding curriculum, economic pressures and healthcare reforms negatively impact on pathology teaching. Profile raising engagement with medical students as part of existing clinical placements can increase appeal and demonstrates the value of the profession. Our experience shows how short integrated postings in a DGH Histopathology department can be delivered via vertical integration of pathology teaching during clinical placements (General Surgery, Special Senses, Oncology and General Medicine) in year 3 and above. This process of vertical integration allows the development of a spiral curriculum, which reinforces pathology concepts in year 3 and above. This approach also allows pathology to be revisited with an appropriate clinical context. Our short program includes a guided tour of the laboratory, specimen assessment in the cut up room, a posting-specific talk and a multi-header microscopy session. third year medical students have a day in the mortuary to observe post mortems and refresh their knowledge of anatomy and pathophysiology whilst encouraged to postulate a cause of death. Feedback is acted upon through peer review to improve teaching sessions and student feedback is shared to ensure continuous improvement. Students are invited to return in a voluntary capacity to shadow Consultants. We have introduced a Poster Presentation programme for students with the aim of presenting a poster at the annual Pathsoc scientific meeting. The programme has been assessed centrally and has been recommended to other DGH Histopathology Departments across Yorkshire. We are able to encourage students to view their learning in the context of both service delivery and clinical research.
We were awarded a Clinical Teaching Development Award 2014, nominated by students for Outstanding Clinical Team 2018 and awarded a Clinical Teaching Excellence Award 2019.

P7

This abstract has been withdrawn

P8

Adopting Digital Pathology into Clinical Practice: Perceived Advantages and Challenges

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Introduction: UK histopathologists are familiar with digital pathology in the setting of EQA schemes, and in an educational context. Few currently use this platform in the clinical diagnostic setting. As an NHS Department in the process of introducing a fully digital diagnostic histopathology service, we face challenges, both those that are described by others, and those associated with our own expectations and fears around digital pathology. These need to be recognised in order to increase the chances of success. At the start of our project we therefore sought to examine the perspectives, opinions and attitudes of the multiprofessional team most involved with introducing digital pathology.
Methods: A survey was sent out to members of the local Digital Pathology Steering Group. The survey incorporated questions to ascertain the opinions and attitudes of the team to the transition to digital pathology, and sought to identify areas of potential concern.
Results: 9 people completed the survey. Responses were positive with regard to the transition as a whole, with a general consensus about projected benefits for improved workflow, patient safety, and workforce (including network working). There was no real perceived concern about the process of transition in terms of time taken to become confident in digital reporting, or to report digitally compared with glass slides. Whilst generally the improved ease of sharing of cases was seen as benefit to patients and for education, concern was raised that facilitation of access to specialist histopathologists for second opinion might significantly increase referral work. It will be important that clear pathways for access to opinions are established and to ensure time is appropriately recognised and funded.
Conclusion: Overall the attitudes and opinions around the transition to digital pathology were positive. The perceived challenges are not insurmountable, and as identified at this stage they can be addressed.

P9

Inspiring the Next Generation: The National Academic Trainees' Network

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Purpose of the study: Over the last 15 years there has been a well-documented decrease in academic pathology capacity in the UK. Initiatives such as the CM-Path programme by the National Institute of Cancer Research are designed to remedy this decline. We recognised a lack of training in non-technical skills for junior academic pathologists and the lack of an effective network for sharing success, advice and opportunities. This led us to create the National Academic Trainees' Network (NATN).
Methods: The network meets three times per year and comprises a networking dinner and inspirational speaker with a full day of tutorials and workshops the following day. Sessions have included: advice on grant writing, social media strategy, legislation including research ethics and working with human tissue, lay communication, intellectual property, three-minute thesis presentations, research impact and the introduction of a mentoring scheme. We evaluated feedback from the meetings and the content of the network WhatsApp group.
Results: Feedback was available from eight of the ten NATN meetings that have taken place to date. The mean attendance was 22 delegates per meeting. Academic clinical fellows, lecturers and trainees from the alternative academic pathway were equally represented. Across all meetings the mean weighted average rating of all sessions was 4.28/5. A common theme from the feedback was that trainees found the networking opportunities and peer support elements of the meetings very useful. A NATN WhatsApp group has been active for 5 months. This has 33 members and has been used for advertising conferences and resources, discussing grant application logistics and developing future NATN sessions.
Conclusions: To our knowledge the network is unique amongst academic medical specialities. Trainees have given favourable feedback and the network appears to offer training not routinely provided by academic training programmes.

P10

Addendum Pathology Reports: Amended, Corrected or Supplementary? Does it matter?

Ⓟ AL Leeming; M O'Donnell

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Background: Addendum pathology reports are issued subsequently to a final report when additional details or a change to the report is required. Their rates may be used as a departmental key performance indicator of accurate reporting. However, not all such reports are due to departmental inaccuracies. Published guidance from the Faculty of Pathology, Royal College of Physicians of Ireland recommends that addendum reports be subcategorised into amended, corrected and supplementary types. Of these, amended reports are those which contain a change to the report significant enough to affect patient management such as an amendment to the diagnosis or stage. Corrected reports are used to correct typographical or other minor errors. We performed this study to analyse our amended reports to see if we could re-classify them using the above criteria focusing on the possible benefit of separating amended from corrected reports.
Methods: All amended reports issued by our department over a three month period were identified, reviewed and reclassified using the above criteria.
Results: A total of 19,523 specimens were reported over the study period with 63 amended reports issued (0.3%). When reclassified, only 18 (28%) of these amended reports detailed a change in diagnosis/stage from the original report. Other reports corrected minor errors and were better classified as corrected (57%), a few contained new information and should have been classed as supplementary (9%).
Conclusions: Clearer categorisation may help receiving clinicians recognise the importance and clinical relevance of amended reports as distinct from corrected reports. In addition it would allow departmental reporting error rates to be more accurately represented as only a quarter of issued amended reports actually contained true reporting errors in our cohort.

P11

Glutaminase Expression Predicts Recurrence in Ductal Carcinoma *In Situ*

ML Craze[1]; A Oldfield[1]; M Toss[1]; Ⓟ BK Masisi[1]; R El Ansari[1]; L Alfarsi[1]; I Miligy[1]; AC Al-Kawaz[1]; H Nicholls[1]; CC Nolan[1]; IO Ellis[2]; EA Rakha[2]; AR Green[1]

[1]University of Nottingham, Nottingham City Hospital, Nottingham, UK; [2]University of Nottingham, Nottingham Universities NHS Trust, Nottingham City Hospital, UK

Purpose of the study: Ductal Carcinoma in Situ (DCIS) shares many morphological and molecular similarities to invasive breast cancer (IBC) and precise predictors of DCIS recurrence and progression are still lacking. In order to meet increased metabolic demands of growth and proliferation, many cancers reprogram their metabolism to rely on glutamine. Glutaminase 1 (GLS1) is a key enzyme, which converts glutamine to glutamate in the glutaminolysis process. GLS1 expression is upregulated in IBC, but has not yet been studied in DCIS. In this study, we evaluated the expression of GLS1 in a large cohort of DCIS and assessed its prognostic significance.
Methods: In this retrospective study, GLS1 expression was assessed immunohistochemically in a large, well characterised DCIS cohort, consisting of 779 pure DCIS cases and 239 cases of DCIS-mixed with IBC. GLS1 expression was correlated with clinicopathological parameters, and outcome analysis was evaluated using local recurrence free interval (LRFI).
Summary of results: GLS1 expression was associated with features of high-risk DCIS, including symptomatic presentation (p=0.04) and tumours that were higher grade (p=0.014), ER negative (p<0.001), PR negative (p=0.003), as well as high Ki67 (p=0.036) and high hypoxia inducible factor 1α (HIF1α) (p<0.001) expression. GLS1 expression was higher in DCIS mixed with IBC compared to pure DCIS (p=<0.001). GLS1 expression was an independent prognostic feature in predicting both invasive (HR=4.1, 95%CI 1.4 to 12.0, P=0.012) and DCIS recurrence (HR=19.8, 95%CI 4.3 to 92.0, P<0.001).
Conclusion: We show for the first time, that GLS1 may play an important role in DCIS progression as well as predicting recurrence in DCIS patients. This study also highlights the potential therapeutic role of GLS1 inhibitors in DCIS patients.

P12

Over Expression of Retinoid X Receptor Gamma (RXRG) Predict Good Prognosis in Oestrogen Receptor Positive Breast Cancer

C Joseph; Ⓟ S Al-izzi; M Alsaleem; S Kurozumi; MS Toss; M Arshad; FQ Goh; MA Aleskandarany; NP Mongan; AR Green; IO Ellis; EA Rakha

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Background: Breast Cancer (BC) is globally one of the most prevalent malignancies and a leading cause of cancer-related death. Retinoid X Receptor Gamma (RXRG) is a member of the nuclear receptor superfamily, which interacts with other nuclear receptors and plays a role in tumour suppression. This study aims to investigate the prognostic role of RXRG in BC.
Methods: RXRG protein expression was evaluated using a large well-characterised BC cohort (n=923) prepared as tissue microarrays. The association with different clinicopathological parameters and patient outcome were investigated. Prognostic significance of RXRG mRNA expression was also assessed using breast cancer gene miner (bc-GenExMiner v4.2).
Results: High nuclear RXRG expression is associated with good prognostic features including good Nottingham Prognostic Index group (p<0.05), lower histological grade (p=0.04) and smaller tumour size (p=0.036). Strong positive associations were observed with oestrogen receptor (ER) positivity and ER-related biomarkers: GATA3, FOXA1, STAT3 and MED7 (p<0.00001), and reduced expression of the proliferation marker Ki67 (p=0.014). RXRG overexpression was associated with longer BC-specific survival (p<0.0001) and less probability for the development of distant metastasis (p=0.003). In ER-positive tumours, high expression of RXRG showed significant survival advantage regardless of adjuvant systemic therapy (p=0.04). RXRG expression is an independent prognostic factor associated with improved survival, particularly in ER-positive BC. In the external validation cohorts, RXRG mRNA expression was associated with improved patients' outcome (p=0.025). Differential gene expression evaluation identified ER signalling pathway as the principal predicted master regulator of RXRG expression (p=0.005).
Conclusion: The findings support the proposed role for RXRG as a prognostic marker in ER-positive BC. Exploring the utility of RXRG as a potential therapeutic marker is warranted.

P13

Keratin 24 (KRT 24) Confers Poor Patient Outcome in Invasive Breast Cancer

Ⓟ Y Kariri; C Joseph; S Kurozumi; I Miligy; S Al Saeed; A Aljohani; PL Narasimha; IO Ellis; NP Mongan; M Aleskandarany; AR Green; EA Rakha

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Background: Keratin (KRT) 24 is cytoskeletal protein playing a major role in the formation of the intermediate filaments that provide mechanical stability. The components of the cytoskeleton mediate tumour cell migration, invasion and metastasis. Metastasis is the major cause of breast cancer (BC) related deaths. Through stringent bioinformatics analysis we identified KRT24 overexpression as strongly associated with poor patient outcome; however, its role in BC remains unclear. The study investigates the clinicopathological significance of KRT24 at transcriptomic and proteomic levels using large cohorts of BC patients with long term follow-up.
Methods: KRT24 mRNA expression was assessed in the METABRIC (n=1980) cohort and externally validated in BC Gene miner v4.0. Primary BC tissue microarrays (n=827) were immuno-stained for KRT24 and correlated with clinicopathological features, patient outcome and with other BC related markers.
Results: KRT24 mRNA was associated with poor prognostic features, including high histological grade, ER negativity, HER2 positivity and is overexpressed in luminal B and HER2+ subtypes (all; p<0.05). KRT24 protein expression was significantly associated with features of aggressive phenotype including HER2-positivity (p=0.003), cycline E (p=0.026), N-cadherin (p=0.012), and epidermal growth factor receptor (EGFR; p=0.043) and basal phenotype (p=0.016). Using BC Gene miner (n=3163), high KRT24 mRNA expression was associated with poor patients' outcomes (p=0.027). High KRT24 protein expression was associated with poor BC-specific survival (p=0.029). Cox proportional multivariate analysis revealed that high KRT24 is a predictor of shorter BC-specific survival, an independent of other clinicopathological factors (p=0.023).
Conclusion: This study confirmed the association between KRT24 expression and poor prognostic features and metastasis related biomarkers; results warranting further functional validation.

P14

CD133 Over-Expression in Breast Cancer: A Marker of Poor Prognosis

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University of Nottingham, Nottingham, UK

Purpose: The cancer stem cell marker, CD133, is associated with poor prognosis in various solid tumours, but its role in invasive breast cancer (BC) remains unclear. Thus, this study aims to evaluate the prognostic importance of CD133 expression utilising large well characterised BC cohorts with long-term follow up.
Methods: CD133 mRNA expression were assessed in the METABRIC cohort and externally validated in BC Gene miner v4.0. Primary BC tissue microarrays (n=687) were immuno-stained for CD133 and correlated with clinico-pathological features, patient outcome and with other stem cell markers.
Results: CD133 protein expression showed a positive correlation with CD133 mRNA (Spearman's coefficient 0.505; p<0.00001). CD133 immunopositivity was observed in the cytoplasm / membrane of invasive cancer cells. Similar to mRNA expression, high CD133 protein levels were associated with high grade, larger tumour size, poor Nottingham Prognostic Index, HER2 positivity and hormonal receptor negativity (all; p<0.001). CD133 protein overexpression was significantly correlated with features of aggressive phenotype including basal cytokeratins CK5/6, CK14 and CK17, Epidermal Growth Factor Receptor (EGFR) and proliferation marker Ki67 (all; p<0.05). A strong positive association with other BC stem cell markers such as CD24, CD44, SOX10, and ALDHA3 were observed (p=0.020). High expression of CD133 protein was associated with shorter BC-specific survival in the whole cohort (p<0.001) and Her2+ subgroup (p=0.04). Cox proportional multivariate analysis showed that that CD133 protein expression was an independent indicator of shorter BC-specific survival (p=0.038).
Conclusion: This study provides evidence for the prognostic value of CD133 in invasive BC particularly in the aggressive HER2+ subtype of BC, and is, therefore, a potential therapeutic target.

P15

Flap Endonuclease 1 (FEN1) is a Prognostic Biomarker in Ductal Carcinoma *In Situ* (DCIS)

Ⓟ A Al-Kawaz; K Mesquita; M Toss; I Miligy; AR Green; IO Ellis; S Madhusudan; EA Rakha

University of Nottingham/ City Hospital, Nottingham, UK

Background: Carcinogenesis could be driven by impaired DNA repair which clinically promotes aggressive behaviour in breast carcinoma. Damaged DNA bases can be removed accurately by base the excision repair (BER) pathway. FEN1 is a major component in the BER which has important roles in genomic stability maintenance through the rescue of stalled replication forks, maintenance of telomere stability and apoptosis. FEN1 also plays a role in replication by controlling the Okazaki fragment maturation. High expression of FEN1 is associated with poor outcome in invasive breast cancer (IBC), however, it is not confirmed in the pre-invasive stage. We hypothesised that FEN1 overexpression is an early event in breast cancer pathogenesis. The aims are to assess the role of FEN1 in DCIS with the potential to predict DCIS progression to invasive disease.
Methods: 779 pure DCIS and 239 mixed DCIS/IBC cases were constructed. The expression of FEN1 was assessed using immunohistochemistry and correlated with the clinicopathological parameters and patient outcome.
Results: In a pure DCIS cohort, high expression of FEN1 was associated with higher DCIS grade (P<0.0001), size >20mm, P=0.008), presence of comedo necrosis (P<0.0001), ER and PR negative tumours (P<0.0001) and high expression KI67 labelling index (>14%) (P<0.0001). DCIS component of the mixed cases showed higher expression compared with DCIS in the pure DCIS cohort (P=0.019) but showed significantly lower expression compared with the coexisting invasive component (P<0.0001).
Conclusion: These results suggest that overexpression of FEN1 is associated with aggressive DCIS. We concluded that overexpression of FEN1 in pre-invasive DCIS is linked to the invasive breast cancer, but it's not linked to local recurrence. FEN1 in DCIS is a poor prognostic factor that can potentially help in in situ risk stratification.

P16

High SLC1A5 Expression Predicts Resistance to Endocrine Therapy in ER+ Breast Cancer via Interacting with Metabolic Pathway

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University of Nottingham, Nottingham, UK

Background: Identification of effective and reliable biomarkers to predict the efficacy of endocrine therapy and understanding the molecular pathways that contribute to the development of endocrine resistance are of crucial importance to the management of oestrogen receptor positive (ER+) breast cancer (BC). Glutamine-dependence is an established hallmark of cancer and the transport of glutamine into the cell via SLC1A5 has an emerging importance as a diagnostic and therapeutic target.
Methods: We investigated the biological impact of SLC1A5 expression in a clinical samples of ER+ BC at the mRNA, using METABRIC and KM-Plotter datasets, and the protein levels using immunohistochemistry in a large annotated series of ER+ BC with long-term follow-up. Additionally, bioinformatics analysis were used to study the interacting networks of SLC1A5, and its biological processes and enriched pathways.
Results: SLC1A5 expression was associated with poor clinicopathological parameters (vascular invasion, large tumour size and high grade; (P<0.05), and poor clinical outcome (P<0.05). SLC1A5 expression also significantly correlated with tamoxifen resistance in patients treated with adjuvant tamoxifen monotherapy (P<0.05). In silico analysis, showed the majority of SLC1A5 correlated-genes within the biological interacting network were were significantly enriched within the metabolic and MAPK signalling pathways. In the top-25 genes of interacting network with SLC1A5, TALDO1 was involved in metabolic pathway and associated with poor clinical outcome in ER+ BC and resistance to endocrine therapy.
Conclusion: This study shows that the glutamine transporter SLC1A5 could act as potential predictive biomarker of poor benefit from endocrine therapy in BC and a potential therapeutic target

P17

The Prognostic Significance of PDZ Domain-containing 1 (PDZK1) in Invasive Breast Cancer and its Association with ER Heterogeneity

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Background: Endocrine therapy is the standard systemic treatment for oestrogen receptor (ER) positive breast cancer (BC), however approximately 40% of these patients develop recurrence. ER intra-tumour heterogeneity is a potential reason for resistance to endocrine therapy and subsequent recurrence. The protein PDZ domain-containing 1 (PDZK1) is encoded by an oestrogen-responsive gene present in BC cells. The study aims to investigate the prognostic role of PDZK1 and its association with ER heterogeneity.
Methods: PDZK1 mRNA expression was evaluated [BC Gene miner and the METABRIC cohort] against clinicopathological variables and patient outcome. Full-face BC sections were stained for ER and PDZK1 using immunohistochemistry (IHC). Intra-tumoural heterogeneity was assessed on high definition digital images divided into 4 quadrants and an association between ER and PDZK1 expression was also assessed.
Results: In the METABRIC cohort, PDZK1 mRNA expression was associated with low histological grade, smaller tumour size, good Nottingham Prognostic Index, hormone receptor positivity and HER2 negativity (all p<0.001). High expression of PDZK1 mRNA was also associated with longer BC-specific survival (p<0.001). Using BC Gene miner, PDZK1 mRNA expression was associated with improved patients' outcome (p=0.001). However, PDZK1 at protein level displayed scatter and cluster heterogeneity. Association between ER and PDZK1 expressions revealed that 18.75% of cases showed similar expression percentage and intensity, 43.75% showed an association with percentage, while 6.25% showed an association with intensity. Distribution pattern, cluster and scatter heterogeneity of ER and PDZK1 were also shown to be associated.
Conclusion: This study provides evidence for the prognostic value of PDZK1 mRNA in invasive BC. The heterogeneous pattern of expression of both ER and PDZK1 further establishes the link between PDZK1 and ER; warranting further validation of its clinical utility.

P18

Transcriptomic Profiling of Triple Negative Breast Cancer Identifies SPDYC as a Novel Independent Predictor of Outcome

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Background: The lack of robust prognostic markers for the aggressive triple negative breast cancer (TNBC) leads to unselective treatment of patients. Transcriptome profiling of TNBC identified several subclasses but no single gene was reported as a risk classifier. In this study, using next generation sequencing (NGS) we identified SPDYC as a strong prognostic marker in TNBC. SPDYC is a member of the speedy\Ringo cyclin- dependent kinase (CDK) family, which promotes progression through cell cycle by binding and activation of CDK1 and CDK2. We further examined the clinicopathological significance of SPDYC at the protein level using a large annotated TNBC cohort.
Methods: Supervised artificial neuronal network (ANN) analysis of gene expression was applied on RNA-Seq depository utilising the HiSeq2500 instrument (Illumina, Inc) to identify differentially expressed transcripts with respect to distant metastasis-free interval (DMFS) and breast cancer specific survival (BCSS). Primary TNBC tissue microarrays (n=305) were immuno-stained for SPDYC and correlated with clinicopathological features and patient outcome.
Results: High SPDYC mRNA expression was significantly associated with shorter BCSS (P=0.014) and DMFS (P= 0.018). SPDYC mRNA expression was also independent poor prognostic transcript for BCSS and DMFS (both; P<0.01). High SPDYC protein expression in tumours was observed in 234 out of 305 cases (59%, H score >90). High SPDYC protein expression was significantly associated with poor BCSS and DMFS (both; P<0.01). Cox proportional multivariate analysis revealed that high SPDYC expression is a predictor of shorter BCSS and DMFS, independent of other clinicopathological factors (P=0.015).
Conclusion: Our study identified SPDYC as an independent predictor for prognosis and outcome in TNBC thus can be a potential guide for therapeutic decision

P19

Methods of Nucleolar Assessment in Invasive Breast Cancer and their Prognostic Significance

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Background: Size and number of nucleoli are attracting considerable attention for its potential role in cancer development and progression. Analysis of prominent nucleoli is considered as one of the several important considerations for cancer diagnosis and progression. This study aims to investigate (a) methods of nucleoli scoring of optimal performance (b) their prognostic significance in invasive breast cancer (BC) and (c) the added value of nucleoli scoring to enhance performance in BC grading.
Methods: Hematoxylin and eosin (H&E) stained sections from invasive breast carcinoma cohorts with long-term clinical follow-up split into training (n = 400 cases) and validation (n =1200 cases) set. Four different scoring methods were applied to the training set to identify the most optimal objective and reproducible method associated with a high prognostic value.
Results: Among the four methods used, counting nucleoli in one high power field (hot spot) provided a higher significant association with outcome and highest concordance rate [intraclass correlation coefficient = 0.988]. Prominent nucleoli score in 10 hpf , 5 hpf as well as evaluation of nucleoli in 20% of the tumour were practically less reproducible, more subjective and showed lower concordance rates than nucleoli score in a hot spot. Within the validation set, high nucleoli score were associated with younger age, larger size, higher tumour grade, advanced stage, estrogen receptor negativity, progesterone receptor-negativity and HER2 positivity as well as shorter survival, shorter time to distant metastasis and shorter recurrence-free interval (p =0.000004, p= 0.00001 and p=0.008, respectively). Also adding nucleoli score as a part of BC grading components, grade showed highly significant association with survival (p = 7.48 × 10-13).
Conculsion: Nucleoli scoring in hot spot as an assessment method for nucleoli in H&E stained full-face sections is a reproducible and practical method to predict tumour behaviour

P20

The Prognostic Significant of the Stem Cell Marker ALDH1A1 in Breast Cancer

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Background and aims: Aldehyde dehydrogenase family 1 member A1 (ALDH1A1) has been identified as a cancer stem cell marker in several cancers. In this study, we evaluated the prognostic and biological significance of ALDH1A1 in breast cancer (BC).
Methods: ALDH1A1 was assessed using immunohistochemistry in a large (n=900) well-characterised annotated series of early-stage BC patients with long term follow-up prepared as tissue microarrays. Expression was also characterized using full-face sections of excision specimens (n=28).The associations between ALDH1A1 and clinicopathological parameters and patients outcome as well as with other relevant BC stem cell markers (CD44,CD24, CD133, SOX9,SOX10,EPCAM, and CD133) were determined in the different molecular subtypes.
Result: ALDH1A1 showed homogenous expression in cytoplasmic tumour cells. High cytoplasmic ALDH1A1 expression was associated with poor prognostic features including high grade, high mitotic count, increased nuclear pleomorphism, poor Nottingham Prognostic Index, high nodal stage, and highly proliferative ER+ and Triple negative (TNBC) subtypes and ki67 (P<0.05). High ALDH1A1 expression was significantly associated with poor BC specific survival (BCSS) for 20 year (P=0.000). Based on molecular classes, high ALDH1A1 was significantly associated with poor BCSS (20 year) in highly proliferative ER+ and TNBC subtypes (P=0.04 and 0.002 respectively). High expression ALDH1A1 also predicated the response of chemotherapy in TNBC (P<0.05). ALDH1A1 was positively correlated with the expression of the stem cell markers EPCAM and SOX9 (P<0.05).
Conclusion: ALDH1A1 expression is associated with poor prognostic characteristics in BC particularly in luminal B and TNBC. Moreover, ALDH1A1 can predict the response of the chemotherapy in TNBC.

P21

The Role of Carnitine Palmitoyltransferase-1 in the Progression of Breast Cancer

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Purpose of the study: Tumour cells exhibit a range of metabolic adaptations which fuel tumour growth under conditions of metabolic stress. Carnitine palmitoyltransferase-1 (CPT-1) catalyses the rate limiting step of fatty acid oxidation and is known to be overexpressed in invasive breast carcinoma. It is also suggested that CPT-1 translocates to the nucleus and interacts with histone deacetylases to deacetylate and thereby silence pro-apoptotic genes. The aim of this study was to determine the prognostic significance of the cellular localisation of CPT-1 and the methylation status of the CPT-1 gene in breast cancer patients.
Methods: We measured CPT-1A and CPT-1C expression in a retrospective (MOBCAT) and prospective (BREACAST) cohort, and correlated the nuclear expression level with the methylation status of the CPT-1 promoter, measured using Pyrosequencing, in tumour and background tissue.
Summary of results: Nuclear expression of the CPT-1A isoform is seen in the majority of the tumours. There are differences in the methylation status of CPT-1A between tumours showing low, intermediate and high nuclear expression of CPT-1A, which are associated with differences in the methylation status of ALU repeat elements and IGF-2. There is no differential methylation between tumour and corresponding background tissue. Expression of CPT-1C, the brain isoform of CPT-1, is seen in human breast cancers, and increased cytoplasmic expression is an independent predictor of survival in multivariate analysis, predicting a favourable outcome.
Conclusions: CPT-1 isoforms are predictors of breast cancer progression. CPT-1A expression is associated with changes in the methylation status of the CPT-1A gene which may sit within a network of epigenetically-regulated genes, representing a field-cancerisation effect and possible early event in breast cancer development.
This work was supported by a Pathological Society Grant from the Trainees Small Grant Scheme.

P22

This abstract has been withdrawn

P23

Up-Converting Nanoparticles as a Tool for Histopathological Tissue Evaluation with Multiplexing and Machine Learning Potential

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In the field of histopathology, pathologists diagnose patients by assessing imaged tissues. Even with the pathologists' trained eye, there is a great risk for misdiagnosis. For decades haematoxylin and eosin (H&E) stain has been a standard way to visualise morphology of the cells. It is also common to detect proteins using a DAB chromogenic stain and combine it with a single counterstain to visualise cell nuclei. However, this method suffers from narrow dynamic range, problems with quantitation and difficulties with multiplexing and co-localisation. Fluorescent IHC techniques generate a more quantitative readout but suffer from photobleaching. Here we present that the use of up-converting nanoparticles (UCNPs) allows to overcome problems associated with commonly used imaging techniques. Novel luminescent UCNPs were used together with a prototype instrument to image selected markers, e.g. Her2, in the human tissue. Formalin-fixed paraffin-embedded human colon and breast cancer tissues were sectioned and stained using autostainer. UCNP fluorescence imaging of the human tissue sections was compared with a standard DAB based IHC. Pulsed excitation and gated detection were explored to improve the scanning speed. UCNP and H&E co-staining and co-imaging were also investigated. Images obtained with our novel device clearly show that developed by us antibody-UCNP conjugates can be used to successfully stain human tissues. Brightfield images show that UCNPs are not visible in white light and hence do not interfere with standard tissue evaluation by a pathologist. Additionally, brightfield and luminescent images can be merged to provide better understanding of tissue morphology. Emerging field of UCNPs opens up new possibilities. Staining solutions and a novel device developed by us give hope for more accurate diagnosis by keeping the advantage of H&E staining and combining it, in one image, with luminescent data, ideal for generating ground truth for machine learning algorithms.

P24

Evaluating Steroid Hormone Receptor Profiles in Triple Negative Breast Cancer and their Association with Clinical Outcome

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Triple negative breast cancer (TNBC) accounts for 15% of all breast cancers diagnosed. TNBC lacks expression of the three receptors which are required for response to the current targeted endocrine and biological therapeutics - oestrogen receptor alpha (ERα), progesterone (PR) and human epidermal growth factor receptor 2 (Her2). A targeted treatment option for TNBC remains a significant unmet clinical need in breast cancer care. ERα and PR belong to the steroid subfamily of nuclear receptors, which includes the glucocorticoid receptor (GR), mineralocorticoid receptor (MR), androgen receptor (AR) and oestrogen receptor beta (ERβ). These receptors share functional similarity to ERβ and PR, and so if expressed, targeting these steroid receptors may offer an alternative therapeutic target for TNBC. This study explores the hypothesis that expression of other steroid receptors may predict prognosis in TNBC. The profiles of GR, MR, AR and ERβ were determined in 39 TNBC cases. TNBC tumours are heterogeneous, and steroid receptors have variable expression in different cell types. To control for this, QuPath software[1] was used to specifically identify tumour regions in tissue sections immunostained for each steroid receptor. The proportion of tumour cells expressing each receptor were quantified. Correlations between receptor status and patient survival were determined using clinical data for each sample. Kaplan Meier survival analysis revealed that high GR expression and low AR expression were linked to poor outcome in our TNBC cohort. Pairwise analysis identified no correlation between receptor expression in TNBC tumour cells. Future studies will determine the effect of pharmacological modulation of AR and GR alone and in combination to determine the effect on TNBC cell fate. Through this, we hope to identify alternative therapeutic strategies to treat TNBC.

[1]Bankhead P. et al. (2017) QuPath: Open source software for digital pathology image analysis. Scientific Reports.

P25

Audit of Breast Fine Needle Aspiration Cytology Diagnoses at a Large Teaching Hospital Unit

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Purpose of the study: Fine needle aspiration (FNA) of the breast is used as part of triple assessment of patients presenting via the NHS Breast Cancer Screening Programme (NHSBSP) and the symptomatic breast service. Cases are coded as C1-C5 (inadequate-malignant). An audit was completed to evaluate the accuracy of FNAs at Hospital A.
Methods: Information was collected for all FNAs from July-December 2016 using pathology reports.
Summary of results: Coding was; 49%, 38%, 2%, 1%, 4%, 6% for C1-C5 and other respectively. Twenty-two percent had further testing. Of these, the cytological diagnosis was correct in 86%. The sensitivity and specificity was 65% and 98% respectively. The false-positive rate for C4 was 25% and the PPV for C5 was 100%. Considering only clinically/radiologically suspicious lesions, the rate of inadequate samples dropped to 3%. The rate of C1 diagnoses is larger than documented (7.8-32%) and NHSBSP recommendations. However, this drastically decreases for suspicious lesions. Sensitivity is lower than other reports (71.5-97.5%) and NHSBSP recommendations. Cytological diagnoses and diagnosis of suspicious/malignant lesions show good to perfect accuracy.
Conclusions: Hospital A produces a high number of inadequate/C1 FNAs and has a low sensitivity, but performs well diagnosing malignant lesions. It could be considered that reserving FNA for more clinically appropriate patients could improve standards.

P26

Is Bigger Always Better? X-ray Guided Biopsies in Breast Cancer Screening

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Introduction: In March 2018 the regional breast cancer screening service changed first line X-ray guided biopsies from 12 gauge vacuum assisted biopsy (12G VAB) to 9G VAB. Whilst on the face of it, more tissue seems better as it is more likely to be a representative diagnostic sample, there were two concerns raised (i) With bigger diameter biopsies the standard practice of 4 levels for calcifications may not be sufficient. Further levels may be required on an increased number of cases with implications on turn around times, laboratory and pathologist workload and logistical issues with clinic appointments for final results to be ready. (ii) More sample tissue could result in increased pick up of incidental B3 lesions (e.g. in situ lobular neoplasia (ISLN)) requiring further sampling, increased patient anxiety and radiology and pathology workload.
Methods: We measured the indication for sampling, size of tissue sampled, number and % requiring extra work, type of extra work and the diagnosis and B-codes and compared with a 12G VAB cohort.
Results: The change in diameter of 12G to 9G is 1 mm to 1.5 mm resulting in an increased volume of 193%. In March-November 2018 451 9G VABs were taken. 273 (61%) were taken for calcification and 178 (39%) for mass/asymmetry/nodule/density. For those with calcifications 65 (24%) needed further work, 49 (18%) required additional levels as calcifications not adequately seen initially. 10 biopsies for calcification (4%) ended up as B3 due to the prevalence of incidental ISLN. In comparison, of 273 consecutive 12G VABs for calcification from 2017 40 (15%) required additional levels and 5 (2%) had incidental ISLN.
Conclusions: This study has highlighted that additional levels are needed in a high percentage of calcification cases and that incidental ISLN is not a rare occurrence. However, this study has also shown that the increase in cases requiring additional levels and in incidental ISLN is not as big as concerns initially suspected.

P27

Breast Implant Associated Anaplastic Large Cell Lymphoma (BIA-ALCL): A Local Case Series and the Introduction of a Proposed TNM Staging Criteria in Reporting

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Purpose of study: BIA-ALCL is a relatively new entity, first recognised in 1997, and a TNM staging system has recently been proposed (J Clin Oncol. 2016;34(2):160-8). There is currently no mention of BIA-ALCL in the RCPath datasets for the reporting of breast cancer or lymphoma. We aimed to review cases reported in a large teaching hospital in relation to the proposed TNM staging system, along with evaluation of follow-up data for these patients.
Method: Review of cases of BIA-ALCL reported at Leeds Teaching Hospitals NHS Trust (LTHT) and retrieval of follow-up data from the hospital EPR system.
Results: We identified three patients diagnosed with BIA-ALCL, comprising five pathology specimens, including seroma fluid, capsulectomy specimens, lymph nodes and recurrence samples. These have all been reviewed and diagnosis agreed by both the Histopathology Department and Haematological Malignancy Diagnostic Service (HMDS) provided at LTHT. The patients presented at different pathological stages of the recently proposed TNM staging criteria – including T1, T2 and T4 staging. The T2 case also had nodal disease, and the T4 case has since had further surgery for recurrent disease. Both of these two ladies received chemotherapy post the initial surgery.
Conclusions: Our limited case series supports the proposed TNM staging, and patients with a higher T stage are more likely to have metastatic disease or recurrences requiring on-going follow-up and management. At LTHT we have introduced the TNM staging in our histology reports as this is purported to provide improved prognostic information to our surgical and haemato-oncological colleagues on overall survival, recurrence rates, and the requirement for adjuvant chemotherapy and radiotherapy. We will continue to use the proposed TNM staging system in our reports locally, and would also encourage its use in other centres to allow data collection and comparison going forward. We would also encourage the College to introduce guidelines for assessment and reporting, along with the TNM system in future breast and lymphoma datasets.

P28

Prognostic Impact of Tumour Infiltrating Lymphocytes (TILS), Forkhead Boxp3 (FOXP3) and Cyclooxygenase-2 (COX2) Immunohistochemical Expression in Breast Cancer

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Objectives: To evaluate the prognostic significance of TILs, FOXP3 and COX-2 expression in breast cancer cases and correlating their expression with the clinicopathological data. Background:FOXP3 is a forkhead box transcription factor, playing an important role in the function of immune regulatory T cells (Tregs). Tumour-induced increase in FOXP3+ Tregs represents a potential barrier to attempts at cancer immunotherapy. COX-2 is rapidly induced by mitogenic and inflammatory stimuli. Inflammation-induced COX-2 has been reported to participate in the development and survival of cancers and linked to poor prognosis in a variety of malignant tumours.
Patients and methods: This retrospective study was conducted on 66 Egyptian breast carcinoma (BC) patients. Immunohistochemistry using FOXP3 and COX-2 antibodies was done. Factors of possible prognostic significance and survival data were analyzed.
Results: Higher FOXP3 H. scores in tumour cells were in younger age, low grades of BC and non-triple negative BC cases (p value=0.032 ,0.039 and 0.026 respectively). Higher FOXP3 H. scores in peri-tumoural lymphocytes is more in younger (p=0.010). PR negative cases had higher COX2 H. scores (p=0.025). Median overall survival (OS) was 23 months. There was improved OS with high infiltration by peritumoural infiltrating lymphocytes and high FOXP3 positive intratumoural lymphocytes scores (p=0.018 and 0.014 respectively). FOXP3 positive intratumoural lymphocytes was independent prognostic factors affecting patients' OS (p= 0.043).
Conclusions: High FOXP3 expression in peritumoural and intratumoural infiltrating lymphocytes is considered as a good independent prognostic factor, while COX2 might be considered to have poor prognostic role in studied breast cancer cases.

P29

The Handling and Reporting of Breast Cavity Shave Specimens: A National Survey of UK Practice

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Purpose of the study: Cavity shave (CS) specimens are integral to breast conservation surgery and contribute significantly to breast pathology workloads. We investigated current UK sampling and reporting practices for CS taken during wide local excision (WLE) surgery with reference to existing national guidelines.
Methods: UK breast pathologists were sent an online questionnaire during May – July 2018 followed by 3 reminders.
Summary of results: 110 pathologists completed questionnaires (response rate 20%). 7% of respondents receive CS with all WLE specimens. Targeted CS specimens are more common than 4 standard CS (inferior, superior, lateral, medial). The average CS weight is estimated as less than 10g (59%) and 10-20g (35%). The commonest sampling methods are to embed all tissue (39% always; 47% often; 13% sometimes) or sample areas of macroscopic abnormality only (15% always; 7% often; 34% sometimes). Few shave the new margin only (4% always; 0% often; 6% sometimes). Most pathologists ink the new margin only (54%) and slice perpendicular to this (89%). Commonest definitions for complete local excision for invasive cancer were 1mm (73%); not at ink (14%); 2mm (5%) and for high grade DCIS were 1mm (52%); 2mm (33%); not at ink (6%). 46% of pathologists felt more specific national guidelines for CS would be helpful.
Conclusions: The current sampling, handling and interpretation of cavity shave specimens appears to be inconsistent. Updated national guidance could assist UK breast pathologists and standardise clinical practice.

P30

Adenomyoepithelial Adenosis of the Breast Presenting as a Palpable Mass Mimicking Malignancy on Imaging

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Introduction: Adenomyoepithelial adenosis (epithelial myoepithelial adenosis) is an extremely rare lesion, with asymmetry, irregular margins and calcifications mimicking malignancy on imaging. These biphasic lesions includes spectrum of adenomyoepithelial adenosis, adenomyopithelioma and their malignant counterpart. Here we report a case of adenomyoepithelial adenosis proved to be difficult to diagnose pre-operatively.
Case History: A 46yr old woman presented with right breast distortion and mass like lesion was diagnosed as an intraductal papilloma with florid epithelial hyperplasia in 14G core biopsy and followed by local excision.
Pathology: The lesion was 24mm in diameter had ill-defined margins, nodular architecture with areas reminiscent of intraductal papilloma. In places there were small sized tubules focally giving microcystic appearance, composed of two cell type; an inner layer of ductal cells and outer layer of CK14 and CK5/6 positive myoepithelial cells. Areas with apocrine change and columnar cell change were also present.
Discussion: Our case confirms the rarity of the lesion and illustrates the difficulty to achieve a pre-operative diagnosis. As cited in literature, this is a benign lesion which may progress to malignancy, with few cases that were associated with intraductal carcinoma and malignant myoepithelioma. No correlation was found between tumour size and progression of malignancy. The reported cases were <50mm and predominantly a disease of young women as in our patient. It is important to ensure excision is complete because of the tendency to recur with inadequate excision margins.
Conclusion: This is a rare lesion with limited data showing an association with malignant progression and local recurrence, which needs complete excision with adequate margins. Accurate diagnosis requires immunohistochemistry for basal/myoepithelial markers.

P31

Case Series to Highlight the Clip Site Reaction in Breast Pathology

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Introduction: Biopsy marker placement is an important step in the breast biopsy procedure mostly in screen detected lesions. These marker clips are important to indicate the location of lesions with increased rate of negative margins. The markers are most commonly metallic in various shapes but more recently new types of clips have become available on the market. They may be accompanied with embedding material such as collagen, hydrogel or PVA polymer. This material helps to decrease clip displacement and can aid ultrasound identification of the clip site. Little is known of the changes induced by the modern type markers in breast tissue. Therefore, we decided to review our series of cases demonstrating the clip site reaction in breast tissue and its relation with the targeted lesion removed with both vacuum and surgical type of specimens. In addition, we have looked at the interval of time between clip insertion and tissue reaction.
Case series: A series of breast excisions following a biopsy containing a clip site reaction were collected. These were characterised by a clear cut cavity, which was associated with a multinucleated giant cell reaction and surrounding fibrosis. Some contained jelly-like material.
Discussion: The tissue reaction at the clip site is dependent on the clip material and the interval of time from clip insertion to resection. In our series the typical changes were noted 3–6 weeks after clip insertion. Awareness of tissue changes due to the clip is helpful for several reasons: 1. Accurate assessment of breast specimen resected for small screen detected lesions; 2. Confirming presence of clip site in the pathology report is valued by surgeons as proof of precise localisation; 3. Increases patients satisfaction as it provides reassurance the targeted breast lesion has been removed.
Conclusion: This case series has highlighted the histological appearances of clip sites. This is important in aiding to confirm the correct site of sampling.

P32

A Dedicated Breast Cancer Prevention Clinic: Reducing Breast Cancer Incidence and Identifying Biomarkers Indicative of Therapeutic Response

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Purpose of study: NICE recommends breast cancer preventive therapy for women at high or moderate risk based on their family history. Data from the IBIS prevention trials suggest tamoxifen can reduce the incidence of all breast cancer by 38% and oestrogen receptor (ER) positive cancers by 50%. Anastrazole reduced the incidence of all breast cancer by 53% and ER positive cancers by 58%. Despite these promising data, uptake remains low due to concerns regarding side effects, inadequate consultation time and lack of awareness amongst clinicians. In addition, the mechanism of action of preventive therapy is poorly understood – not all women receive the protective effect. In the IBIS I trial, reduction in mammographic density (MD) after one year of tamoxifen was associated with protective effect in a proportion of women. There is an urgent need to determine novel biomarkers indicative of therapeutic response and to develop novel prevention associated with fewer side effects.
Methods: A dedicated breast cancer prevention clinic was established within the family history service at our NHS breast unit, offering half hour appointments to discuss the evidence for available preventive agents and potential side effects. Those keen to proceed were offered the option to provide serial blood samples and an additional mammogram after 6 months, to assess change in MD and identify serum biomarkers indicative of therapeutic response.
Results: This is the first dedicated breast cancer prevention clinic in the UK embedded within routine NHS services. 198 women have attended over the past three years. Uptake of preventive therapy has been substantially higher (52%) than those reported in previous studies (10%). To date, 20 women have enrolled in the biomarker study.
Conclusions: Uptake of preventive therapy can be substantially improved if women are adequately counselled regarding potential risks and benefits. The clinic has provided a valuable resource for future preventive biomarker discovery.

P33

Two Cases of Metaplastic Breast Carcinoma with Osteosarcomatous Differentiation

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We describe two cases of metaplastic breast carcinoma with osteosarcomatous differentiation diagnosed at our Trust.
The first case was that of a 61 year old woman who underwent mastectomy for a large fungating tumour which had replaced most of the breast tissue. On gross pathological examination the tumour was solid, white, with hard areas and areas of necrosis. Histology showed an atypical spindle cell tumour with areas of malignant chondromatous and osteogenic differentiation. There was prominent mitotic activity, with large areas of necrosis and ulceration of the overlying epidermis. The appearance and immunoprofile was of a metaplastic carcinoma with heterologous differentiation (chondromatous and osteogenic). One of the lymph nodes in the axillary clearance showed metastasis from the main tumour.
The second case was that of a 54 year old woman who underwent mastectomy following a biopsy which had been reported to show osteosarcoma. The nipple was depressed overlying a 43mm tumour. The tumour was haemorrhagic and friable upon specimen slicing. Histology showed a metaplastic carcinoma with predominant heterologous differentiation (osteogenic sarcoma) and a minority component of grade 3 invasive ductal carcinoma with associated high grade ductal carcinoma in-situ. No evidence of malignancy was seen in palpable nodes taken from the axilla.
Metaplastic breast carcinoma is a rare and generally aggressive form of breast cancer and both of these patients died within 12 months of diagnosis. Both cases showed osteosarcomatous differentiation. The main differential diagnoses of phyllodes tumour and primary breast sarcoma are discussed alongside important pathological prognostic factors in these types of cases.

P34

A Study of Symptomatic Breast Needle Core Biopsies Categorised as B3 Over a Period of Three Years with Review of Surgical Follow-Up and Comparison with Outcomes from Breast Screening Programme Cases

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Purpose of the study: Breast needle core biopsies within the B3 category represent a heterogeneous group of lesions with a positive predictive value (PPV) for malignancy ranging from 9.8% to 35.1%. Much of the data is drawn from cohorts of screening cases; this study provides insights from symptomatic patients by comparing the range of cases categorised as B3 within our institution with those previous studies and by reviewing the subsequent histology.
Methods: Breast needle core biopsies performed at our institution from 2012 to 2014 were identified from the pathology records. Biopsies that had been reported as B3 were identified by reviewing the reports. Cases reviewed as part of the NHS Breast Screening Programme were excluded. Clinical and radiological indications were recorded from the details provided on the pathology request forms. Follow-up in these cases was recorded by reviewing subsequent histology reports or electronic patient records if there was no further surgical intervention. The slides for the initial needle core biopsies and the subsequent excisions were retrieved from file and reviewed.
Summary of results: B3 cases represented 3.97% of all symptomatic breast needle core biopsy cases. Papillary lesions made up the highest proportion at 46%; these were upgraded to DCIS or invasive carcinoma in 7.9% of cases. Atypical intraductal epithelial proliferations represented 19% of cases but were upgraded to DCIS or invasive carcinoma in 36% of cases. 17% of B3 cases were fibroepithelial proliferations, of which 37% were phyllodes tumours, however, these were rarely malignant. Lobular in situ neoplasia and radial scars made up the remainder of B3 diagnoses.
Conclusions: Our study has shown a proportion of B3 cases (3.97%) and PPV for malignancy (8.75%) at the lower end of the ranges in screening-identified cases (3-9.2%, PPV 9.8-35%) reflecting differing populations and pathology. Atypia was associated with increased likelihood of upgrade to malignancy on excision.

P35

Clinicopathological Characterization of Breast Cancer in Africa: A Tissue Microarray Study of Two African Cohorts

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Introduction: While breast cancer (BC) is a major health issue in Africa the disease, is still poorly understood. We aim to study the presentation and molecular profile of African breast cancer as compared to the European counterpart.
Methods: Two breast cancer cohorts representing North Africa (Egypt, n=84) and West Africa (Nigeria, n=88) were assembled into tissue microarrays. Sections were stained for hormone receptors, Androgen receptor (AR), CK14, Ki67, GATA3, and PDL-1 and scored by two pathologists.
Results: 46.4% of Egyptian and 52.3% of Nigerian patients were under the age of 50. 64.2% of the Egyptian cases showed luminal and 23.5% a triple negative phenotype. 57% Of Nigerian patients were triple negative and only 37.5% of them were of luminal phenotype. PDL-1 showed positive expression in 53.7% of Egyptian and 15% of Nigerian tumours. AR was positive in 38.5% and 47% of Egyptian and Nigerian cohorts respectively. GATA3 was positive in 78.2% of Egyptian and 10.3% of Nigerian cases. 18.7% of Egyptian and 24.3% of Nigerian patients were of the basal phenotype.
Discussion and conclusion: Around half of African breast cancer present at a young age (<50yrs). Compared with the Caucasian BC, Breast cancer in Nigerian was predominantly of the triple-negative phenotype. GATA3 was remarkably low in the Nigerian cohort. The proportions of luminal cancers, as well as AR positive cancers, were low in the two African cohorts. PDL-1 was expressed in tumour cells within a larger proportion of tumours compared with the known Caucasian data. Our data highlight differences in the presentation, immunophenotype and molecular profile between the African and European breast cancer. We are investigating in detail the role of tumour microenvironment and PDL-1 in the African tumours.

P36

Moving to a Digital Pathology Supraregional Germ Cell Tumour Service

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Patients with testicular cancer are managed in supraregional networks serving a population of 2–4 million, seeing at least 100 new patients/year. Patient management includes the review of diagnostic glass slides from local sites for the supraregional MDT. Digital reporting of testis cases was piloted as part of a move to full digitisation of the cellular pathology laboratory at our institution. Feasibility of digital referral from the local centres to the supraregional site centrally was assessed, avoiding the need for postage of slides. Four Philips slide scanners were deployed in 2018 (2 in the central site and 1 in each of two peripheral centres). The service was evaluated as a traditional glass-slide based service in preparation for the switch to digital. A central site validation of digital reporting SOP based on the RCPath guidelines was created. Two specialist germ cell tumour pathologists reported 57 cases on glass slides from 6 sites (benign and malignant). The number of slides ranged from 3–75/case (mean 17), with an average of 13 H&E slides/case. 36 cases had IHC. Mean reporting was 18 minutes/case (range 7–49). The retrospective digital training cases (n=26) were rated on a Likert scale from 1–7 (not at all-very confident). Confidence in digital diagnosis for one pathologist was 4–7 with 23 cases rated 6 or 7. Glass was preferred in 6/26 cases. In 20/26 cases there was no preference between digital and glass diagnosis. Searching for small foci of germ cell neoplasia in-situ was reportedly easier on glass. Prospective validation started with the first digitally reported case of Seminoma in November 2018. The feasibility of running a fully digital supraregional germ cell tumour service across our region shows great promise. Digital reporting of testis cancer cases is feasible and has the potential to improve the availability of slides for supraregional review.

P37

Comparison between Glass and Digital Scans for Maximum Cancer Core Length (MCCL) Measurement and Prostate Cancer Gleason Scoring

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Purpose of study: The use of digital pathology has the potential to improve patient care. We aimed to compare the estimation of MCCL and Gleason sum between glass and digital scans in a cohort of patients at at London hospital.
Methods: 30 patients with Gleason 3+4 (n+15) and 4+3 (n=15) from the PROMIS study were included. A total 192 slides were scanned using NanoZoomer-SQ digital slide scanner. Using NDP.View 2 software each core compromised by cancer was manually measured in mm, if the core was not straight the measurement was added following the shape of the core. For each patient each pathologists provided an overall Gleason sum, blinded to the original glass diagnosis. Wilcoxon matched pairs signed rank test was performed using R.
Summary of results: 426 cores were analysed, the average MCCL was 9.53 mm (5-15) and digital MCCL (dMCCL) was 9.88 (5.01-15.7). In 20 cases the dMCCL was higher than the MCCL ($p = <0.0001$). In 10 cases the MCCL was higher than the dMCCL ($p=0.002$). When taking into account a difference of more than 1 mm between the two measurements 12 were had a higher dMCCL and 7 a higher MCCL. It is important to note that the difference between the two measurements was less than 2 mm in 22 cases (73%), confirming good accuracy for the glass measurement. When estimating the overall Gleason sum per patient, when examining the digital images one patient was downgraded from 4+3 to 3+4 by both pathologists. One patient was downgraded by the more experienced pathologist, and two were upgraded by the less experienced pathologist.
Conclusions: Digital scoring is comparable to glass examination of prostate biopsy specimens. With the possibility of more in depth analysis of digital images and potential reclassification of patients.

P38

Extra-Mammary Paget's Disease of the Penis with Underlying Poorly Differentiated Adenocarcinoma: A Case Report

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Extra-mammary Paget's disease (EMPD) is a rare and slow growing neoplasm and is seen much less often than mammary Paget's disease.1 EMPD presents in regions with a large number of apocrine glands and the ano-genital region is the most common site to be involved. It can be associated with an underlying carcinoma or distant metastases. Patients commonly present with pruritus and a slow growing crusting, scaling, non- healing lesion.2 The symptoms can mimic benign inflammatory skin conditions leading to a misdiagnosis. EMPD is more commonly seen in the elderly with more cases reported in the vulval region of females. Cases involving men are much less common. EMPD in men usually involves the scrotum and involvement of the penis is rare.
We present a case report of an elderly male whose diagnostic biopsy was initially reported as a poorly differentiated squamous cell carcinoma. The patient underwent a penectomy. Histology demonstrated EMPD with characteristic pale cytoplasm and large pleomorphic nuclei with an underlying poorly differentiated adenocarcinoma. Penile involvement with EMPD and associated poorly differentiated adenocarcinoma is an extremely rare entity with few cases reported in the literature.4 Surgical excision is recommended but EMPD is difficult to manage due to the multifocal nature of the disease.5,6 When associated with invasive malignancy reports suggest there is up to a 46% higher risk of mortality highlighting the need for the timely diagnosis and management.6,7

P39

Concordance and Discordance of Gleason Scores in Prostate Biopsies with Matched Radical Prostatectomy Specimens

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Background: Prostate adenocarcinoma is the most common male cancer in the UK and accounts for 26% of new cancer cases (1). The Gleason Score offers key prognostic information, directs the management of patients with prostatic adenocarcinoma (2). Research has shown that discrepancies exist between the Gleason Score of the preoperative prostate biopsies (PB) and that of radical prostatectomy (RP). Prostatic cancer characteristically shows histologic heterogeneity which contributes to the grading discrepancy between PB and RP (3).
Methods: Data was retrospectively collected for RP cases performed in University College London Hospital (UCLH) from 1st January to 18th March 2019 using Co-Path Database. The Gleason scores of RP and the matched PB were reviewed and compared.
Results: 134 cases of RP were performed at UCLH during the period. In 73 cases (54.5%), the Gleason score was concordant with the preoperative Gleason Score, while 61 cases (45.5%) were discordant with the original biopsy Gleason Score. Of the 54 cases, 50 cases were upgraded and 11 cases were downgraded from the original Gleason Score.
Conclusion: A significant proportion of prostatic adenocarcinoma is under-graded in PB compared to RP, which may impact the patient management and clinical outcome.
References: 1. Cancer Research UK, https://www.cancerresearchuk.org/health-professional/cancer-statistics/statistics-by-cancer-type/prostate-cancer; Accessed March 2019. 2. Epstein JI, Egevad L, Amin MB, Delahunt B, Srigley JR, Humphrey PA. The 2014 International Society of Urological Pathology (ISUP) consensus conference on Gleason grading of prostatic carcinoma. The American journal of surgical pathology. 2016 Feb 1; 40(2):244-52. 3. Evans SM, Bandarage VP, Kronborg C, Earnest A, Millar J, Clouston D. Gleason group concordance between biopsy and radical prostatectomy specimens: A cohort study from Prostate Cancer Outcome Registry–Victoria. Prostate international. 2016 Dec 1;4(4):145-51.

P40

This abstract has been withdrawn

P41

An Unusual Renal Pelvis Tumour

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Large cell neuroendocrine carcinomas (LCNEC) are high grade malignancies which display characteristic morphological features and immunohistochemical evidence of neuroendocrine differentiation. They occur across a range of organ systems but are extremely rare in the upper urinary tract, with very few reported cases in the literature. We present a case of an 80 year old man who presented with right iliac fossa pain and haematuria. Imaging showed an obstructing mass within the right renal pelvis. Subsequent nephrouretectomy revealed a polypoid renal pelvis tumour composed of sheets of medium cells with scant cytoplasm and elongated, granular nuclei. Marked nuclear pleomorphism was present, with scattered multinucleate giant cells, and the tumour showed abundant mitoses, apoptotic bodies and coagulative necrosis. Focal rosette formation was also identified. Immunohistochemistry revealed evidence of neuroendocrine differentiation, with the tumour cells staining positively for CD56, Chromogranin and Synaptophysin. The proliferation index was measured at >90%. Dot positivity was seen with AE1/3 and CK7. Focal Gata3 positivity was also identified. These morphological appearances and immunohistochemical profile are consistent with a large cell neuroendocrine carcinoma of the renal pelvis. This diagnosis carries a poor prognosis.

P42

The Relationship Between the Tumourigenicity-Promoting Function of C-FABP and its Fatty Acid-Binding Ability in Prostate Cancer

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To study the possible relationship between the tumourigencity-promoting activity of cutaneous fatty acid binding protein (C-FABP) and its fatty acid-binding capability, mutations were generated on the fatty acid-binding motif of the C-FABP cDNA. The wild type and mutated C-FABP cDNAs were transfected respectively into the LNCaP prostate cancer cells, which do not express C-FABP prior to the transfection, to generate a clone expressing a wild type C-FABP which can bind to fatty acids; and two clones expressing mutated C-FABPs which are less capable of binding to fatty acids. The increased wild type C-FABP expression in LNCaP cells significantly increased cell proliferation, invasiveness, and tumourigencity in vitro, whereas, the increased expression of both mutant forms of C-FABP did not significantly change these characteristics. When inoculated in nude mouse, 7 out of 8 mice produced tumours in the wild type group, with an average tumour weight of 342.9±144.8mg. In single mutant group, 4 out of 8 mice produced tumours with an average tumour weight of 217.5±69.5mg. Whereas in the double mutant and control group, only 3 out of 8 mice produced tumours with average sizes of 59.33±19.0mg and 46.7±15.3mg, respectively. The increased expression of wild type C-FABP, which is able to bind fatty acids, produced 4 more tumours and significantly increased the tumour size by 7.3- fold, comparing with the control. In contrast, increased expression of the mutant C-FABP, which only has a similar ability of binding fatty acids to the control cells, produced the same number of tumours with similar sizes with those produced by the control cells. Our results suggest that the biological ability of C-FABP to promote tumourigencity of the prostate cancer cells depends on its ability of binding to fatty acids. Thus C-FABP may facilitate cancer development through transporting fatty acids into cells.

P43

The Immunohistochemical Profile of Urothelial Carcinoma Based on CK5 and CK20 Expression, Impact on Prognosis and Patient Outcome

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Background: In Egypt, bladder cancer represents the third most common cancer in both sexes with a male predominance, which makes identification of valuable predictive and prognostic markers mandatory.
Purpose of study: The aim of the present study is to investigate the pattern, distribution and significance of the immunohistochemical expression of CK 5 and CK 20 as surrogate markers for basal-like and luminal-like subtypes, respectively in urothelial carcinomas.
Methods: Using the standard immunohistochemical (IHC) technique, CK 5 and CK 20 expression in 90 primary bladder carcinoma was assessed.
Summary of results: Four groups of urothelial carcinoma were identified according to the results of CK5 and Ck20, CK5+/CK20+ group comprised 42%, CK5-/CK20- group comprised 13%, CK5+/CK20- group comprised 39% and CK5-/CK20+ group comprised 6% of the studied cases. The CK5-/CK20+ group showed the shortest disease free survival (28.4 months), advanced stage (80%) and high grade(100%) among the 4 groups.
Conclusions: Immunohistochemical assessment of both CK 5 and CK20 can divide urothelial carcinomas into different prognostic categories with CK5-/CK20+ as the worst profile.

P44

Comparative Study between Immunohistochemical Expression of Erythroblastosis E26 Rearrangment Gene (ERG) and Membrane Associated Guanylate Kinase (MAGI-2) in Prostatic Carcinoma

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Prostate carcinoma (PC) is the commonest non cutaneous cancer in men in the USA. In Egypt, it represents 61.63% of all male genital tract malignancies. The relative lack of specificity and sensitivity of p63 and AMACR in morphologically equivocal cases resulted in significant over and under -diagnosis of PC. Thus it has been important to search for other diagnostic markers with high sensitivity and specificity. The aim of this study is to evaluate the diagnostic and probable prognostic value of immunohistochemical expression of MAGI-2 in comparison to ERG in prostatic adenocarcinoma (PAC). This prospective study included 56 cases of PAC and 29 cases of nodular prostatic hyperplasia (NPH). Results revealed significant difference between NPH and PAC regarding ERG expression ($p< 0.001$). While all NPH cases were negative for ERG, 29 of studies cases of PAC (51.8%) were positive. Thus, the diagnostic power of ERG in PAC revealed 53.6 % sensitivity and 100% specificity. Furthermore, it was noted that ERG expression was statistically significantly related with Gleason grading in PAC ($p=0.005$) and with Ki67 expression ($p<0.001$). Regarding MAGI-2 expression there was a highly statistically significant difference between NPH and PAC ($p<0.001$). While 51 cases (91.1%) of PAC were positive, positive MAGI-2 expression was detected in only 5 cases (17.2%) of NPH. Thus, the diagnostic power of MAGI-2 in PAC revealed 91.1 % sensitivity and 86.2% specificity. Regarding probable prognostic role of MAGI-2, there was no statistically significant relation with any histopatholoic prognostic parameters including Ki67 expression. In conclusion, this study recommend to add both ERG and MAGI-2 to the diagnostic panel of PAC due to different sensitivity and specificity

P45

Prognostic Significance of Regulator of Cullins-1(ROC-1), Carbonic Anhydrase IX (CAIX), and P21 in Bladder Cancer

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Objectives: To investigate the role of Regulator of Cullins-1(ROC-1), in regulating cell cycle via P21 in bladder cancer expressing the hypoxic marker carbonic anhydrase IX (CAIX)and how can they act in developing and progressing this disease.
Background: Bladder cancer is one of the most common malignancies in Egypt representing about 6.94% of cancers which makes identification of valuable predictive and prognostic markers mandatory. ROC-1 is a key subunit of Cullin-RING ligases (CRL) which plays an important role in the ubiquitination of cell cycle-related proteins or other proteins like p21. CAIX has gained much interest as a hypoxic marker.Hypoxia is an important regulator of gene expression and resistance to chemotherapy and radiotherapy.
Patients and methods: Using the standard immunohistochemical technique, ROC-1, CAIX and p21expression in 80 primary bladder carcinoma and in 15 normal bladder specimens were assessed . The carcinoma cases included 50 cases with muscle invasive bladder cancer and 30 non-muscle invasive bladder.
Results: ROC-1 and P21 overexpression were significantly associated with muscle invasive, (p=0.04, p=0.001) and high grade (p=0.04,p=0.02) bladder cancer respectively. While CAIX was significantly expressed in primary bladder cancer than normal specimens (P= 0.01) and overexpressed in high grade urothelial carcinoma (P=0.01). CAIX expression was predominant in perinecrotic areas than away from necrosis (P=0.014).
Conclusions: ROC-1,P21 and CAIX expression could be a promising potential predictive biomarker for identifying patients with poor prognostic factors in bladder cancer which may serve as potential target for cancer therapy. ROC-1 expression in early non-invasive BC could help in predicting the progression to invasive type.

P46

Implementation of BxChip for Prostate Core Samples in Histology

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Purpose of the study: This was a quality improvement project which involved an innovative method of handling and processing prostate core biopsies, with the primary aim of reducing the risk of transposition errors. The conventional method involves multiple steps requiring handling of the fragile cores, with significant risk of fragmentation, tissue loss and significantly, tissue transposition at every step. The implementation of the Bx Chip was undertaken in order to reduce this risk.
Method: The Bx Chip is a patented sectionable matrix which allows the cores to be placed into a multiplexed grooved matrix constructed from a protein gel by the biopsy taker in clinic itself. The prostate cores are placed into specific channels of the BxChip in grooves between dotted bands with tissue orientation enabling markers. This matrix is inserted into a pre-labelled cassette and directly processed without any further manipulation. The BxChip replaced the conventional cassettes used in prostate core biopsies in our laboratory following a pilot study.
Summary:
Implementation of this new technique has led to multiple advantages, which include - significantly reduced the risk of sample mix-up and transposition; minimal fragmentation of biopsies and tissue loss of potentially malignant samples obtained from an invasive procedure; reduced handling of samples in the laboratory with reduced time taken in processing; reduced turnaround time for reporting due to better presentation of biopsy cores and reduced slide numbers; it provides an ideal template for digital pathology; less space needed for archiving as fewer blocks (2) and slides (4) produced; ideal for future research projects as more tissue is left within paraffin block; cost neutral to implement. No significant risk was identified.
Conclusion: In our experience, this new method has provided multiple benefits in the processing and reporting of prostate core biopsies compared with the conventional method of processing

P47

Clinicopathological Features of Incidental Prostate Cancer in Cystoprostatectomies

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Introduction: Incidental prostatic adenocarcinoma (CaP) in radical cystoprostatectomy specimens is frequently encountered, its incidence ranging from 15 to 54%1,2. Our aim was to determine how many clinically significant incidental CaPs were identified at our centre 2016–2018 to assess whether complete prostate sampling is justified.
Material and methods: Cystoprostatectomies received at our centre 2016–2018 were retrievedand cases with concomitant CaP were analysed fortumour diameter, Gleason grade, stage andmargin and nodal status. Cases with a previous diagnosis of CaP were excluded.
Results: Incidental CaP was identified in 61/135 (45%) cases. Gleason grade was 3+3 in 17 (28%), 3+4 in 32(52%), 4+3 in 6(10%), 4+4 in 2 (3%) and 4+5/5+4 in 4 cases (6%). 48/61 of cases were organ confined (pT1a-pT2c, 79%), 11 cases showed extraprostatic extension (pT3a, 18%) and 2 cases had SV involvement (pT3b, 3%). Margin involvement was present in 4 cases (6%), and one had nodal spread (1.5%).18/61 cases (30%) had features indicative of clinically significant disease, as defined as showing any of the criteria of predominant Gleason pattern 4, pT3a/b status, positive surgical margin or nodal involvement. Additionally, 13/61 further organ confined cases (21%)of Gleason 3+3/3+4 cases had a tumour diameter >10mm.
Conclusion: In our institution a significant proportion of incidental CaPs diagnosed in cystoprostatectomies possess features indicative of clinically significant disease. This supports complete rather than partial sampling of the prostate, previously shown to improve the detection of clinically significant CaP3.The clinicopathological features may however evolve with time due to increasing emphasis on early detection of CaP.

P48

A Study on the Value of Frozen Section Examination of Margins in Penile Resections (2014–2016)

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Introduction: We noticed an increase in the number of penile resection specimens received by our unit. These resections are associated with multiple frozen sections (FS) of margins. We wished to examine the utility of this.
Aim: To determine the caseload associated with FS examination of penile margins, the rate of positivity, false positive and negative rates and comparison with resection specimen and risk of local recurrence.
Method: We assessed our records to identify all penile cancer cases with associated FS examination over a three year period. We compared the FS report with subsequent paraffin sections and local recurrence data for this cohort.
Results: A total of 142 FS on 33 patients were studied (27 in 2014, 25 in 2015 and 90 in 2016 representing 6, 6 and 21 patients respectively) with increased technical and consultant hours noted over the study period. An average of 4 FS were submitted per patient (range 2–6) representing 34% of all uropathology FS in 2014 compared to 86% in 2016. Only one patient had a positive FS (0.7% positive rate) with 100% concordance rate to paraffin blocks. Comparing the results of the FS with subsequent margins in the resection specimen, there were 6 cases with negative FS with an involved margin status of which two developed local recurrence in the follow up period.
Conclusions: Most FS specimens were negative in our cohort. Replacement of random sampling of resection margins with targeted sampling of periurethral/fascia tissue in larger tumours of higher stage is likely to be a more precise and appropriate.

P49

The Matter of Margins: An Audit on the Reporting of Margin Status in Radical Prostatectomies

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Introduction: Positive surgical margins in radical prostatectomy cases are of prognostic significance as well as providing feedback on surgical technique. The Royal College of Pathologists' core data items for the reporting of margin status include margin positivity, the extent of involvement, location and whether involved margins are intraprostatic or extraprostatic. This audit aimed to assess adherence to the core data items in reporting.
Methods: All radical prostatectomy cases for prostate cancer reported between 1st August 2017 and 31st July 2018 were identified and audited on adherence to the following criteria within reports: 1) Margin status; 2) Extent of margin positivity measured; and 3) Location, including whether involvement was intraprostatic or extraprostatic. A standard of 100% for all criteria was used.
Results: 168 radical prostatectomy cases were identified. Margin status was reported in 100% of cases, revealing 64 cases (38.1%) to have positive margins. The extent of margin involvement was measured in 53 cases (82.8%). The location of margin involvement was reported in 100% of cases, whilst intraprostatic margin positivity was reported specifically in 19 cases (29.7%) overall.
Conclusions: The standard was not met for reporting the extent of margin involvement. Most reports omitting this involved basal or multiple margin positivity (72.7%). Intraprostatic margin positivity, which may be reflective of surgical technique, was formally commented upon in only 45.4% of cases with positive radial margins and this is an area which requires further focus within our reports.

P50

An Audit of Renal Tumour Needle Core Biopsy: Single Centre 10 Year Consecutive Case Series

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Purpose: An audit of histological diagnoses, ancillary tests and correlation with final resected diagnoses for a consecutive series of needle core biopsies of renal masses underpinning Scottish Quality Performance Indicator (QPI) for Histological diagnosis.
Methods: All renal tumour needle biopsies were identified by departmental computer search for the period January 2008 to June 2018. Pathology reports were assessed for diagnosis and use of ancillary diagnostic techniques including immunohistochemistry (IHC) and whole chromosome flourescent in situ hybridisation (FISH). Concordance of biopsy diagnosis with final diagnosis was assessed for those cases subsequently undergoing partial or radical nephrectomy.
Results: 220 separate cases were identified, with the following histological diagnosis: 142/220 (65%) biopsies were malignant, 40/220 (18.1%) benign. The remaining 38/220 (17.2%) were not diagnostic. 28.2% of cases (62/220) underwent subsequent surgical resection. The concordance between cases diagnosed as malignant on biopsy which were then resected was 100% (62/62). There were no benign-malignant reversals. Specific biopsy diagnoses were as follows: 40.4% clear cell RCC; 12.3% oncocytomas; 10% papillary RCC; 4.5% invasive urothelial cell carcinomas; 1.8% RCC not otherwise specified. 3.6% each for Chromophobe RCC, metastatic carcinomas from other sites and angiomyoliypoma. All resected clear cell RCCs were reported with Fuhrman/ISUP grade. IHC aided diagnosis on 56.4% of cases and FISH was requested on 14.1% of biopsies.
Conclusion: Our single institution experience demonstrates that histology of needle biopsies of renal tumour masses in conjunction with ancillary tests, where appropriate, is an effective diagnostic tool that plays a significant role in informing treatment decisions. The QPI for histology in renal cancer are consistently achieved.

P51

Renal Biopsy Findings in Recurrent POEMS Syndrome

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Glomerular disease is an uncommon manifestation of POEMS (Peripheral neuropathy, Organomegaly, Monoclonal protein and Skin changes) syndrome found in about 4% of cases. We present here the glomerular changes with recurrent POEMS syndrome after apparently successful treatment. A 53 yr old male had previously presented with peripheral neuropathy and was found to have POEMS syndrome. At presentation he was found to have proteinuria, a renal biopsy showed glomeruli with diffuse endothelial swelling. He received an autologous stem cell transplant resulting in complete remission. On routine follow up after 11 years he was found to have developed renal impairment and significant proteinuria, this was investigated by a second renal biopsy. The biopsy showed 20% of glomeruli were globally sclerosed. All other glomeruli were enlarged and showed a diffuse proliferative glomerulonephritis appearance with lobular architecture and both endocapillary and mesangial proliferation. Focal GBM splitting was seen on silver stain. No deposit was identified by IHC or EM. Electron microscopy showed extensive endothelial swelling with luminal occlusion. On further investigation circulating vascular endothelial growth factor (VEGF) was found to be significantly elevated and although no other manifestations of the syndrome were present this was considered evidence of a relapse of his POEMS syndrome. He was subsequently treated with lenalidomide with significant improvement in renal function. Only one previous case of a glomerular relapse of POEMS has been reported, this showed similar renal histology with endotheilal swelling and hyperplasia without either thrombosis or inflammation; this renal pathology is thought to be relatively specific to POEMS and a result of the circulating cytokines, particularly VEGF.

P52

A Comparative Audit of Current and Previous Renal Biopsy Diagnoses

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Primary diagnosis and request form data from May 2017–Nov 2018 of medical native renal biopsies received in a tertiary centre were audited and compared with previous audits from 1990, 2003 and 2011. There were 159 biopsies (M: 95, F: 64), ages range from 1 to 88 (median: 53). All cases had a documented clinical reason for biopsy: proteinuria (nephrotic range and sub-nephrotic) and suspected AKI being the most common. IHC was available in 157, and EM in 150. The primary diagnoses were: IgA nephropathy: 25 cases (16%); FSGS: 25 cases (16%); membranous GN: 12 cases (8%); minimum change and immune complex GN: 10 cases (6%) each; and vasculitis: 8 cases (5%). A range of less common diagnoses were recorded including acute interstitial nephritis, lupus, diabetes, and amyloid. No case was reported as normal or failed in this period. The distribution of diagnoses showed differences to the distributions seen in previous similar audits undertaken in the years 1990, 2003 and 2011. There was a reduction in the incidence of diabetic nephropathy, thin membrane nephropathy and IgA nephropathy; this reduction likely reflects changes in the criteria for biopsy. More puzzling was a reduction in the incidence of vasculitis and membranous GN as clinical suspicion for these diseases are strong indications for biopsy. It is possible that this represents true changes in the incidence of these disease. In contrast there was increase in the incidence of FSGS; the majority of the FSGS cases were not associated with nephrotic syndrome and while several cases were biopsies of suspected relapsed vasculitis this does not fully explain this increase. In summary the audit demonstrates consistent and complete examination of medical renal biopsies and a wide range of primary diagnoses, but with changes in the frequency of some diagnoses over time that require explanation.

P53

Pleomorphic Rhabdomyosarcoma of the Uterus: A Molecular and Immunohistochemical Analysis of a Rare but Aggressive Gynaecological Malignancy

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Purpose of study: Pleomorphic rhabdomyosarcoma (PRMS) of the uterus is an aggressive sarcoma associated with poor prognosis. Cases are rare and treatment regimens are not well established. The aim of this study was to identify cases of PRMS at our hospital and perform molecular and immunohistochemical tumour analysis.
Patients and methods: Two cases of PRMS of the uterus were identified at our institution over a ten year period (2009–2019). The women were aged 84 and 68 years old respectively. The patients underwent total abdominal hysterectomy and bilateral salpingo-oophorectomy. We reviewed the clinical data and histology of both cases. An immunohistochemical panel was performed on selected tumour blocks. Tumour DNA was sequenced using a targeted next generation sequencing (NGS) panel consisting of 50 genes.
Summary of results: Both tumours were positive for desmin and myogenin. They were negative for CD117 and PDL1. Analysis of the tumour immune response demonstrated a dense tumour infiltration by CD68+ macrophages. A pauci-immune response from CD4 and CD8+ T cells was noted. Somatic DNA sequencing failed in one case due to excessive fixation. In the other case, two mutations were identified: a PIK3CA p.(His1047Arg) activating mutation and a TP53 p.(Arg248Gly) loss of function mutation.
Conclusions: We present the first evidence of an actionable mutation in uterine PRMS. P13K inhibitors are currently a focus in multiple clinical trials for other malignancies. Activation of the P13K/AKT/mTOR pathway by mutations in the PIK3CA gene has been documented in other subtypes of rhabdomyosarcoma at sites outside of the gynaecological tract; blockade of this pathway has revealed promising evidence for further therapeutic strategies. Furthermore, this study shows for the first time that uterine PRMS is a macrophage-rich tumour. If this correlates with poor survival, as in many other cancer-types, the therapeutic potential of macrophage inhibitors could be explored.

P54

Exceedingly Rare Pure Large Cell Neuroendocrine Carcinoma of the Endometrium: Report of Two Cases

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Introduction: Less than 1% of endometrial carcinomas are neuroendocrine carcinomas (NECs). Most endometrial NECs are small cell neuroendocrine carcinomas, with ~90 cases reported to date. Endometrial large cell neuroendocrine carcinoma (LCNEC) is rare with <20 cases reported to date. LCNEC has a poor prognosis with early hematogenous/lymphogenous metastasis.
Case presentation: We present two cases of endometrial LCNEC.
Case 1: A 64 year old hospital secretary presented with postmenopausal bleeding. Ultrasound revealed a 3 x 4 cm intrauterine mass. Hysteroscopy confirmed a fundic mass with necrotic edges. The biopsy and subsequent total abdominal hysterectomy, bilateral salpingo-oopherectomy (TAHBSO) and lymphadenectomy revealed a 60 mm FIGO stage 2 endometrial LCNEC with extensive lymphovascular space invasion.
Case 2: An 85 year old lady with Alzheimer's disease presented with postmenopausal bleeding. Transvaginal ultrasound suggested an endometrial tumour invading >50% of the myometrium. MRI pelvis revealed a grossly enlarged uterus measuring 17x 10cm with a distended endometrial cavity containing a large and poorly enhancing soft tissue mass. The patient underwent a hysteroscopic biopsy followed by TAHBSO and omental biopsy. The histology showed a 150 mm FIGO stage 2 endometrial LCNEC with extensive lymphovascular space invasion.
Discussion: Pure endometrial LCNEC is rare and diagnostically challenging. As most endometrial NECs are admixed with other endometrial carcinoma subtypes; extensive sampling is essential to confirm the rare pure NECs. The differential diagnosis includes a solid component of endometrioid carcinoma, undifferentiated carcinoma, a sarcomatoid component of carcinosarcoma or primitive neuroectodermal tumour . Pathologists should be aware of the histological features of these rare tumours and the potential pitfalls with immunohistochemistry.

P55

This abstract has been withdrawn

P56

Leydig Cell Tumour of the Testis and Ovary with Rare Metaplastic Changes: Case Comparison and Review of the Literature

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Leydig cell tumours are a type of sex chord stromal tumour found in the testis; however they can also occur in the ovary. Leydig cell tumours represent 1–3% of all testicular tumours but comprise less than 0.1% of ovarian tumours. In men, they arise at a broad age range between 20–70 years with some tumours occurring in prepubertal children. In contrast, Leydig cell tumours of the ovary occur mostly in post-menopausal women with an average age of 60. In both sexes these tumours commonly present as an incidental finding, although they can produce endocrine symptoms such as feminisation or virilisation in males and virilisation in females. Macroscopically they tend to be small and unilateral but 10% of testicular Leydig cell tumours can be bilateral. Importantly 10% of Leydig cell tumours of the testis are malignant compared to their ovarian counterparts which are almost entirely benign. Microscopically they are typically uniform and composed of sheets of polygonal lipid laden cells but unusual architectural features and metaplastic differentiation such as ossification and adipose metaplasia can be seen. These metaplastic changes are rare and have been previously described in a small case series of testicular Leydig cell tumours but not in Leydig cell tumours of the ovary. We present two cases of a Leydig cell tumour, one arising in the testis of a 50 year old man and the other found incidentally in the ovary of a 70 year old woman. Both cases show a benign Leydig cell tumour with areas of ossification and adipose differentiation. We compare and discuss the histological appearances and diagnostic challenges associated with these unusual features. Critically we describe these changes for the first time in an ovarian Leydig cell tumour and explore the possible pathogenesis and diagnostic implications.

P57

Rare Primary Leiomyosarcoma of the Uterine Cervix: 4 Cases and Review of the Literature

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Case report: A 65 year old woman presented with a 7.2 cm cervical mass. At macroscopic examination the mass was based in the cervix with no involvement of the uterine corpus. Microscopically, the lesion consisted of atypical spindled cells with necrosis and a high mitotic index (40/10HPF). Immunohistochemically the tumour showed expression for smooth muscle markers, confirming the diagnosis of a cervical leiomyosarcoma. Retrospectively, we only found 4 cases (including the current case) of primary cervical leiomyosarcoma over the last 10 years in our institute. The ages at diagnosis were between 23 and 65 years and one patient was pregnant. The tumour diameters were between 5 and 8 cm. The oestrogen receptor was negative in 3 cases while one case showed weak staining. The progesterone receptor was negative in 2 cases and strongly positive in 2 cases. We have follow up data for 3 patients all of which were treated with radical hysterectomy without lymph node dissection. One patient had local recurrence after 2 years and died 3 years after diagnosis. Two patients are disease free with follow up of 8 years and 2 months respectively.
Discussion: With these 4 cases we would like to draw attention to this rare entity. Primary cervical sarcomas account for less than 1 % of all tumours of the cervix. Cervical Leiomyosarcoma represents 0.21% among all invasive tumours of the uterine cervix. A leiomyosarcoma of the uterine corpus invading in the cervix must be excluded, since this is much more frequent. Rare carcinosarcoma of the cervix should also be excluded by thorough sampling. Leiomyosarcomas of the cervix have a worse prognosis than carcinomas and are therefore important to recognise. Most patients are in their 4th to 6th decade. Due to the rarity of the disease, therapeutic options are extrapolated from uterine leiomyosarcomas but are not well investigated. In view of the rare instances of the involvement of lymph nodes, routine pelvic lymphadenectomy is not recommended.

P58

The Association of Human Papilloma Virus Subtypes with Lymph Node Metastases and Recurrence of Vulval Squamous Cell Carcinoma

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The incidence of global vulval squamous cell carcinoma (VSCC) is 2.5 in 100,000 women per year and it represents between 2–5% of all gynaecological cancers. The incidence of VSCC is rising, specifically in younger women and this has been attributed to rising Human Papilloma Virus (HPV) infection rates. The disease is associated with high morbidity including lymphoedema, incontinence and psychosocial disability. Once the disease has metastasised to the lymph nodes, the prognosis is dismal. In this study we will identify the subtypes of HPV that drive VSCC and correlate them with clinical data including disease recurrence and lymph node metastases. We identified 29 cases of VSCC from our Co-path database at UCLH from 2015 to 2016. We collated the clinical data and diagnostic histopathology reports. All H&E slides were reviewed and an appropriate FFPE block of tumour per case was selected. These were sent to our diagnostic laboratory for p16 immunohistochemistry and for HPV genotyping by PCR array (Zytovision visionarray chip). There were 40 HPV genotypes assessed (12 high risk, 11 probable high risk and 17 low risk subtypes). 31% of these patients are now deceased (9/29) and 89% of the deceased patients had pelvic lymph node metastases (8/9). 7/29 patients experienced one or more disease recurrences post-surgery. In total, 58% of patients had lymph node metastases (17/29), 28% had negative lymph nodes (8/29) and 14% of patients did not have lymph nodes sampled (4/29). The results of the HPV genotyping of each tumour will provide us with important information about HPV driven VSCC and the subtypes that may confer a worse prognosis. A significant number of patients had lymph node metastases. Most patients without lymph node metastases are still alive after 3 to 4 years. This study will improve our understanding of the pathogenesis of HPV-driven VSCC by identifying HPV subtypes associated with metastasis and progression.

P59

Primary Malignant Melanoma of the Female Genital Tract: An Audit of Histopathology Reporting

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Aims/objectives: Primary malignant melanoma of the female genital tract (FGT; vulva, vagina or cervix) is rare, and carries a poor prognosis compared to cutaneous melanoma. Histological prognostic factors are not defined and validated for these tumours. In the absence of published reporting guidelines for FGT melanoma, we assumed that the Royal College of Pathologists' cutaneous melanoma dataset would be used to report them. We performed this audit to investigate whether core dataset items were being reported at our institution.
Methods: All cases of malignant melanoma occurring in the FGT since 2007 at our tertiary referral hospital were identified via a text search of the pathology reporting database (CoPath). Primary FGT origin was confirmed via electronic clinical notes. Excision/resection reports were reviewed, and core dataset items were recorded as present/absent from the report.
Results: 31 FGT melanomas were identified: 20 vulval, and 11 vaginal or cervical. Mean age at diagnosis was 64. 45% of patients were Caucasian. 25% had a history of other previous malignancy, including one BRCA2 mutation carrier. Excision or resection specimen reports were available for 25 cases. Dataset parameters reported for these cases included: tumour subtype (56%), Breslow thickness (91%), ulceration (68%), mitotic index (23%), lymphovascular invasion (58%), regression (23%), stage (32%), completeness of excision (92%).
Conclusions: Key melanoma dataset items are often absent in reports in primary FGT melanomas. In view of the rarity of these tumours and the paucity of known prognostic indicators, we recommend routine use of the RCPath dataset for cutaneous melanoma to increase the knowlege base in this area. Reporting could be improved by the use of proformas and specialist dermatopathology input.

P60

Distribution of Leiomyosarcomas in the Female Genital Tract: A 10 year Retrospective Study

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Introduction: Of all sarcomas in the female genital tract, leiomyosarcoma of the uterine corpus has the highest incidence (54% of all sarcomas). The myometrium is the commonest location because of the abundance of smooth muscle. Leiomyosarcomas at other locations in the female genital tract are much less frequent and are thought to arise from vascular smooth muscle bundles. With this study we aim to investigate the distribution of leiomyosarcomas in the female genital tract.
Methods: We performed a retrospective search over the last 10 years in the archive of our tertiary hospital. All patients with a biopsy or resection from a primary tumour were recorded and when there was a biopsy or resection of a metastasis, the location of the primary tumour was noted.
Results: In total 76 patients were diagnosed with a leiomyosarcoma in the female genital tract over the last 10 years. Of these, 64 were primary uterine leiomyosarcoma. The other locations of primary leiomyosarcoma were vagina (5), cervix (4), vulva (2) and fallopian tube (1).
Discussion: Although leiomyosarcoma are most frequent in the corpus uteri, more than 15% of our cases had another primary origin in the female genital tract. These included all other organs, except the ovaries. Ovarian leiomyosarcomas are described in the literature but are very rare. This distribution appears to have remained stable over time as our data is concordant with older literature that showed that 86% of the gynaecologic leiomyosarcomas are uterine. With this report we want to emphasise that leiomyosarcomas have a more heterogeneous site distribution than mostly thought and that a smooth muscle neoplasm must be considered in every spindle cell lesion of the female genital tract.

P61

A Rare Diagnosis after Investigation for Infertility

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We present this case of a 38 year old female patient who underwent an exploratory laparoscopy for investigation of infertility and abdominal pain. At the time of surgery multiple solid white nodules were noted and biopsies were obtained from the gynaecological tract, bladder and peritoneum. Histology showed that all the biopsies comprised a papillary tumour with fibrovascular cores lined by pleomorphic epitheloid tumour cells. Immunohistochemistry was performed and the tumour cells were positive for WT1, Calretinin, podoplanin and CK5/6, confirming mesothelial origin. The tumour cells were negative for Pax-8, p53, p16, ER and PR. The clinical, histological and immunohistochemical features were consistent with a diagnosis of primary peritoneal mesothelioma. Primary peritoneal mesothelioma is a rare but aggressive malignancy, which comprises only a small proportion of all mesotheliomas diagnosed. It is poorly described and the knowledge of its natural history is very limited. Only 20% to 33% of all mesotheliomas arise from the peritoneum itself; the pleura is the most common site of origin. Other locations include tunica vaginalis testis, and pericardium. This tumour is most frequently seen in middle aged adults but can present at any age and usually has a male preponderance. Primary peritoneal mesothelioma is associated with asbestos exposure in approximately half of cases. A reported association with simian virus (SV) 40 remains controversial. This tumour tends to spread throughout the peritoneal cavity, producing a diffuse form of the disease. Treatment options include debulking surgery and chemoradiotherapy. The main histopathological differential diagnosis lies between metastatic carcinoma and reactive mesothelial hyperplasia.

P62

Does Epithelial Mesenchymal Transition Play a Role in Progression of Surface Epithelial Ovarian Carcinoma? Evidence from an Immunohistochemical Study

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Ovarian carcinoma (OC) is the 10th most common cancer and the fifth leading cause of cancer related death among women worldwide and in Egypt.
Purpose of the study: In order to evaluate epithelial mesenchymal transition (EMT) that may occur in OC and affect prognosis, this study used expression of SIX1, EYA2 and E-cadherin.
Methods: Formalin-fixed, paraffin-embedded of 65 surface epithelial ovarian carcinoma cases and 20 atypical proliferative (borderline) tumour were stained immunohistochemically. Scoring was done using H-score method and results of scoring were statistically analysed and correlated with clinicopathologic parameters.
Results: Results revealed SIX-1 over-expression in studied malignant cases which was significantly associated with high mitotic count (P=0.017) and large tumour size (P=0.045). Furthermore, the malignant invasive group showed significantly higher intratumoural stromal SIX-1 expression (p= 0.0001) and lower H-score of E-cadherin (P=0.020) when compared with borderline group. In contrast, expression of EYA-2 did not differ between malignant and borderline tumours (P = 0.215). In terms of therapy, there was a significant association between platinum based chemotherapy sensitivity, solid cut section (P=0.044), early stage (I, II) (P=0.05), long overall survival (p=0.003) and progression free survival (P=0.0001).
Conclusions: This study suggests that SIX-1 may contribute to ovarian epithelial carcinogenesis by increasing proliferation and associated with poor prognosis. SIX-1 over-expression may be helpful in predicting stromal invasion in borderline tumours. The most independent prognostic factor in surface epithelial ovarian carcinoma is response to platinum based therapy.

P63

Phyllodes Tumour of the Vulva: A Rare Case Report

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Introduction: Phyllodes tumours of the vulva are uncommon neoplasms with a leaf like architecture and biphasic component histologically. This tumour shares morphological similarities with phyllodes tumour occuring in the breast supported by the immunohistochemical expression of breast markers, oestrogen and progesterone receptors.
Case Report: A 64 year old female presented with lump on her right vulva for a duration of 3–4 months. On examination a 5cm exophytic lump was seen but no lymph nodes were palpable bilaterally. The findings were concerning and a fungating vulval lesion was in the differential diagnosis. A biopsy showed pseudopolypoid fibroconnective tissue stroma covered by epithelium displaying variable appearance ranging from a double epithelial layer to areas with more complex micropapillary tufting with cross-linking, and squamous epithelium. The lesion was subsequently excised and histology showed a biphasic tumour with an epithelial and stromal component. The epithelial component comprised columnar cells with areas of papillary tufting. The stromal component was collagenous and hypocellular with admixed spindle stromal cells lacking cytological atypia. Mitotic figures were scarce and the lesion appeared excised. Immunohistochemistry on both specimens showed positive staining with GATA-3, ER, PR and Vimentin in the epithelial component. SMA, CK5/6 and S100 stained the myoepithelial cells but showed negative staining in the stromal component. The diagnosis of a benign phyllodes tumour was made.
Discussion: Clinically, these tumours are unilateral, solitary, pain less and located in the labia majora. The management is surgical excision with clear margins. Recurrence is rare and no record of metastasis has been reported. In conclusion this rare tumour should be considered in the differential diagnosis for women presenting with a slow growing vulval mass.

P64

Cystic Endosalpingiosis-Associated Clear Cell Carcinoma Arising in a Caesarian Section Scar

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A 48 year old woman was admitted for investigation of a longstanding 3.5cm fungating abdominal skin lesion, which was unresponsive to antibiotics. Past medical history includes laparoscopic investigation for endometriosis and Caesarean section. Ultrasound imaging showed a lobulated soft tissue mass in the right iliac fossa region, extending from ulcerated skin to deep subcutaneous tissue. Skin punch biopsy demonstrated multiple cysts lined by simple columnar to cuboidal epithelium. These cells were positive for CK7, ER, CA-125 and BerEP4, with negative staining including calretinin, WT1, CK20, CK5, p63, TTF1 and CD10 (with patchy stromal staining). There was no atypia, necrosis or mitoses. Consensus diagnosis was of a 'mullerianosis', most in keeping with cystic endosalpingiosis. Abdominal CT imaging identified bilateral inguinal lymphadenopathy. Pelvic organs were normal. Due to the lesion size and associated lymphadenopathy, she underwent excision of the lesion with lymphadenectomy. Macroscopically, there was residual Caesarean section scar abutting the 16 x 12 x 11cm lesion, which had cystic, solid and papillary components. Histology showed clear cell carcinoma, confirmed by positive staining for CK7, CA125 and napsin A, with negative staining for ER, WT1, p16 and p53 (wild type). Staining was repeated on areas histologically similar to the biopsy to confirm the cystic endosalpingiosis component. Clear cell metastases were present in multiple local lymph nodes. Ongoing active treatment options are under consideration.

P65

Total Processing of the Omentum to Identify Microscopic Implants in Patients with Serous Borderline Ovarian Tumours

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Aims: Serous Borderline Ovarian Tumours (SBOTs) may form extra-ovarian deposits known as implants, commonly in the omentum. Thorough omental sampling to detect microscopic implants is crucial for staging SBOTs. However, there is little information for a specific sampling threshold that must be reached to detect implants. We aimed to determine if total omental sampling is necessary to detect microscopic SBOT implants.
Methods: The trust histology database was searched for pathology reports of patients with SBOTs between 2008 and 2019. Completeness of omental sampling, the number of omentum blocks processed, and the detection of implants was noted.
Results: Of 69 cases with total omental sampling, 19 had microscopic implants, compared to 17 in 30 cases that did not undergo total omental sampling. In 23 cases where the omentum was incompletely sampled when initially received, no implants were found and the rest of the omentum was processed. In 3 such cases, implants were found in the extra blocks. On average, 1 further block was taken in cases that contained an implant compared with those that did not contain implants. Results were compared to the recommendation of 10 blocks for a grossly normal omentum. Of 6 cases in which exactly 10 blocks were initially taken with tissue remaining, 2 cases contained no implants and in one of these cases implants were found in further blocks.
Conclusions: Sampling 10 blocks of grossly normal omentum is insufficient to avoid missing implants. Complete sampling of the omentum is recommended to accurately stage SBOTs.

P66

An Unusual Complication of Tension Free Trans Vaginal Tape

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Objectives: We report an unusual complication of Tension-Free Vaginal Tape (TVT) use for posterior vaginal wall prolapse. Histopathologists are not used to seeing complications of the use of TVT in specimens sent to them as these specimens are usually received at certain centres in the UK. We feel that an awareness of the rather serious possible complications that may ensue following the use of TVTs should be known by histopathologists so that there is no confusion with other primary gastro-intestinal pathology. Thorough examination and documentation of these specimens with clinico-pathological correlation is required.
Method: 64 year old female, underwent posterior vaginal wall prolapse repair with mesh and sacrospinious fixation in 2009. Following which, the patient became clinically symptomatic with vaginal and anal pain and dyspareunia. The Magnetic Resonance Imaging revealed migration of the mesh which had launched in to the posterior vaginal wall close to rectum. This was partially removed under emergency colorectal operation in 2015. A year later, she suffered intermittent episodes of systemic sepsis. The 3D ultrasound scan located residual mesh between the lower posterior vaginal wall and the rectum. This resulted in a rectovaginal fistula. She required an ultra-low anterior resection to excise the mesh and fistula. Gross examination of the bowel revealed the mesh in situ within the bowel wall and histological confirmation of microscopic erosion of the mesh into the intestinal mucosa was confirmed. Recto-vaginal fistula was not obvious macroscopically.
Conclusion: Complications of Tension-Free Vaginal Mesh have been associated with medicolegal issues and there are national enquiries about safety of this procedure. We have described here a rare but serious complication of bowel injury following the use of TVT, and feel it is important for the pathologists to be aware of its complication so that appropriate examination and documentation can occur.

P67

A Comparison of Pre- and Post-Operative Histology Findings in Patients with Atypical Endometrial Hyperplasia Diagnosed by Endometrial Biopsy

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Purpose of the the study: To assess the concordance between pre and postoperative histological diagnosis in patients surgically treated for atypical endometrial hyperplasia.
Methods: A search and analysis was performed using the service's electronic reporting database over a two year period. Cases of atypical hyperplasia diagnosed on endometrial biopsies were identified, correlated and compared to subsequent hysterectomy specimen reports.
Summary of results: 48% of the cases diagnosed preoperatively as atypical hyperplasia were found to have endometrioid adenocarcinoma on subsequent hysterectomy. When separated into biopsies reported as "atypical hyperplasia suspicious but not diagnostic of grade 1 endometrioid adenocarcinoma" 73% were diagnosed as endometrioid adenocarcinoma on hysterectomy while only 14% of the cases reported only as "atypical hyperplasia" were found to have concurrent endometrioid adenocarcinoma.
Conclusions: Sometimes the features in a biopsy may be at the cusp between atypical hyperplasia and grade 1 endometrioid adenocarcinoma; these patients have a higher incidence of cancer in the subsequent hysterectomy. Such cases should not be reported merely as a conventional atypical hyperplasia but the strong suspicion for cancer should be mentioned in the pathology report. All suspicious cases should be discussed at Multidisciplinary team meetings and receive special attention compared to those reported only as atypical hyperplasia. These cases should be offered imaging in a similar manner to biopsy proven cancer with abdomino-pelvic MRI, chest X-ray and according to the 62 day target cancer pathway.

P68

An Audit of Atypical Complex Hyperplasia Reporting and Management

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Background: This audit was carried out to determine the proportion of women diagnosed with atypical complex hyperplasia (ACH) who undergo hysterectomy as recommended by the RCOG/BSGE Joint Guideline "Management of Endometrial Hyperplasia". The degree of histological agreement between the initial biopsy and hysterectomy specimen was also investigated.
Method: This was a retrospective selection of all cases for a 14 month period between 01/01/17 and 01/03/18. We reviewed histology reports on the laboratory information system and recorded the biopsy and hysterectomy specimen diagnoses.
Results: 39 cases of ACH were identified during the search period. Of these, 34 patients underwent hysterectomies as per the guidelines. The 5 patients who did not undergo hysterectomies had medical reasons. 16 patients were found to have endometrial cancer after undergoing a hysterectomy. The compliance between the biopsy and hysterectomy diagnosis was 82% (28/39). This figure includes cases whereby the initial biopsy diagnosis was ACH, and in the hysterectomy specimen a diagnosis of at least ACH was present. 15% (6/39) of hysterectomies did not show features of ACH.
Conclusion: The results of this audit have demonstrated that we are meeting the recommended targets outlined by the RCOG/BSGE with the exception of patients who are not medically fit for surgery. The cases showing a lack of correlation between the biopsy and resection specimen were re-reviewed. All six cases had thorough sampling of the endometrium. Reasons for non-compliance included poorly preserved endometrium, progesterone effect in patients started on progesterone therapy, and cases where ACH was identified within a polyp. This audit has highlighted the importance of adequate tissue fixation to enable accurate histological assessment. In addition the use of progesterone therapies appears to be having a therapeutic effect on cases with ACH as demonstrated in 3 cases from our cohort where no residual ACH was identified.

P69

Oligometastasis of Lobular Breast Carcinoma to the Uterine Cervix: A Case Report

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A case report of lobular carcinoma of the breast, metastatic to the uterine cervix, and detected in a routine cervical smear test. A 56 year old female underwent a routine cervical smear. The smear contained groups and sheets of dyscohesive, round to oval cells with pleomorphic, eccentric nuclei and granular cytoplasm, some with targetoid intracytoplasmic inclusions. Abnormal mitoses were noted and tumour diathesis was present in the background. A review of the medical records revealed a history of bilateral breast carcinoma, including a grade 2 lobular carcinoma of the right breast. Histology from the left breast tumour was not available for review. A diagnosis of non-cervical adenocarcinoma was made on the smear with a comment suggesting lobular carcinoma. A tissue biopsy from the cervix was obtained, and a diagnosis of metastatic lobular breast carcinoma was confirmed morphologically and immunohistochemically. Imaging of the chest, abdomen and pelvis revealed no evidence of further metastases. Metastases to the female genital tract from extragenital primary tumours are uncommon. The most common site for genital tract metastases is the ovary. The uterus, especially the uterine cervix, is rarely involved by metastatic tumours. In extragenital tumours metastatic to the female genital tract, the breast is the second most common primary site, behind the gastrointestinal tract. Invasive lobular carcinoma of the breast is renowned for exhibiting unusual metastatic patterns. Invasive lobular carcinoma spreads more frequently to the genital tract than invasive ductal carcinoma. Correct diagnosis is important as management of primary genital tract malignancy and metastatic breast carcinoma differ dramatically. It is therefore important for the pathologist to consider metastatic carcinoma in the differential diagnosis of a cervical mass or abnormal cervical smear.

P70

A Retrospective Analysis of Negative Large Loop Excisions of the Transformation Zone (LLETZs)

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Purpose of the study: The increased risk of preterm birth, mid-trimester miscarriage and ectopic pregnancy following large loop excision of the transformation zone (LLETZ) makes minimising unnecessary procedures critical. This audit quantified and confirmed the histological status of negative LLETZs and determined the indication for, and management pathway to, excision.
Methods: Retrospective analysis of LLETZ specimens (n=1146) from 2016 across two Yorkshire centres identified 81 negative cases which were compared with a randomly selected, size-matched positive comparator group. The indication, punch biopsy dimensions, biopsy-to-LLETZ time interval and the number of levels examined were analysed.
Summary of results: The negative LLETZ rate was 7%. Indications for LLETZ fitted into three categories: see-and-treat at colposcopy following screening (30 vs. 63% in the negative and positive LLETZ groups, respectively), treat despite negative biopsy (15 vs. 1%) and treat following positive biopsy (55 vs. 36%). Mean pre-LLETZ punch biopsy volume was smaller in the positive LLETZ cohort (64.3 vs. 108.1mm3). More negative LLETZs (79%) had multiple levels cut than their positive counterparts (62%). There was no significant difference in biopsy-to-LLETZ time interval across positive and negative LLETZ groups.
Conclusions: These findings suggest that the larger biopsies taken in the negative group have excised the lesions, in essence a therapeutic biopsy. The finding of a negative LLETZ following a positive biopsy adds no clinical value, as the management will be test of cure regardless. This raises the question of whether further levels should be routinely cut and/or tissue reorientated on every block for negative LLETZs, or whether they need multidisciplinary team discussion, as this will not change management. The results also suggest that colposcopists may be more confident in their clinical impression in the positive LLETZ group, with fewer/smaller biopsies being taken.

P71

Risk of Vulval Squamous Cell Carcinoma Amongst Patients with Lichen Sclerosus

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Purpose of study: Vulval Lichen Sclerosus (VLS) is an inflammatory condition of unclear aetiology charaterised by the development of itchy plaques, scarring and local histoarchitectural destruction. Of particular concern is the allied risk of vulval squamous cell carcinoma (VSCC) arising from VLS. Although this risk has been approximated to circa 5%, there is a paucity of large scale studies with appropriate follow-up to corroborate this figure. The aim of this study was therefore to determine the risk of malignant transformation in a large retrospective cohort of patient with VLS.
Method: This study examined the clinical records of 762 women suffering from VLS across St James's University Hospital, Leeds, and Bradford Royal Infirmary across the period spanning 1993 to the present day. Data collected included incidence of VSCC and recurrence rate.
Summary of results: Eighty-eight (11.5%) women with VLS developed VSCC. Of these, 48 (55%) developed a recurrence post intended curative treatment.
Conclusions: These figures markedly exceed current published data relating to malignant transformation rates of women with VLS; these range from <1-5%. Recurrence rates were also higher in our study compared to published data (12–50%). These findings highlight that current figures relating to the malignant transformation of VLS are likely to significantly underestimate true risk. It also underscores for clinicians to manage VLS aggressively in order to prevent the development of VSCC. Further follow-on work will assess the impact of stage, grade and age at diagnosis as well as human papillomavirus status in our current population in order to determine their impact on VLS transformation to VSCC.

P72

Adenocarcinomatous Transformation of a Retroperitoneal Teratoma Mimicking an Adrenal Incidentaloma in an Adult Female: A Case Report and Literature Review

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Retroperitoneal teratomas are extremely rare in adults and are typically benign tumours. We describe a case of a 48-year-old lady who initially presented with abdominal discomfort 18 years ago and was found to have a large right-sided supra-renal mass suspected to be an adrenal lesion. Following initial biochemical and imaging studies a diagnosis of non-functioning adrenal adenoma was made and she was managed conservatively. After being lost to follow up and remaining clinically well for many years, she once again presented with worsening abdominal pain. A right adrenalectomy was performed and the histology surprisingly confirmed a mature cystic teratoma with malignant transformation into a moderately-differentiated intestinal-type adenocarcinoma as well as having a focus of carcinoid tumour. She was admitted shortly afterwards with widespread metastatic disease and sadly passed away before any treatment could be instituted. Only a handful of cases of primary retroperitoneal teratomas have described adenocarcinomatous transformation. A carcinoid tumour within a teratoma is even less common and typically does not manifest systemic features. This case underlines the broad differential diagnosis of an adrenal mass and highlights several unusual aspects of a teratoma. We present the current literature and discuss the appropriate investigations, management and challenges involved in such rare cases.

P73

Diagnostic Value of TROP-2 and Ck19 Expression in Papillary Thyroid Carcinoma in Both Surgical and Cytological Specimens

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Background: Several immunohistochemical markers are used individually or in combination to help in differentiation of papillary thyroid carcinoma (PTC) from mimickers. Purpose of the study: The current study aims at assessment of the diagnostic value of TROP-2 and CK19 in differentiating PTC from mimickers both singly and combined.
Methods: The current study was carried out on 77 surgical specimens (56 PTC and 21 non neoplastic cases) and 12 cytological specimens (4 THY2, 6 THY4 and 2 THY5).
Summary of results: TROP-2 was negative in 81% of non-neoplastic surgical specimens and in 100% of THY2 cytological specimens while it was positive in 71.4% of PTC surgical specimens and 100% of TY4/THY5 cytological specimens. Sensitivity and specificity of TROP-2 positive expression for diagnosis of PTC in surgical specimens reached 71% and 81%, respectively while it reached 100% for both in cytological specimens. CK19 was expressed in 85.7% of non-neoplastic surgical specimens and in 92.9% of PTC surgical specimens. CK19 showed negative expression in 75% Thy2 while it was positive in all Thy4 and Thy5. Sensitivity and specificity of CK19 total estimated score for diagnosis of PTC in surgical specimen were 78.6 % and 66.7%, respectively while it reached 100% and 75% in cytological specimens. Positive TROP-2 and CK19 in PTC were associated with lymph node metastasis.
Conclusions: TROP-2 is a specific rather than sensitive marker while CK19 is a sensitive than specific marker in differentiating PTC from mimickers in surgical specimens. The diagnostic validity of both markers was superior in diagnosis of classic PTC compared to follicular variant PTC. TROP-2 is superior to CK19 in diagnosis of PTC in cytological specimens. Both TROP-2 and CK19 could be used preoperatively in adjunct to hematoxylin and eosin for more confident diagnosis of thyroid cytology and beside radiology as predictors of lymph node metastasis.

P74

Platform-Independent Prediction of Malignant Transformation Within Ten Years of Confirmed Oral Epithelial Dysplasia Using Image Analysis

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Objectives: Oral Squamous Cell Carcinoma (OSCC) is one of the more frequently diagnosed cancers worldwide. Some cases of OSCC are preceded by changes in the oral epithelium known as dysplasia. Reported transformation from oral epithelial dysplasia (OED) into malignancy ranges from 3.2-50.0%. Currently there is no reliable method of predicting which lesions progress into malignancy and which don't. The overall aim was to test CD3 and CD8 as potential biomarkers for predicting malignant transformation which was assessed by correlation of CD3 and CD8 expression with progression of OED into OSCC. An additional objective of this study was to compare four image analysis approaches (Qupath, Halo, Visiopharm and Definiens) for quantifying levels of CD3 and CD8 in OED in order to determine inter-platform reproducibility.
Methods: 49 cases of OED with at least two biopsies from the same oral site and with ten years follow up were subcategorised into two groups; (a) patients whose dysplasia progressed into malignancy within ten years and (b) patients whose dysplasia remained as dysplasia within ten years. The levels of CD3 and CD8 were quantified in the two groups of patients using four different image analysis platforms: Halo, Qupath, Visiopharm and Definiens. The inter-platform reproducibility of the test was also observed to compare the four image analysis software's utilised in this study.
Results: Levels of CD3 in the two subgroups of patients demonstrated no significant differences between the two patient groups ($p>0.05$). There was a significant difference noted in the level of CD8 between the two subgroups of patients ($p<0.05$). There was strong correlation of the results produced by the four software ($R>0.7$).
Conclusions: CD8 was confirmed as a potential biomarker for predicting the malignant transformation of OED. The inter-platform reproducibility suggests that the analysis of the biomarkers tested in this study should be similar when performed by any platform.

P75

Identification of Novel Copy Number Alterations in Ameloblastoma and Ameloblastic Carcinoma from Nigeria

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Purpose of study: Ameloblastoma is a benign odontogenic neoplasm, characterized by local invasiveness, facial deformity, tooth displacement, a high rate of recurrence, and malignant transformation. It accounts for 63% of odontogenic tumours in Nigeria. Recently, studies in the genomic landscape of ameloblastoma have identified a number of consistent alterations that may be useful for therapeutic intervention. To date, no whole genome survey of ameloblastoma and ameloblastic carcinoma has been published.
Methods: DNA was extracted from RNALater stored tissue using the DNeasy Tissue Kit (QIAGEN), from a cohort of ten ameloblastoma and three ameloblastic carcinoma from UCH, Ibadan, Nigeria. Whole genome analysis was performed using the Oncoscan FFPE Assay Kit (Affymetrix). Data was analysed using Nexus Express for Oncoscan 17.0 and Somatic Mutation Viewer 1.0.1.
Results: Ameloblastoma (n=10) showed a mean genome change of 9.7%, with a mean of 88.7 copy number (CN) aberrations and 7.5% of loss of heterozygosity (LOH), whereas the ameloblastic carcinomas (n=3) had a mean genome change of 6.8% with a mean of 87.3 copy number (CN) aberrations and 3.6% of loss of heterozygosity (LOH). All tumours (benign and malignant) showed CN gain at 8q23.3, affecting the CSMD3 gene. Other commonly affected regions included LOH at 1p34.2-p34.1 and 2q11.2, among others. Ameloblastoma and ameloblastic carcinomas shared somatic mutations in BRAFV600E, EGFR, KRAS and PTEN genes. One ameloblastoma showed a mutation in TP53 and two (66.7%) ameloblastic carcinomas showed a mutation in the PIK3CA gene, which was not observed in the ameloblastoma cohort.
Conclusions: Ameloblastoma and ameloblastic carcinoma do not show extensive genome changes indicative of genomic instability. We have identified novel areas of CN gain and LOH that require further investigation. The mutational profile of these lesions is similar to that reported in the literature.
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P76

Hypoxia and HPV Status in Oropharyngeal Squamous Cell Carcinoma

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Hypoxia attributable to disorganised vascularisation variously features in the microenvironment of solid tumours and adversely affects response to treatment and prognosis. The present investigation aims at exploring whether tumour-cell phenotypes related to micro-environmental oxygen levels are influenced by the integrated human papilloma virus (HPV) infection in oropharyngeal squamous cell carcinoma (OPSCC). Microarrays or full sections from formalin-fixed, paraffin-embedded tissues of 41 HPV(+) and 34 HPV(-) OPSCCs were examined by means of immunohistochemistry for 'anaerobic' phenotypes (hypoxia inducible factor 1 alpha, HIF1α), glucose transporter 1 (GLUT1) and (on a smaller cohort) monocarboxylate transporter 4 (MCT4); mitochondria (TOMM20); and 'angiogenic' phenotypes (vascular endothelial growth factor, VEGF). Membranous expression of MCT4 was seen in the front of 40/41 and 26/34 HPV(+) and HPV(-) tumours, respectively ($p = 0.023$). Nuclear expression of HIF1α was seen in the centre of tumour cell aggregates in both front and core of 4/10 HPV(+) and 8/10 HPV(-) tumours ($p = 0.08$). Membranous GLUT1, cytoplasmic granular TOMM20 and cytoplasmic diffuse VEGF expression were seen throughout the tumour parenchyma, independently of the HPV status (10/10 and 10/10). The results suggest that while the tumour cells in HPV(+) and HPV(-) OPSCC show similar mitochondrial load and transport glucose, their 'anaerobic' profile differs. Tumour cells in HPV(+) tumours more frequently release lactate via expression of MCT4, whereas hypoxia inducible transcription factors are more commonly synthesised by HPV(-) tumour cells. 'Angiogenic' phenotypes appear similar in both groups of tumours and not directly related to those differences.

P77

Is Human Papillomavirus-Related Carcinoma with Adenoid Cystic-like Features Limited to the Sinonasal Tract? A Review of Two Cases from the Palate

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Introduction: Human Papillomavirus (HPV)- related carcinoma with adenoid cystic like features is a recently described entity limited to the sinonasal tract. It is associated with high risk HPV infection namely HPV type 33, 35 and 16. Histology usually show basaloid myoepithelial cells, ductal cells, solid, cribriform and tubular growth patterns may be seen. Abrupt keratinisation within tumour nest and dyspasia of surface epithelium can also be seen in this entity. We review two cases of adenoid cystic carcinoma (ACC) of the palate in two female patients presenting with palatal swelling.
Methods: Two middle aged females 34 and 42 years respectively had excision biopsy of a soft tissue palatal swelling. Specimen was fixed in 10% buffered formalin. Slides were cut and stained with H&E. Immunohistochemistry was performed using DB Biotech (Slovakia). Two antibodies were used p16 and AE1/AE3.
Results: Morphology of the tumour in the younger patient (34 years) showed features of HPV-related carcinoma with adenoid cystic-like features. A predominant basaloid myoepithelial-type cells with ductal cells arranged in solid and cribriform pattern. Microcystic areas containing basophilic material and surface epithelial dysplasia was seen. Diffuse nuclear and cytoplasmic staining with p16 was seen in >90% of tumour cells. The second patients tumour morphology was that of adenoid cystic carcinoma of the salivary gland with p16 staining been negative. Both tumours were positive for AE1/AE3.
Conclusion: This study suggests that HPV related carcinoma with adenoid cystic features may occur outside the sinonasal tract. It may be very important to do MYB immunostaining and FISH in some cases in other to differenciate it from adenoid cystic carcinoma and other lesions.

P78

Recurrent Pleomorphic Adenoma in the Maxillary Sinus: An Unusual Location

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Introduction: Primary and recurrent Pleomorphic Adenoma is most commonly found the in the parotid gland. Minor salivary glands can harbour PA but recurrences involving the maxillary sinus are very rare. Here we report a 33 year old male who presented with a mass in the right maxillary sinus.
Case History: A 33 year old man presented with a right floor of nose lesion causing obstructive sypmtoms of the maxillary sinus blocking the right maxillary sinus. Computer Tomography revealed a 40mm mass involving the hard palate and maxillary sinus. Following an incisional biopsy diagnosis of pleomorphic adenoma, right maxillectomy and reconstruction with a fibula free flap. Bone allograft was performed.
Pathology: The incisional biopsy showed a cellular tumour with focal ductal differentiation and large sheets of myoepithelial cells. The features were consistent with pleomorphic adenoma without signs of malignant transformation. The right maxillectomy specimen was mostly replaced by pleomorphic adenoma where tumour islands were permeating bone tissue. Squamous metaplasia was noted but there was no evidence of carcinoma ex pleomorphic adenoma. A histologic diagnosis of recurrent PA was rendered.
Discussion: Recurrent PA is usually seen in the parotid gland mostly in women younger than 40 yrs. Our patient developed a PA of minor salivary glands of the palate at the age of 24 yrs treated elsewhere. At 33 yrs a massive recurrence was detected in the right maxillary sinus but the radiologic findings and clinical symptoms were not specific. Definitive histological was made on open biopsy and confirmed on subsequent surgical specimen.
Conclusion: Maxillary sinus recurrence of PA of palate is rare and it should be considered in the differential diagnosis of benign and malignant expansile masses of the Sinonasal Tract.

P79

Spatial Relationships Between Immune Infiltrate and Tumour Buds Improves Prognostic Accuracy in Stage II Colorectal Cancer

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Purpose of the study: Within the tumour microenvironment, cancer cells coexist and interact with the heterotypic immune system. Tumour budding and the immune infiltrate are established prognostic factors in stage II colorectal cancer (CRC), though the importance of their spatial interaction is less studied. The objectives of this research were to determine the prognostic value of the dynamic interplay between tumour buds (TBs) and infiltrating immune cells.
Methods: Multiplexed immunofluorescence for TBs, CD3+, CD8+ lymphocytes and CD68+, CD163+ macrophages was performed across two sequential whole tissue slides (n=232). Automated image and spatial analysis was applied to quantify the distinct cell populations and their spatial interactions. Machine learning was used for the development of a prognostic risk model.
Summary of results: TB ($p=0.001$) and lymphocytic density ($p<0.001$) were found to be significantly correlated with disease-specific survival. However, a novel prognostic model which also incorporated their spatial relationship was shown to better stratify stage II CRC patients at high risk for disease-specific death ($p<0.001$) compared with the clinical gold standard of pT stage ($p=0.003$). The model was developed using data from 114 patients and validated in two independent cohorts (cohort 1: n=56 and cohort 2: n=62). Furthermore, a low ratio of CD68+/CD163+ cells at the tumour core was associated with better survival ($p<0.001$). A prognostic model integrating the above-mentioned features allowed the identification of patients with low-risk of disease-specific death with 100% sensitivity.
Conclusions: We demonstrate an automated machine learning workflow that captures the cellular interactions present in the tumour microenvironment and which, may lead to improved and personalised clinical decision making.

P80

Intestinal Tumour Modelling and DNA Damage: Investigating the Interaction of Deficient DNA Mismatch Repair and Ethanol in Colorectal Carcinogenesis

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Lynch Syndrome (LS) confers inherited cancer predisposition due to germline mutations in one of the DNA mismatch repair (MMR) genes. MMR is a DNA-damage repair pathway involved in the removal of base mismatches and insertion/deletion loops caused by several endogenous and exogenous factors. Loss of MMR through somatic alteration of the wild-type allele in LS results in defective MMR (dMMR). Ethanol and its metabolite acetaldehyde are classified as group one carcinogens by the IARC. Aldehydes are very reactive molecules that constitute a serious threat to cellular integrity by causing a range of DNA lesions. However, DNA repair pathways responsible for correcting such lesions remains unknown. We hypothesized that MMR is involved in the repair of certain forms of ethanol/acetaldehyde-induced DNA damage. In this study, we aim to determine if there is a gene-environment interaction between dMMR and ethanol/acetaldehyde that accelerates colorectal tumourigenesis. We used a conditional Msh2 knockout mouse model that mimics the LS patients' pattern of MMR gene inactivation. The LS model mice (6-8 weeks of age) were fed either with 20% ethanol in drinking water or normal drinking water. Most of the ethanol-treated mice demonstrated large intestinal hyperproliferation, adenoma formation and, in some cases, invasive adenocarcinoma within 7 months (6/10), compared with one case of intestinal tumour formation after 16.5 months in the water-treated mice (1/10). The quantification of the dMMR crypts in LS mouse colon has shown an increased number of dMMR foci in ethanol-treated mice compared with the control group. Preliminary results indicate that long-term ethanol treatment induced acceleration of dMMR-driven large intestinal tumour formation. Possible mechanisms may include ethanol-induced mucosal crypt epithelial proliferation and ethanol/acetaldehyde mediated DNA damage that would usually be repaired, at least in part, by DNA MMR.
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P81

Differences in Negative Lymph Node Size in Rectal Cancer Resections are Related to Patient's Immune Reaction

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Introduction: The number and size of lymph nodes without metastasis (LNneg) is an independent prognostic factor in rectal cancer (RC) patients. It is unclear what influences LN size in RC patients. To explore whether there is a relationship between LN size and LN architecture in RC patients we focussed our pilot study on LNnegs using haematoxylin/eosin (H&E) stained slides.
Method: 50 RC patients treated between 2012 and 2015 with surgery (n=17) or neoadjuvant treatment (NAT) followed by surgery (n=33) were included. All H&E slides were digitalised and LNnegs were outlined manually using image analysis software to calculate the LN area. In addition, LNs were reviewed under the microscope with a 2.5x objective to establish number of primary and secondary lymphoid follicles and presence of intranodal fat (inF). LNnegs were grouped in five groups based on primary and secondary follicle count. The relationship between LN morphology and LN area was analysed.
Results: 677 LNnegs were analysed. The median size of LNs with NAT is 98.48mm^2 (range 2.74mm^2 – 1107.71mm^2) compared to 119.33mm^2 (range 9.26mm^2 – 1214.89mm^2) in patients without. 150 (22.5%) LNnegs contained inF. Presence of inF was related to smaller LNneg area in patients after NAT (median (range) LNneg area without inF 103.9mm^2 (3.5mm^2 – 1007.7mm^2) versus 82.2mm^2 (2.7mm^2 – 642.9mm^2) with inF) p=0.04. Increasing numbers of lymphoid follicles were related to larger LNneg area in all patients ($p < 0.05$).
Conclusion: This pilot study in LNnegs from RC resections showed that patients with larger LNneg appear to have higher number of lymphoid follicles and interestingly less inF. This study seems to confirm a previously proposed interaction between the immune system and fat tissue. Thus, larger LNs in cancer patients could be related to patient prognosis and treatment response, this need to be investigated in future studies.

P82

The Relationship Between Tumour Immune Profile and Response to FOLFOX-Based Pre-Operative Chemotherapy in the International Phase III FOxTROT Trial

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Recent strong evidence suggests that the immune system influences cancer prognosis in a number of cancers including colorectal cancer. The prognostic importance of tumour infiltrating lymphocytes (TILs) in colon cancer has long been recognised. However, the importance of TILs and other immune cells following preoperative chemotherapy is unknown and a validated methodology for assessment is lacking. Patients were randomly assigned in a 2:1 ratio to pre- and post-operative chemotherapy comprising three 2-week cycles of FOLFOX then surgery followed by nine more cycles, or to post-operative chemotherapy consisting of surgery followed by twelve 2-week cycles. H&E slides were collected for central pathological review in 904 out of 1052 cases (86%). The primary tumour immune profile was assessed for TILs (stromal and intratumoural), neutrophil and eosinophil infiltration at the invasive edge, tertiary lymphoid structures (TLS), abscess formation and necrosis. In addition, we assessed the area of all lymph nodes (LNs) identified and the area of metastatic tumour deposits in LNs. Significantly higher numbers of stromal TILs (14% vs. 9%, p<0.0001) and eosinophils (6% vs. 3%, p<0.0001) were observed in the pre-operative group, however, intratumoural TILs were equivalent (5% vs. 5%, p=0.63). Significantly lower numbers of neutrophils (5% vs. 10%, p<0.0001) and reduced abscess formation (11% vs. 21%, p<0.0001) was seen in the pre-operative group. The average area of all uninvolved LNs was smaller in the pre-operative group as was the average tumour area in metastatic LNs. There were no significant differences in TLS between the two groups. This is the first study of the tumour immune profile in colon cancers treated with pre-operative chemotherapy, and demonstrates fascinating differences in immune cell populations between the chemotherapy and control groups. Ongoing work will assess the importance of tumour immune cell populations in the chemotherapy group according to response.

P83

The Effect of Pre-Operative FOLFOX Chemotherapy in Advanced Colon Cancer on Histopathological Features: Analysis of the International Phase III FOxTROT Trial

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FOxTROT is the first international phase III randomised clinical trial to evaluate the effectiveness of pre-operative chemotherapy in locally advanced colon cancer. Patients were randomly assigned in a 2:1 ratio to pre- and post-operative chemotherapy or to post-operative chemotherapy alone. Central pathological review of the core histopathological features was performed in 904 out of 1052 cases (86%). Additional histopathological features were also assessed during the central review including restaging according to TNM8, extra-nodal spread, tumour budding, poorly differentiated clusters (PDC), and tumour deposits (TDs). All histopathological features generated during the central pathological review were then compared between the pre-operative chemotherapy group and control arm. Pre-operative chemotherapy was associated with a significant reduction in tumour diameter (40mm vs. 51mm, p<0.0001), lower pT stage (pT0-pT2 rate 18% vs 8%, p<0.0001), lower pN stage (pN0 rate 64% vs. 52%, p=0.0002) and greater R0 rate (99% vs. 96%, p=0.02). Apical node metastases were less common in the pre-operative chemotherapy group (3% vs. 8%, p=0.002) as was extracapsular nodal spread (8% vs. 19%, p<0.0001), intramural venous invasion (20% vs. 33%, p<0.0001), extramural venous invasion (35% vs. 44%, p=0.004) and lymphatic invasion (46% vs. 55%, p=0.002). There was no significant difference in perineural invasion (12% vs. 14%, p=0.69). The percentage of the patients with high grade budding was lower in preoperative group (5% vs. 14%, p<0.0001), however, no significant difference was found in the number of PDCs (4.0 vs. 4.7, p=0.08). Pre-operative chemotherapy in locally advanced colon cancer is associated with significant effects on the primary tumour leading to improved complete resection rates. There is also a significant reduction in many high risk features and mechanisms of metastatic spread, all of which are expected to lead to an improvement in patient outcomes.

P84

An Assessment of the Mutation Rate or Normal Colorectal Epithelium in Patients with Cancer Compared to Patients Without

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It is thought that half of somatic mutations present in colorectal cancers have arisen previously in the epithelium. In order to become fixed, these mutations must occur in colonic stem cells which can then replace the crypt. To study the mutation rate we used a neutral clonal marker, MAO-A. It is located on the X chromosome and truncating mutations result in loss of staining of the protein with immunohistochemistry allowing for direct visualisation of fixed mutations. Normal colonic mucosa was examined from cancer patients (cancer-associated normal, CAN) N=9 and patients who had resections for non-neoplastic and non-inflammatory indications (non-neoplastic normal, NNN) N=6. Slides were stained for MAO-A and digitally scanned. The total mucosal area was measured and 300 random points were scored as either epithelium, lamina propria or non-relevant. Next 50 randomly selected crypts per slide were measured to estimate the average crypt size and the total crypt number. For the CAN group the average mutation rate was 1 in 2646 crypts. The 6 samples of NNN had an average mutation rate of 1 in 6737; this meant a 2.6 fold difference for CAN compared to NNN. This difference was significant (p=0.0198). The average age of the patients in the two groups was no significantly different; CAN=72years, NNN=69years, p=0.516. The mutation rate increased with increasing age in both groups; the lowest mutation rate was 1 in 12842 for a patient aged 44 and the highest rate was 1 in 901 crypts for patient aged 88. Mutations accumulate throughout the colorectal epithelium during a person's lifetime and are present in histologically normal mucosa before cancer occurs. By using a neutral clonal marker, MAO-A, we have shown a difference in the mutation rate of the normal mucosa from patients with cancer and without. Although a relatively low sample size, we have still demonstrated a clear difference of 3.6-fold. This may indicate more genetic damage occurring in the colorectum of cancer. patient

P85

Lynch Syndrome Screening for Colorectal Cancer: The Provisional Results of a Two-Year Regional Programme

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Around 3% of colorectal cancer (CRC) is associated with Lynch syndrome (LS). All newly diagnosed CRC should be screened for LS according to NICE guidance yet most centres do not routinely offer this service due to lack of identified funding. Here we report the two year results of a regional LS screening programme offered to 16 multidisciplinary teams serving a population of 5.7 million. Screening was available for all patients diagnosed with CRC over the age of 50 years. Local centres were asked to refer a formalin fixed paraffin embedded tumour block (preferably biopsy) to the central laboratory for MLH1, PMS2, MSH2, and MSH6 immunohistochemistry. This was followed by BRAF codon 600 pyrosequencing and MLH1 promoter methylation analysis as appropriate. Twelve hospitals referred blocks from 2415 patients between May 2017 and March 2019 (66% biopsies/polyps and 34% resections). In total, 335 showed deficient mismatch repair (13.9%) of which 253 showed loss of MLH1/PMS2 (10.5%), 20 loss of MSH2/MSH6 (0.83%), 15 loss of isolated PMS2 (0.62%) and 13 loss of isolated MSH6 (0.54%). In addition, less common combinations of protein loss were observed in 34 cases (1.4%). Of the cases showing MLH1 loss, 75.5% contained a BRAF mutation. Of the wild type BRAF patients, 82.3% showed MLH1 promoter hypermethylation. Following screening, 68 patients were strongly recommended for LS germline testing (2.8%). Through a large regional LS screening programme, we have demonstrated a 14% rate of deficient mismatch repair and recommended 3% of patients to be referred for genetic counselling and consideration of germline testing. This will ensure that patients and their families are managed appropriately with regard to adjuvant chemotherapy decisions and the identification and subsequent management of LS.

P86

Analysis of TP53 Mutation Status and p53 Protein Expression in a Subset of 296 Patients Enrolled in the FOCUS4 Clinical Trial

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It is well recognised that the correlation between *TP53* mutation status and protein expression is limited. With *TP53* mutation status being determined by next generation sequencing (NGS), to guide patient randomisation in the FOCUS4 mCRC clinical trial, we took the opportunity to also assess, in parallel, p53 protein expression. *TP53* mutation status was determined using the Qiagen Clinically Relevant GenePanel, and run on a MiSeq sequencing platform. Protein expression was determined using the DAKO mouse monoclonal anti-human p53, clone DO-7. Both mutation and protein expression data were available for 296 patients. 212/296 (71.6%) of tumours contained a *TP53* mutation, with the remaining 84/296 (28.4%) being wild-type (WT). The ratio of biopsy to resection tumours in the *TP53*-WT cohort was 65.5:34.5, and in the *TP53*-mutant cohort was 52.8:47.2 (p=0.047). 60.8% of mutations were missense, 19.3% nonsense, and 11.3% in-frame deletions, with in-frame variants, intronic substitutions and splice acceptor variants constituting 2.8%. 5.7% were unknown. Minimum and maximum mutant allele frequencies (MAFs) were 6% & 86% and 6% & 85% in biopsy and resection tumours respectively. 136/212 (64.2%) of *TP53*-mutant tumours showed protein overexpression, 48/212 (22.6%) were negative, 19/212 (9%) showed patchy staining, with the remainder showing cytoplasmic expression. The nonsense mutation c.916C>T p.Arg306Ter was present in 8/9 tumours displaying cytoplasmic staining. Patchy staining was seen in 42/84 (50%) of WT tumours, but 31/84 (36.9%) showed overexpression, 9/84 (10.7%) showed no expression and 2/84 (2.4%) showed cytoplasmic staining. We have demonstrated aberrant staining patterns in 91% of *TP53*-mutant tumours, however, we have also seen this in 50% of the *TP53*-WT tumours, clearly demonstrating that protein expression is not a suitable surrogate when determining mutation status.

P87

Investigating the Effects of Radiotherapy on the Bowel Cancer Microbiome: Reanalysing the MRC CR07 Trial

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Purpose of study: In recent years, the importance of the gut microbiome in the biology of colorectal cancer is being increasingly recognised. However, there is a paucity of data with which to build a baseline measure of what constitutes a cancerous microbiome. Fortunately, bacterial sequences can be detected in existing genomic data, such as that produced for the MRC CR07 trial, a phase III trial studying total mesorectal excision +/– short course pre-operative radiotherapy.
Methods: Low coverage whole genome sequencing data of FFPE resection samples from 470 CR07 patients (224 RT+ versus 246 RT–) was reanalysed using the PathSeq algorithm, which searches for bacterial sequences.
Summary of results: A median of 9,783,000 sequence reads were processed per sample of which median 9139 were assigned to bacterial taxa. The numbers of bacterial reads did not significantly differ between the RT+ and RT– arms. Common gut bacteria such as *Bacteriodes* and taxa associated with cancer such as *Fusobacterium* were frequently observed. The frequencies of the taxa varied between individuals, but the frequencies within the cohort as a whole were similar to published results. Comparing the RT+ versus RT– arms, 19 taxa were differentially abundant. These were mostly isolated species, but did include several taxa in the *Firmicutes* phylum, found in the treated arm. Some of these, such as *Peptiniphilus* are known to infect wounds and other inflamed tissue.
Conclusions: We have shown that old data can be re-analysed in order to study bacterial populations. Any next-generation sequencing data could be examined in this way, or any trial cohorts could be sequenced with this analysis plan. In the case of the CR07 trial, we have shown that the profiles obtained previous findings, with a number of taxa linked to bowel cancer. The two arms of the trial were distinguished by taxa known to inhabit damaged tissue, which is entirely plausible given the radiotherapy treatment regime.

P88

The Potential Role of Raman Spectroscopy in Predicting Response to Pre-Operative Radiotherapy in Rectal Cancer

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High risk rectal cancer patients frequently receive neo-adjuvant radiotherapy (RT). However, there are currently no approved methods of predicting patient response, and a significant proportion of patients show no response at all. Raman spectroscopy is a non-destructive technique able to provide the unique chemical fingerprint of tissues, detecting changes in molecular composition prior to visible morphological changes. Using Raman spectroscopy, we aimed to build a classification model capable of predicting patient response to pre-operative short course RT, in order to guide personalised patient treatment. High quality Raman spectra were collected from formalin fixed paraffin embedded (FFPE) biopsy and resection samples from 10 rectal cancer patients, who had received pre-operative short-course RT. Cases were selected based on response to RT, determined by calculating the percentage reduction in tumour cell density (TCD), following pre-operative RT. Regions of interest were selected using digitalised H&E stained sections and the spectra collected from the corresponding unstained sections using an inVia Raman confocal inverted microscope. The same regions were then re-mapped using a Renishaw benchtop RA816 Raman spectrometer. The cases that showed a poor response to pre-operative RT had a TCD reduction of less than 36%, and those showing a good response had a reduction in TCD greater than 75%. Our preliminary results, as presented in 2018 showed a principal component analysis - linear discriminant analysis (PCA-LDA) classification model was able to differentiate areas of tumour and stroma within patient resections. These results have since been supported by data from additional patient samples, and current analysis is underway to confirm reproducibility of the model using an alternative Raman spectrometer. From this, further work is ongoing to build a classification model capable of predicting response to pre-operative RT, from spectra collected from patient biopsies.

P89

Central Pathological Review of the International Phase III FOxTROT Trial: Comparison with the Local Pathological Evaluation

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FOxTROT is the first international phase III randomised clinical trial to evaluate the effectiveness of pre-operative chemotherapy in locally advanced colon cancer. Patients were randomly assigned in a 2:1 ratio to pre- and post-operative chemotherapy or to post-operative chemotherapy alone. Local pathology evaluation was undertaken in 94 centres in the UK, Sweden and Denmark. H&E stained slides were available for central pathological review in 904 out of 1052 cases (86%). All slides were scanned at 20x magnification using an Aperio XT and viewed digitally using Aperio ImageScope. The central assessment was performed blinded to the original local evaluation by a single observer for all of the microscopic pathological features captured locally on the case report form, where these were assessable. Staging was performed according to TNM version 5. The overall concordance rates between the central pathological review and the local pathological evaluation were very good. Specific items assessed and the associated concordance included: pT stage (95%), pN stage (94%), resection margin status (99%), intramural venous invasion status (89%), extramural venous invasion status (89%) and Dukes' stage (96%). The local pathological evaluation in the FOxTROT trial has been confirmed to be carried out to an excellent standard across a large number of centres in three countries. Concordance with the central pathological review is excellent for most histopathological features, with a slightly lower rate of agreement for venous invasion, which is recognised to be frequently missed by pathologists. Central pathological review should ideally be built into all clinical trials using histopathological endpoints to confirm the quality of local reporting.

P90

Alcian Blue: A Rediscovered Biomarker of Poor Prognosis in Gastric Cancer Patients

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Background: Gastric carcinoma (GC) is one of the major causes of cancer related deaths worldwide. GC mucin phenotype has been related to tumour invasion and genetic alterations. However, the relationship between mucin phenotype, clinicopathological variables and GC patient survival remains a matter of debate.
Methods: Tissue microarrays from 709 GC resections were stained immunohistochemically for MUC2 (intestinal-type mucin) and MUC5AC (gastric-type mucin) and histochemically for Alcian Blue (AB) periodic acid-Schiff (PAS) (acidic and neutral mucin). Stainings were scored using a 10% cut off to define positivity. The relationship between marker, clinicopathological variables and survival was analysed.
Results: 16% GC were MUC2 positive, 36% MUC5AC positive, 6% AB positive, 3% PAS positive and 11% AB and PAS positive. 4% GC were triple positive, 49% triple negative. Expression of MUC2, MUC5AC and ABPAS staining was related to GC histological phenotype ($p<0.05$). AB positivity was related to deeper invasion ($p=0.006$) and poorer grade of differentiation ($p<0.001$). Patients with AB negative GC (n=543) survived significantly longer than those with AB positive GC (n=112), $p=0.001$. Survival of patients with PAS positive GC (n=20) was similar to those with AB negative GC. There was no relationship between MUC2 or MUC5AC and patient survival.
Conclusions: This is the first study to show that patients with AB positive GC are more likely to have locally very advanced disease, poorly differentiated GC and poorer survival. The underlying biological mechanisms related to the switch from PAS positive mucin in the normal gastric mucosa to acidic/intestinal type AB positive mucin in GC are currently unclear and warrant further investigations.

P91

Routine CT Scan One Year After Surgery can be Used to Estimate the Level of Arterial Ligation in Colon Cancer Surgery and May be More Accurate Than Standard Histopathological Measurements

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Complete mesocolic excision (CME) is increasingly being used to optimise colon cancer surgery and involves central ligation of the tumour feeding vessel at its origin. We have previously shown that measuring the arterial stump on a CT-scan performed two days after surgery renders a reliable estimate of the level of artery division. The aim of this study was to identify and measure the arterial stump on a routine CT-scan one year after surgery, and compare the results with those obtained two days after surgery. 52 patients had surgery for colon cancer followed by a CT-scan two days after surgery. One year after surgery, 47 patients had a CT-scan as a part of the standard follow up. Both the images were evaluated by the same specialist radiologist. The vessels were identified and measured from the origin to the point of ligation. In 38 cases (81%) we were able to identify and measure the arterial stump on both scans. Overall, we found no difference in the length of the vessel one year after surgery compared to the length after two days (mean difference -1.7 mm; 95% CI (-3.8 to 0.5 mm), $p=0.13$). However, vessels categorized as thrombosed or as a fibrotic line were shorter after one year (mean difference -4.5 mm; 95% CI (-8.9 to -0.1 mm), $p=0.05$). Routine CT-scans obtained one year after surgery can be used to estimate the level of arterial division following colon cancer surgery. These measurements are highly likely to give a more accurate impression of the level of arterial division when compared to standard histopathological measurements performed on the specimen, given that vascular anatomy is highly variable and that histopathologists cannot comment on what has been left behind in the patient. The level of arterial division (assessed radiologically) should be combined with the plane of mesocolic surgery (assessed pathologically) to ultimately determine the quality of the specimen.

P92

The Creation and Validation of a Global Microbiome Colorectal Cancer Research Network

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Research investigating the colorectal cancer (CRC)-associated microbiome has been almost entirely conducted in 'Western' high CRC incidence countries. Funded by the UK Academy of Medical Sciences Global Challenge, we established a global research network to compare the CRC-associated microbiome of high (UK and Argentina), intermediate (Chile) and low (India and Vietnam) CRC incidence countries. Faecal samples were collected using bowel cancer screening cards from 10 CRC patients and 10 healthy volunteers from each country and transported to the UK at room temperature. Replicate control samples from 5 UK healthy volunteers were generated to assess for the effect of transportation and storage abroad. V4 16SrRNA sequencing was performed. Here we present the results of samples from India, Vietnam and Argentina, as samples from Chile were not available at the time of sequencing, but are currently being processed. There were no significant differences in bacterial community between any of the UK replicate controls, indicating that transport and local storage of samples does not alter microbiome results. There was no significant difference in the alpha diversity of samples from the different countries, but the beta diversity differed significantly for both weighted and unweighted UniFrac distances. Beta diversity differed significantly between the combined healthy volunteer group and the combined CRC group for unweighted but not weighted UniFrac distance, indicating that rare taxa account for this difference. These include *Fusobacterium*, *Parvimonas*, *Porphyromonas* and *Escherichia-Shigella* which are CRC-associated bacteria described in the literature. We have demonstrated a robust method of conducting global CRC microbiome research. The microbiome differs by country and differences in rare taxa exist between the combined microbiome of healthy volunteers from India, Vietnam and Argentina compared with CRC patients. Plans are underway to expand the network and sample collection.

P93

The Use of Stochastic Optical Reconstruction Microscopy (STORM) in Formalin Fixed Paraffin Embedded Tissue

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Background: Colorectal cancer (CRC) with amplification/over-expression of cell surface receptors or ligands are specifically targetable (e.g. HER2 and anti HER2 therapy, EGFr and anti EGFr therapy) but most patients selected for these therapies fail to respond due to unknown mechanisms of resistance. Developments in advanced fluorescence microscopy have made it possible to resolve protein localisation at up to 5nm resolution. In this pilot study we aimed to develop a robust routine methodology for Formalin Fixed Paraffin Embedded tissue (FFPE) that exploits this technology and explore its potential for visualisation of ligand-receptor pathways in CRC to increase our understanding of resistance mechanisms at a cellular level.
Methods: To establish the protocol on FFPE tissue we selected colorectal cancer cases with strong HER2 and negative HER2 receptor expression. Previously described cell culture protocols (Creech et al. 2017) were modified, optimised and imaged by confocal microscopy using HER2 (1:250) with affinity purified secondary antibody (1:500). The optimised protocol was used in 3D dSTORM on FFPE.
Results: Protocols for HER2, EGFR, RAB5 and RAB11 expression on FFPE samples were determined using the confocal microscope. In 3D dSTORM, high levels of HER2 were localised to aggregates in the membrane and lower levels in the cytoplasm. Further work will focus on imaging and quantification of further components of the MAPK/ERK pathway in 3D dSTORM and imaging multiple proteins in combination to assess ligand-receptor and receptor-adapter interactions as well as receptor cycling. STORM microscopy opens up subcellular microscopy in FFPE to Histopathologists.

P94

Comparison of Two Methods to Analyse Components of the Microbiome from FFPE CRC Tissue: Low Coverage WGS and qPCR

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Microbiome research is rapidly advancing, with particular focus on the association with colorectal cancer (CRC). Prospective studies, which typically perform metagenomic or 16SrRNA sequencing on faeces or fresh-frozen tissue from CRC patients and healthy volunteers, have identified specific bacteria as being significantly enriched in CRC patients. One, *Fusobacterium nucleatum* (*F.nucleatum*), is associated with chemo-resistance and poor prognosis. Additional clinically-relevant associations could be discovered through the assessment of archival FFPE CRC clinical trial material. For trials which have had low coverage whole genome sequencing (LCWGS) performed, it is possible to interrogate the WGS data for reads which map to the genomes of the specific bacteria of interest or to perform quantitative polymerase chain reaction (qPCR). Here we compare the two methods, taking TaqMan qPCR as the gold standard. qPCR amplification of *F.nucleatum* was performed in triplicate using DNA from 148 CRC samples, for which LCWGS (less than 1x) data was also available. qPCR amplification of the human prostaglandin transporter gene was used as a reference and (2^-deltaCt) was calculated. The LCWGS data was aligned to a composite artificial genome of human and bacterial sequences using BWA aligner and *F.nucleatum* load was calculated.
The Pearson correlation coefficient for the relative abundance of *F.nucleatum* between the two methods was strong (0.74). Compared with qPCR, LCWGS had a sensitivity of 66% (63/96) and positive predictive value of 69% (63/91) for the detection of *F.nucleatum*. We have shown that existing LCWGS data from FFPE CRC clinical trial material is valuable for the investigation of the presence and relative abundance of *F.nucleatum* but has a lower sensitivity than TaqMan qPCR. We recommend performing analysis of existing LCWGS data from archival CRC clinical trial material in order to generate hypotheses, with subsequent validation by TaqMan qPCR.

P95

When To Biopsy? Compliance With Local Guidance for Effective Endoscopic Biopsy Practice

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Purpose of study: Our local guidance 'When to Biopsy' was first written in 2002 following the Royal College of Pathologists 'Specimens of limited clinical value', to ensure appropriate use of endoscopic biopsy resources. It has been updated periodically in discussion with endoscopists, and is displayed in all local and external endoscopy suites. New BSG/AUGIS standards in upper gastrointestinal endoscopy were published in 2017 (Beg S et al, Gut 2017). Before updating our own biopsy guidance, we audited compliance with existing guidance.
Method: The endoscopic biopsies reported during 1 week in March 2018 were assessed prospectively for compliance with the guidance provided. The indication, site, number of biopsies, endoscopist and pathologist were recorded. Following this, further consecutive biopsies taken in the independent endoscopy units were assessed in the same way, to compare with the Trust cohort.
Summary of results: 79% (89/112) of Trust endoscopy suite biopsies conformed to the guidance. Of the 23 that did not, 16 were an incorrect or incomplete series, and 7 had no clinical indication. 41% (29/70) of external endoscopy unit biopsies conformed, significantly less than within the Trust ($p<0.0001$). Of the 41 that did not, 11 were an incorrect or incomplete series, and 30 had no clinical indication. An incomplete series for diagnosis of microscopic colitis was the most common 'incorrect/incomplete series'.
Conclusions: Our audit showed a 79% compliance within the Trust. Overall, improved compliance would not have affected biopsy numbers. Compliance was lower in endoscopy outsourced to independent units, usually due to biopsies of normal mucosa without clinical indication. We have updated our local guidance in line with BSG/AUGIS standards and plan to re-audit practice. There is a national shortage of pathologists and focus on efficient working – we have found our guidance effective, but needs to be provided in the context of close working with endoscopists.

P96

Stage is Not Predictive of Time to Recurrence in Colorectal Cancer

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Introduction: Colorectal cancer (CRC) has a variable presentation, treatment options and prognosis depending on multiple underlying factors such as site, molecular signature and invasion. While many of these risk factors have been identified and investigated, their interactions and effects on prognosis are not fully understood. The recent update to Royal College dataset incorporated an updated understanding of a number of these risk factors; we sought to test a number of these risk factors and others using a large tissue-linked pathology database.
Methods: We identified 1000 recent cases of colorectal cancer from our local pathology database, including full pathology reports of surgical biopsies and resections (initial diagnosis from 2008 to 2014). Patient demographics and follow-up data were gathered from the hospital patient information system. Survival analyses including overall survival, cancer-free survival and cancer-related mortality were performed, according to known risk-factors for validation. We tested the effect of overall stage on time to recurrence, overall survival and cancer-related mortality.
Results: High overall stage was highly predictive of adverse outcome, with people with stage 1 CRC having a mean survival of 101 months (95% CI: 95 – 108 months), stage 2: 85 months (95% CI: 80 – 89), stage 3: 75 months (95% CI: 70 – 80) and stage 4: 34 months (95% CI: 28 – 40). Similarly, cancer related death was strongly predicted by higher overall stage. While higher stage predicted number of recurrences, we found that time to recurrence was not related to overall stage. The mean time to recurrence for all stages was 22 months (95% CI: 20 – 24) and after 36 months the likelihood of recurrence was less than 10%.
Conclusion: Almost all colorectal cancer recurrences occur within 3 years of primary diagnosis. Although stage is associated with higher risk of recurrence, it does not predict time to recurrence.

P97

Presence of Vascular Invasion in Colorectal Cancer Correlates with TNM8 Nodal Status Classification

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Purpose of the study: Vascular invasion (VI) is an important prognostic marker in several malignancies including colorectal cancer (CRC). The correlation between the VI status and the revised nodal stage of CRC as per TNM8, was investigated in this study. Methods: 1000 consecutive cases of CRC from a tertiary centre, diagnosed between 2008-2014 were investigated for VI profiles from histopathological reports on IT records and results correlated to clinicopathological variables including the revised nodal stage as defined by the TNM8 classification.
Summary of results: 483 of 985 (49%) cases showed extramural vascular invasion (EMVI), the incidence of which within stages 1-4 of CRC was 1%, 44%, 68%, 72% respectively (p<0.001). Positive VI status was associated with higher grade, nodal status, distant metastases and recurrence (p<0.001). Within the revised nodal stages as per TNM8, positive VI status was seen in 34% (N0), 58% (N1a), 64% (N1b), 85% (N2a), 96% (N2b) of cases (p<0.001) Intramural VI (282/916) also correlated with tumour stage (p<0.0001), distant metastases (p=0.003) and recurrence (p=0.002). Within the revised nodal stages as per TNM8, intramural VI was seen in 25% (N0), 33% (N1a), 30% (N1b), 46% (N2a), and 65% (N2b) cases (p<0.001). Presence of intramural VI in the absence of EMVI however did not correlate with lymph node metastases and recurrence. EMVI conferred poorer 5 and 10 year overall and cancer specific survival (p<0.001) and in N0 cases, was correlated with worse overall survival (p=0.001).
Conclusions: Overall, this study shows that both intramural and extramural VI in CRC have strong correlations with the increasing nodal stage, as defined by the TNM8 classification. Revalidation of prognostic associations of VI with clinicopathological variables and survival in novel datasets of CRC establishes confidence in the the cohort for subsequent biomarker/molecular analysis.
Supported by the Nottingham Molecular Pathology Node.

P98

A Novel 3D Cell Culture System Demonstrates Induced Pluripotent Stem Cells Driven to Intestinal Differentiation are Capable of Spontaneous Crypt Assembly

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Introduction: Induced pluripotent stem cells (IPSC) are an important tool for studying development and disease and give the potential for personalised tissue regeneration in future. While more is known about the fundamental signalling pathways driving IPSC to differentiate along specific tissue lines, much of the evidence for differentiation is derived from molecular assays or organoid assays. Although these form vital evidence, molecular assays lack the morphological context of cells in a 3D environment and organoids do not always faithfully recapitulate the tissue organisation of cells within a luminal organ. The aim of this study was to develop a long-term 3D culture system of the intestinal mucosa and sub-mucosa from IPSC, which would allow conventional histological and immunohistochemical methods to be used for analysis.
Methods: IPSC were driven to intestinal differentiation using a novel serum-free protocol incorporating Wnt and Nodal pathway activators for 8 days. These intestinal progenitors were seeded onto the surface of pre-cast 3D culture gels consisting of purified extracellular matrix components. Cells were maintained in culture medium supplemented with growth factors promoting growth and maturation. After 4 weeks of culture, the gels were formalin-fixed and paraffin-embedded and sectioned for histology and immunohistochemistry for intestinal markers.
Results: Cells formed a polarised epithelium on the surface of the gel with an underlying mesenchymal component. The mucosa was capable of spontaneous intestinal crypt assembly. Cells showed expression of a range of intestinal markers in the epithelium and mesenchymal markers in the submucosa. There was no evidence of aberrant differentiation or retained pluripotency.
Conclusion: Using a novel 3D culture, it has been possible to demonstrate that IPSC driven to intestinal differentiation are capable of forming mature intestinal mucosal and submucosal structures.

P99

Faux-Multiplex Immunohistochemistry (fm-IHC) to Delineate Biomarker Territories and Biomarker Co-Localisation in Colorectal Cancer

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Colorectal cancers (CRC) are heterogeneous. Mapping the geographical expression pattern and the co-localisation of biomarkers within a tumour would allow activated pathways and cellular communities to be defined. This is possible with immunofluorescence but multiplex immunohistochemistry (IHC) is beset with problems such as antibody cross reactivity and tissue degradation. In this project, we aim to build on the registration facility of Histogenic Molecular Mapping (HMM) to establish faux-multiplex immunohistochemistry (fm-IHC) as a novel approach for multiple biomarker analysis. Sequential whole tissue sections (WTS) of FFPE blocks of CRC were subjected to IHC for stromal, epithelial, and various immune cells markers. The staining was carried out automated slide stainer (Ventana) and digital slides were produced using Aperio slide scanner (Leica). For each block, all consecutive IHC images were registered to the middle one in the block to establish accurate correspondence across all biomarkers (a particular landmark in one registered IHC image can be found at the same position in all the other registered image). Each registered image was then downsampled by a factor of 10 to account for small differences across consecutive images and small registration inaccuracies. Subsequently, the images were automatically colour separated and thresholded to obtain expression maps and to establish correlations across combination of biomarkers. Fm-IHC analysis of BerEP4, CD3 and CD20 was undertaken in 20 CRC which were mismatch repair deficient (dMMR, n=10) or proficient (pMMR, n=10). The intra-epithelial lymphocytes (BerEP4+/CD3+ frames) and the stromal lymphocytes (BerEp4-/CD3+ frames) could be identified. Overall, dMMR tumours had a higher density of CD3+ cells but there was no difference in the compartmental distribution (intra-epithelial vs stromal). There was significant negative association of CD3 and CD20 in the medulla and germinal centre mantle of lymph node.

P100

Hepatocyte Nuclear Factor 4A is a Novel Tumour Suppressor in Pancreatic Cancer

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The dismal prognosis of pancreatic cancer due to the delayed diagnosis, rapid metastasis and resistance to current therapeutics, signifies the importance of identifying novel therapeutic approaches.We analysed global alterations at the epigenetic and genetic level, in 20 pancreatic cancer and 14 normal tissues, and explored for overlapping changes in DNA methylation and gene expression. This analysis revealed hepatocyte nuclear factor 4A (HNF4A), a key transcription factor in the development and proper function of the pancreas, as a novel target for aberrant DNA methylation.Functional in vitro studies, using four different pancreatic cancer cell lines, indicate that the HNF4A acts as a tumour suppressor, regulating pancreatic cancer growth, invasiveness and chemoresistance. Additionally, our findings were verified in vivo by employing two different mouse models: a xenograft and a genetically engineered mouse model of pancreatic cancer. To elucidate the clinical significance of our findings, we conducted immunohistochemistry in two different cohorts of tissues: 1) a tissue microarray containing 154 tissue specimens from pancreatic cancer and matched non-neoplastic tissue samples. 2) 8 different tissue microarrays containing in total 225 tissue specimens from pancreatic cancer and 50 normal tissue samples. Scoring of the immunostained tissues was assessed using a double-headed microscope. Multiplication of the percentage of cells with staining intensity ordinal value (scored from 0 to 3) was performed. The results revealed a statistically significant suppression of HNF4A (70%) in human pancreatic cancer. Clinicopathological analysis revealed an inverse correlation between HNF4A expression and tumour size (T) or grade (G). Importantly, low HNF4A expression correlates with poor survival.Our data suggest that HNF4A plays a central role in pancreatic cancer progression and its perturbation holds promise as a novel therapeutic approach.

P101

Cten Ability to Induce Migration is Dependent on its SH2 Domain

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Cten (Tns4) is a member of the tensin family and acts as an oncogene in colorectal cancer where it enhances EMT, migration and invasion but not proliferation. It is normally localized in the cytoplasm but it was found that it translocates to the nucleus where it binds to beta-catenin. The mechanism by which Cten induces EMT, invasion and migration is not fully understood. The structure of Cten has an SH2 domain. The SH2 domain is found in different proteins and has a very important role in regulating signaling cascades. SH2 domains play a vital role in interacting with different receptors in several cancers. Upon interaction, signals are transmitted through several pathways to induce growth and metastasis in different types of cancers. We performed Site-directed mutagenesis to delete the SH2 domain of Cten plasmid. We then transfected HCT116 with Empty vector, wild-type Cten, and Cten(Δ SH2). This was followed by functional assays, protein extraction and western blot. Our data from functional assays confirms Cten overexpression is inducing cellular migration but not proliferation in HCT116. Moreover, we have discovered that the SH2 domain is essential for Cten to induce migration. Moreover, our data illustrate that Cten regulates E-cad, Snail, N-cad and ROCK1 through SH2 domain. We have shown by western blot that Cten is Upregulating Snail, ROCK1 and Ncad and downregulating Ecad. When the SH2 domain was deleted, the expression of these proteins was restored.
Conclusion: Cten induces migration in Colorectal Cancer through its SH2 domain. Cten regulation of downstream targets is controlled by its SH2 domain

P102

Evolution of Chronic Lymphocytic Leukaemia / Small Lymphocytic Lymphoma

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Introduction: Chronic lymphocytic leukaemia/small lymphocytic lymphoma (CLL) is common B cell malignancy with heterogenous behaviour. Certain cases present with atypical morphological/immunophenotypic features; others develop these features over time. Many such features indicate poorer prognoses. The current study aims to characterise these CLL variants.
Methods: Retrospective review of CLL cases performed in Hammersmith Hospital archives. Atypical CLL (atCLL; n=58; cleaved nuclei), aggressive CLL (agCLL; n=17; diffuse/confluent proliferation centres) and Richter transformation to diffuse large B cell lymphoma (RT; n=14) cases were compiled. Comparisons made with classic CLL (cCLL; N=71). Available data collected on the immunophenotype on paraffin-embedded tissue and cytogenetics.
Results: Mean age at diagnosis was 71.1 years. No significant age difference noted across CLL types. Tissue from lymph node (48%, N=78), bone marrow (40%, N=64) and other sites (11.3%, N=18) with more extra-nodal/extra-marrow sites in RT (Chi Square, $p<0.001$). A significant difference in Ki67 index noted across the groups (ANOVA, $F=66.57$), $p<0.001$). Post hoc comparisons showed RT Ki67 (M=70, SD = 16.4) was higher than agCLL (M=54.3, SD=13.2) which was greater than either atCLL (M=22, SD=9.7) or cCLL (M=22.9, SD=10.1). There was a significant difference in CD5 (Chi-Square, $p<0.001$; lower in RT), CD23 (Chi Square, $p<0.001$; lower in RT), MUM1 (Chi Square, $p<0.01$; higher in RT and agCLL) and BCL6 (Chi-Square, $P<0.001$; higher in RT and agCLL) expression across the groups. No significant difference in Lef-1, CD38, BCL3, EBER, Pax5, CD21, IgM or IgD expression was seen across the groups. No significant differences in trisomy12, del(11q), del(17p) or del(13q) between atCLL and cCLL.
Conclusions: Morphological CLL variants have corresponding immunophenotypic changes. Clinical correlation is underway. Further genetic testing will be necessary to define the precise genetic chages across these types.

P103

OCT2 as a Diagnostic Immunohistochemical Marker in Rosai-Dorfman Disease

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Here we characterise a consecutive series of RDD cases diagnosed in our regional haematopathology diagnostic service, demonstrating that histiocytes in all RDD cases in our population are characterised by the strong expression of the transcription factor OCT2. A consecutive series of RDD cases diagnosed between 2008-2018 according to standard practice at the regional HMDS diagnostic service were identified. Diagnostic criteria were based on characteristic morphology and S100+ reactivity of sinus histiocytes. Staining for S100, CD163 , CD1a , CD68, Langherin and OCT2 with mouse mAb. Antigen retrieval on FFPE sections was carried out using DAKO PT links with high antigen retrieval buffer with antigen retrieval at 95°C for 15 min. DAKO Envision Flex+ kit was used with standard detection methods with DAB chromogen and haematoxylin counter stain. The average patient age at diagnosis was 43 years and six months (range 3 years to 81 years). There were slightly more female than male cases (62% female, n=16). 35% (n=9) cases had recurrent disease. 50% (n=13) cases had nodal disease at presentation, 38% (n=10) had extra-nodal disease and 19% (n=5) had both nodal and extra-nodal disease at presentation. The most commonly affected sites were head and neck 62% (n=16), breast 19% (n=5) and skin and soft tissue 31% (n=8). Of the cases stained for S100, 98% were positive, 76% were CD68 positive, 100% were positive for CD163. None of the cases stained for Langherin or CD1a were positive. Two patients with recurrent disease had IHC performed on their recurrence specimens, which also demonstrated clear nuclear positivity for OCT2. Control cases did not show strong nuclear positivity for OCT2. Here, we have demonstrated that OCT2 is a reliable immunohistochemical test for the diagnosis of RDD. This antibody is widely used in clinical practice so can be readily applied in this alternate diagnostic setting.

P104

Identification of Suitable T-Cell Associated Transcripts for the Development of a New Veterinary Diagnostic Test for T-Cell Lymphoma

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Lymphoma is one of the most frequently encountered malignancies in veterinary practice, particularly in cats and dogs, but veterinary pathologists struggle with current diagnostic techniques to distinguish T-cell lymphomas from infiltrates of benign T-cells. Clonality studies are possible for cat and dog, as in human clinical pathology, but these are complex and time-consuming with variable success rates. In human pathology, we have developed a new chromogenic in situ hybridisation (CISH)-based assay for formalin fixed paraffin embedded histological samples to look for T-cell monotypia, in a manner analogous to kappa/ lambda for B-cells. We determine the ratio of the TRBC1: TRBC2 constant segments in T-cell populations. Significant skewing away from the normal 1:1 ratio indicates likely lymphoma. In order to apply this approach to veterinary samples, we set out to identify the animal sequences, to determine their relative levels of expression and to provide preliminary CISH-staining data. By aligning publicly available sequence data with the human TRBC1/2 sequences, the T-cell receptor constant regions, TRBC1 and TRBC2, were predicted for cat, dog and mouse. These were amplified from cDNA samples by PCR and confirmed by Sanger sequencing. Sequences from multiple animals showed greater polymorphism in cat and dog, with implications for CISH probe design. As for the human sequences, the 3' untranslated region shows the greatest variation between TRBC1 and TRBC2, making this a promising site for segment-specific CISH probe design. Q-PCR indicated a TRBC1: TRBC2 ratio close to 1:1 in cat, dog and mouse. TRBC1 and TRBC2 specific probes, produced by PCR and labelled with digoxygenin, gave excellent CISH staining for both TRBC1 and TRBC2 in lymphoid cells and on FFPE mouse spleen tissue. In summary, we have demonstrated that CISH-based detection of TRBC1/2 could have utility as a test for animal T-cell lymphoma as it is likely to have for human T-cell lymphoma.

P105

MYC Translocation-Positive Diffuse Large B-Cell Lymphoma: The Clinicopathological Impact of Copy Number Gain of the Translocated *MYC* Allele

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MYC rearrangement occurs in ~10% of diffuse large B-cell lymphoma (DLBCL). This can occur as an isolated single-hit (SH) or, frequently, in association with *BCL2* and/or *BCL6* rearrangement known as double-hit (DH) or triple-hit (TH). DH and TH are associated with a poor prognosis and routinely screened for by fluorescence in situ hybridisation (FISH) at histological diagnosis. Often, FISH also demonstrates copy number change at the *MYC* locus including gain of extra copies of the translocated allele or amplification of the intact allele. The clinicopathological impact of these changes is unclear. We identified 21 cases of DLBCL with increased *MYC* copy number, including gain of 1-3 extra copies of the translocated *MYC* allele (*MYC/BCL2/BCL6*-TH=5, *MYC/BCL2*-DH=6, *MYC/BCL6*-DH=2, *MYC*-SH=4), or amplification (>4 copies) of the intact *MYC* allele without *MYC* translocation (MYC-amp). The median age was 65 years (range 43 - 83 years) and 57% were male. 6/21 cases showed blastoid morphology with a "starry sky" appearance on histological review. Immunohistochemistry revealed high MYC protein expression indices (≥60%) in the majority of cases (16/21) and GC-phenotype in 95% according to Hans algorithm. Targeted sequencing of a panel of 70 genes associated with DLBCL demonstrated frequent mutations in *KMT2D* (57% of cases), *TP53* (52%), *FOXO1* (33%) and *MYC* (33%). Interestingly, *FOXO1* mutations occurred exclusively in DH/TH cases (64% vs 0%, p = 0.004, Fisher's exact test), whilst *TP53* mutations mostly occurred in the cases with *MYC*-SH or *MYC*-amp (90% vs 18%, p = 0.002). Of the 17 patients that had *MYC* translocation and extra copies of the translocated *MYC* allele, 9 died within the first year of diagnosis and *TP53* mutation was associated with a poorer prognosis (p = 0.05). Our findings warrant more extended investigation of the clinicobiological impact of copy number gain of the translocated *MYC* allele, particularly in comparison to relevant cases without *MYC* copy number change.

P106

Peripheral T-Cell Lymphoma, NOS with Aberrant Expression of CD20: Report of Two Cases

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Introduction: Peripheral T-cell lymphoma, NOS is a heterogeneous category of nodal and extranodal mature T-cell lymphomas that is associated with an aggressive clinical course. Aberrant expression of CD20 in PTCL is rare and the clinical and prognostic implications of this remain largely uncertain.
Case presentation: The first case concerned an 85 year old man who presented with a mediastinal mass. A bone marrow trephine showed heavy marrow infiltration by sheets of small to medium sized atypical lymphoid cells. Flow cytometry showed most of the cells were T cells that were positive for CD2, CD4, CD5 and CD56 with expression of CD20. CD7 was negative. Immunohistochemistry showed similar findings and the cells were also positive for CD20 in addition to expression of cytotoxic proteins TIA1, Granzyme B and perforin. CD79a and PAX5 were negative. The second case concerned an 80 year old man with widespread lymphadenopathy. A lymph node biopsy showed replacement of the architecture by a dense infiltrate of medium to large sized atypical lymphoid cells that surrounded numerous small collections of epitheloid histiocytes. Immunohistochemistry showed the lymphoid cells were BF1 positive T-cells co-expressing CD2, CD3, CD5, CD7 and cytotoxic proteins TIA1, Granzyme B and perforin. The cells showed variable aberrant expression of CD20. CD79a and PAX5 were largely negative. In both cases PCR showed clonal rearrangement of the TCR beta and gamma genes with no evidence of a B cell clone. The features of both were consistent with a peripheral T-cell lymphoma, NOS with aberrant expression of CD20.
Conclusions: CD20 expression in peripheral T cell lymphoma is rare and can be a diagnostic pitfall. Performing a wide panel of immunohistochemistry with flow cytometry and clonality studies is often needed to identify and correctly diagnose these cases. Aberrant expression of CD20 in these aggressive lymphomas can provide an additional therapeutic target for Rituximab.

P107

Composite Hemangioendothelioma: A Clinicopathological Study of Site, Composition and Immunohistochemistry of Three Cases

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Introduction: Composite Hemangioendothelioma (CHE) is a recently described new entity classified under the Hemangioendothelioma group of vascular tumours, it was first described in 2000 by Nayler et al, and it is a rare vascular neoplasm of low malignancy that exhibits a composite of hemangioendothelioma variants. It is a complex heterogenous vascular tumour with infiltrative margins. It usually presents on the skin and soft tissue of upper and lower extremities, especially the lower leg and foot. Other sites are the head and neck region chiefly the tongue, mandibular vestibule, cheek mucosa, scalp, nose, and hypopharynx.
Methods: Three cases of CHE were retrieved from our archives. Blocks were cut and stained with H&E. Immunohistochemical stains were done using DB Biotech protocol. Antibodies used include CD34, EMA and vimentin.
Results: All cases of CHE encountered in this study were in the pediatric age group that is 8, 10 and 14 years respectively. The sites of presentation of CHE are head and neck, upper and lower extremities. Histology shows composite of hemangioendothelioma variants that is, Dabska, retiform, epitheloid and spindle cell hemangioma in varying proportions with brisk mitosis seen. All were positive for CD34 and vimentin but negative for EMA.
Conclusion: CHE can present with composite of hemangioendothelioma variants. In our environment this vascular lesion of low malignant potential is see in the pediatric age group.

P108

Modelling Rhabdomyosarcoma Using the Chick Embryo Model

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Rhabdomyosarcoma (RMS) is the most common soft tissue sarcoma affecting children and adolescents. Localised disease has a 5-year survival rate of >70% following multimodal therapies, however patients with recurrent disease have a much worse overall survival. In other paediatric malignancies (neuroblastoma) adaptations to hypoxia have been shown to results in an aggressive cancer cell phenotype. The role of hypoxia in RMS is not as well understood and the purpose of this study is to investigate the role of hypoxia in RMS tumourigenesis with a view to develop novel therapeutics.
Methods: GFP labelled RD cells (embryonal RMS derived cell line, GFP-RD) were subject to in vitro hypoxic or normoxic environments. Cellular proliferation was assessed using sulforhodamine B (SRB) assay. HIF-1α expression, a marker of cellular adaptation to hypoxia, was analysed using western blotting. Hypoxic and normoxic pre-conditioned GFP-RD cells were transplanted onto the chick embryo chorioallantoic membrane (CAM) and tumour nodule forming efficiency (NFE), size and metastases were assessed using brightfield/fluorescent imaging.
Results: SRB assays demonstrated greater cell proliferation in GFP-RD cells incubated for 24 hours in hypoxic vs. normoxic conditions. HIF-1α expression was greatest in GFP-RD cells incubated for 24 hours in hypoxic conditions. Both normoxic and hypoxic GFP-RD cells formed discreet nodules on the CAM but metastases were not seen. Hypoxic GFP-RD cells appeared to form larger nodules but their NFE was less than normoxic GFP-RD cells
Conclusion: HIF-1α may drive cell proliferation in hypoxic GFP-RD cells which subsequently form larger tumour nodules in the CAM model than normoxic cells but with less NFE. Future work will investigate these findings to further define the role of HIF-1α, angiogenesis and pattern of invasion.

P109

Characterising the Response of a Metastatic Uveal Melanoma Cell Line to Hypoxia Using a Chick Embryo Model

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Uveal melanoma (UM) is a rare disease, but is the most common primary intraocular malignancy found in adults. It has an average incidence of 5 cases per million per year in the US and in Europe. Despite effective treatment of the primary tumour, metastatic disease will affect approximately 50% of UM patients, typically affecting the liver. A lack of adjuvant therapy leads to a poor prognosis for these patients and an urgent need for new therapies. Drivers of metastatic UM disease are poorly understood, however in other cancer types hypoxia is associated with enhanced tumour vascularisation and increased cancer cell survival in tumour microenvironments. This study investigates the role of hypoxia in UM and tumour progression, using the chorioallantoic membrane (CAM) assay in the embryonic chick model. The GFP-labelled metastatic UM cell line, MM66, was analysed for cell proliferation and HIF-1α expression in hypoxic (1% O_2) and normoxic (21% O_2) conditions. MM66 cells were grafted onto the CAM to analyse tumour nodule formation and metastasis using fluorescent imaging and histology. MM66 cells grown in hypoxia were found to have an increased rate of proliferation compared to normoxia. HIF-1α expression in hypoxic cells was greatest at 72 hours incubation and this was used to culture MM66 cells before implantation onto the CAM. Normoxic and hypoxic MM66 cells formed nodules on the CAM. This study was able to conclude that hypoxia is a driver for proliferation of MM66 cells *in vitro*. Both hypoxia- and normoxia-conditioned cells form tumour nodules on the chick CAM and further experiments will investigate the effects of hypoxia on tumour histology and metastatic development.

P110

Distinguishing Neurosarcoidosis from Multiple Sclerosis Based on Cerebrospinal Fluid Analysis and Oligoclonal Bands: A Cohort Study

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Distinguishing neurosarcoidosis from MS can be troublesome, as definitive diagnosis requires invasive brain or meningeal biopsy demonstrating the characteristic granulomas. This study characterises a cohort of neurosarcoidosis patients with a focus on CSF analysis and whether this could help distinguish these two conditions.
This study enrolled 85 patients with a diagnosis of neurosarcoidosis based on stringent diagnostic criteria. The CSF protein, white cell count, and angiotensin converting enzyme levels were measured. The CSF and serum oligoclonal IgG patterns were compared. 80 patients had a probable or definitive diagnosis of neurosarcoidosis. The most frequent findings on MRI were leptomeningeal enhancement (35%) and white matter and spinal cord involvement (30% and 23%). CSF analysis frequently showed lymphocytosis (63%) and elevated protein (62%), but oligoclonal bands were rarely seen (3% in the CSF alone, and 11% matched in the CSF and serum). There was a lack of correlation between leptomeningeal involvement on imaging and CSF OCB. Serum ACE levels were elevated in 51% of patients, but in only 14% of those with isolated neurosarcoidosis. Large elevations in CSF protein, WCC and ACE occur in neurosarcoidosis, but are rare in MS. The diagnostic use of these tests is limited, however, since minimal changes may occur in both conditions. In contrast, intrathecal synthesis of oligoclonal IgG is a powerful discriminator as it is rare in neurosarcoidosis whilst occurring in 95–98% cases of MS. We suggest caution in making a diagnosis of neurosarcoidosis when intrathecal oligoclonal IgG synthesis is found.

P111

Intra Cholecystic Papillary Adenoma Neoplasm of the Gallbladder (ICPN): Report of Two Cases

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Introduction: The definition of ICPN was first given by Adsay et al (Am J Surg Pathol. 2012 ;36(9):1279-301) for polypoid neoplasms arise in the gallbladder (GB). We report two cases of ICPNs.
Case reports:
Case 1: A 65-year old man showed a cystic/solid lesion in the gall bladder (GB) fundus. Microscopy of the GB wall showed chronic inflammation. The fundal lesion revealed an adenomymatosis containing foci of neoplasm comprising papillae lined by tall columnar cells with focal intracellular mucin; features mimicking the pancreatic counterpart lesion called papillary mucinous neoplasm (IPMN). Immunohistochemistry of the ICPN was positive for CK7, MUC1 (EMA) and MUC5AC. There was a focal positive staining with CDX2, CK20, MUC2, MUC4 and CEA. The Ki67 proliferative index was 1%.
Case 2: A 78-year old female with empyema of GB. At microscopy the GB showed severe acute inflammation. The mucosa of the GB showed papillary structures intermixed with solid and cribriform areas where cells had large nuclei. Immunohistochemistry of the papillary neoplasm was positive for CK7, MUC1, MUC4, CEA and MUC5AC. There was a focal positive staining with CDX2, CK20, and MUC2. The Ki67 index was 50%. The predominant cell lineage in these two cases was of biliary and gastric foveolar-type.
Conclusion: ICPN is an intramucosal GB mass showing dysplastic cells distinct from the neighbouring mucosa. ICPNs are pre-invasive neoplastic lesions. Dysplasia and carcinoma have been reported in adenomyomatosis but to the best of our knowledge, no cases of ICPN have been reported in adenomyomatosis of GB.

P112

IgG4 Expression Correlates with Poor Prognosis in Pancreatic Adenocarcinoma

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Purpose of the study: IgG4, a member of the immunoglobulin family, has been implicated in the pathobiology of inflammatory disease (including autoimmune pancreatitis) and cancers. Its role in pancreatic cancers however remains to be elucidated. This study aimed to examine the role of IgG4 in pancreatic cancer, in both tumoural and inflammatory components.
Methods: The expression of IgG4 in pancreatic adenocarcinomas was studied by immuno-histochemical analysis of a tissue microarray (n=142) and analysed for clinicopathological correlations. The tumour inflammatory infiltrate, IgG4 positive lymphocytes (LCA stained) and plasma cells (CD138 stained) were investigated to understand their role in pancreatic malignancies.
Summary of results: Pancreatic tumour cells showed three positive patterns of staining: 47 (33%) golgi, 26 (18.3%) nuclear and 100 (70.4%) cytoplasmic. Significant positive correlations for tumoural IgG4 were observed with pancreatic cancer stage, grade, perineural invasion and recurrence ($p < 0.05$). IgG4 positive lymphocytic infiltrate was seen in 98 (69%) cases and presence at the tumour edge correlated with perineural invasion ($p= 0.03$). 80 (56%) cases exhibited an IgG4 positive plasma cell infiltrate which correlated with T stage ($p=0.02$) and vascular invasion ($p= 0.04$). Tumours with both strong golgi expression in tumour cells and high IgG4 positive lymphocytic infiltrate showed positive correlation with recurrence ($p=0.01$).
Conclusions: Overall, this study shows that IgG4, expressed in pancreatic tumour cells and its associated lymphoplasmacytic infiltrate, has significant correlations with poor prognostic features. Further exploration of the immunological milieu of pancreatic tumours will validate the functional significance of IgG4 associated immune networks in pancreatic malignancies.

P113

Expression of Secretory Leukocyte Protease Inhibitor (SLPI) Detected by Immunohistochemistry in Hepatocellular and Cholangiocellular Tumours

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Introduction: Expression of secretory leukocyte protease inhibitor (SLPI) has been observed in hepatic macrophages and the biliary epithelium of livers affected by massive hepatic necrosis. In extrahepatic tumours SLPI has been considered to have a role as both promoter and inhibitor of cancer in different contexts. Here, we perform a preliminary investigation into the possible role of SLPI in the assessment of benign and malignant liver tumours.
Methods: We studied 10 cases of focal nodular hyperplasia (FNH), 15 large regenerative nodules (LRN), 11 dysplastic nodules (DN), 30 hepatocellular carcinomas (HCC), 17 combined hepatocellular cholangiocarcinomas (c-HCC-ChC), and 30 cholangiocarcinomas (ChC). The ChC cases included 10 intrahepatic, 13 hilar and 7 distal ChC. SLPI expression in both tumour tissue and background liver tissue was qualitatively assessed and characterised as negative, weak, moderate or strong.
Results: 98% of FNH, LRN, DN and untreated HCC showed no (55%) or weak (43%) expression of SLPI. Moderate to strong SLPI expression was observed in the cholangiocellular component of 47% of the c-HCC-ChC and in 73% of the ChC. Strong expression was a characteristic feature of the invasive component of all 13 hilar ChC, but was more variable in intrahepatic and distal ChC with strong expression in 50% and 57% of cases respectively. We observed that SLPI expression was prominent in the neoplastic epithelial component at interfaces with extralesional stroma and intralesional vascularised septa.
Conclusion: The preliminary results of this study indicate a role of SLPI in liver carcinogenesis, particularly in relation to cholangiocellular differentiation, and suggest that SLPI may have a role in the interactions between neoplastic epithelial cells and the adjacent stroma, possibly in terms of stromal invasion. Further studies are necessary to confirm these findings and investigate the role of SLPI in epithelial-stromal interactions in more detail.

P114

An Unusual Solid Pancreatic Tail Lesion: Case Report

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Purpose of the study: To highlight the importance of cytological evaluation in diagnosis and treatment of unusual pancreatic masses.
Methods: A 67 year old lady with a diagnosis of breast cancer underwent staging CT which revealed a small hypervascular pancreatic tail lesion. FNA of the lesion showed a polymorphous lymphoid population, along with platelet aggregates, small capillaries and some spindle cells. No tingible body macrophages or germinal centres were seen. Immunostaining showed the lymphoid cells to be positive for CD45. CD8 was also positive highlighting cytotoxic T lymphocytes and splenic endothelial cells. Cam 5.2, Chromogranin and Synaptophysin were negative excluding a neuroendocrine tumour. Results: The differential diagnosis lay between an intrapancreatic lymph node and intrapancreatic spleen (splenule), but the final diagnosis was a splenule, for which the patient required no further treatment.
Conclusions: Splenules are rare, benign lesions with excellent prognoses, which do not require surgical intervention. It is very important to be able to identify and definitively diagnose them cytologically with the use of appropriate immunostains in order to prevent unnecessary radiological or surgical intervention.

P115

Lymph Node Yield, Nodal Status, and Excision Margin Status in Pancreaticoduodenectomy Specimens: A Seven-Year Audit Assessing the Impact of Preoperative Neo-Adjuvant Therapy

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Purpose: Pathological examination of tumour margin and lymph node dissection following pancreaticoduodenectomy (PD) for resectable pancreatic ductal adenocarcinoma (PDAC) is crucial in tumour staging and disease prognosis. The Irish National Cancer Control Program (NCCP) has set a minimum lymph node yield (LNY) of 10 as a key performance indicator (KPI) in PD specimens. In this seven year audit, we assessed adherence to the KPI and the effect of neo-adjuvant therapies (NAT) on LNY, nodal status and margin status in PD specimens of PDAC.
Methods: All pancreatic specimens at St. Vincent's University Hospital (SVUH) from January 2012 to December 2018 were retrieved via a SNOMED search of the laboratory information system (n=735). Margin status, nodal status, LNY, and pathological treatment response (PTR) data were obtained from pathology reports. Definition of negative margin status (R0) was based on the Royal College of Pathologists pancreatic cancer dataset. PTR was assessed using the College of American Pathologists (CAP) tumour regression grade (TRG).
Results: 183 PD specimens contained PDAC and NAT was administered to 50 patients. The mean LNY for NAT patients (14.02) was lower than for non-NAT patients (17.99). Overall, 160/183 (87%) of specimens achieved ≥10 LNY. Of the 133 non-NAT cases, 122 (92%) achieved a ≥10 LNY and of the 50 NAT cases, 38 (76%) achieved the same minimal LNY. R0 was achieved in 39/50 (78%) of NAT patients, and in 78/133 (59%) of non-NAT cases. Negative nodal status (N0) was achieved in 24/50 (48%) of NAT-patients and 36/133 (27%) of non-NAT cases. A favourable CAP TRG (0 or 1) was observed in 14/50 (28%) cases.
Conclusion: NAT has a negative impact on both LNY and adherence to the KPI. However, NAT has shown a positive impact on the R0 status and better outcomes in nodal status. Overall, adequate adherence to the KPI in PD specimens containing PDAC was met. Despite these findings, the majority of NAT cases did not demonstrate a favourable PTR.

P116

Slides of Many Colours – Use of Tinctorial and Immunohistochemical Stains in Liver Biopsy Reporting: A survey of the UK Liver Pathology Group and BDIAP 2018 Joint Liver Meeting Delegates

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Purpose of the study: To inform the RCPath Tissue Pathways for liver biopsies, we wanted to know current UK practice regarding the use of routine liver special stains and immunohistochemistry for medical and tumour biopsies.
Method: We compiled a SurveyMonkey questionnaire sent to delegates at the UKLPG/BDIAP Liver Meeting November 2018. This asked which tinctorial stains are used routinely for medical liver biopsies and which immunostains for hepatocellular lesions are available in their hospital.
Results: We received responses from 36 histology departments in the UK, with an additional 6 from Belgium and Holland. For medical liver biopsies in the UK the median routine stained sections was 9 (6–12). All but one include 2–4 H&E levels, along with an average of 6 (4–9) tinctorial stains. All centres routinely performed PASD, reticulin and Perls, and all but one do Shikata and/or Victoria Blue. All do one or more collagen stains, most often van Gieson (18/36). Nine routinely include an immunostain, usually K7. In Belgium and Holland, the number of stains is similar but choice varied – all use rhodanine and/or K7 instead of Shikata or Victoria Blue, and van Gieson is not used. Immunohistochemistry: 20/42 departments receive resections for primary liver tumours. For antibodies used in the diagnosis of well differentiated hepatocellular lesions 15 have glutamine synthetase and Glypican 3 (with 13 having both). Other antibodies used for adenoma diagnosis are available in 5/17 UK centres and all 3 in Belgium/Holland. For 22 departments not receiving resections, HepPar1 and/or AFP are available in 21; glutamine synthetase and/or Glypican 3 are available in 2, sent away in 12, and never used in 6.
Conclusions: The study shows that routine use of tinctorial stains is ubiquitous among delegates at the meeting; liver biopsies require a high laboratory technical input. Immunostain use varies reflecting developments in diagnostic practice, and experience should be shared to ensure a common approach to diagnosis. Departments which are not liver surgical centres should have access to IHC or refer biopsies where the differential includes primary hepatocellular neoplasia.

P117

Intrasplenic Epithelioid Malignant Mesothelioma: A Case Report

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Malignant mesothelioma (MM) most commonly arises from mesothelial cells lining the pleura, often related to asbestos exposure. Less commonly it involves the peritoneum and rarely pericardium or tunica vaginalis of testis or ovary. Localised, intraparenchymal MM in abdominal organs is very rare with few cases reported in liver, pancreas and spleen, likely having arisen from mesothelium of the capsule of the organs. A 75 year old lady presented with general malaise and a history of iron deficiency anaemia, hypertension and rheumatoid arthritis. Imaging revealed a 108mm splenic mass and a likely vascular liver mass. Tumour markers CA15-3, CA125, AFP and HCG were normal. Core biopsies of spleen lesion showed a malignant epithelioid tumour with few large bizarre nuclei, frequent mitoses and necrosis. A preliminary diagnosis of metastatic carcinoma of unknown primary origin was made based on positivity for cytokeratins AE1/3, 5 and 7. Positivity for Vimentin and WT1 was noted. Further tests revealed the tumour to be positive for Calretinin, D2-40, thrombomodulin and 34BetaE12 leading to a diagnosis of MM in the spleen. There was no known asbestos exposure. The patient underwent splenectomy for local symptom control. The resected spleen weighed 806grams and was 80-85% involved by tumour. Resection histology including targeted immunohistochemistry confirmed epithelioid MM. CT scan in Sep 2018 showed recurrence in splenic bed with peritoneal involvement. She unfortunately died 2.5 years after her diagnosis. Localised intra and perisplenic MM is very rare and may represent transformation of splenic mesothelial inclusion cysts or splenic capsular mesothelium. To the best of our knowledge this is only the second reported case of intrasplenic MM. Awareness of rare intrasplenic MM will facilitate correct diagnosis when posed with epithelioid tumours of unknown origin at pathological assessment of splenic tissue samples.

P118

The Tale of Two Livers

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Introduction: The liver has a limited number of morphological changes that occurs secondary to pathological insults. In this poster, we present two classic but rare cases of liver biopsies.
Case Presentation 1: A 64 year old man of Asian origin, with a history of treated Hepatitis C, type 2 diabetes mellitus and seronegative arthropathy, was referred to Royal Surrey County Hospital with an isolated conjugated hyperbilirubinaemia. He developed a chest infection and his Liver Function Tests showed an elevated ALT of 90. Biopsies were taken which showed steatosis, focal ballooning and prominent pericanalicular pigmentation,which was negative for Orcein and Perls staining. Features were suggestive of a hereditary cause of conjugated hyperbilirubinaemia. Urine coprophyrin studies are awaiting.
Case presentation 2: A 72 year old Caucasian retired lawyer presented to Royal Surrey County Hospital with weight loss and general malaise. CT thorax, abdomen and pelvis showed a 3cm hepatic mass in segment 8, which was radiologically consistent with a hepatocellular carcinoma. On further enquiry, he revealed a 30 year history of asthma. Liver biopsy revealed PAS positive hyaline alpha-1-antitrypsin globules, mild-moderate chronic periportal inflammation and stage 3 fibrosis. A diagnosis of anti-1-anti-trypsin deficiency was made and upon review by the respiratory physicians, a pulmonary manifestation was also confirmed. PiZ genotype was confirmed on serology.
Discussion: Hereditary causes of conjugated hyperbilirubinaemia include Dubin-Johnson Syndrome and Rotor Syndrome, which cannot be distinguished morphologically, and require urine coproporphyins for definitive diagnosis. The diseases discussed are rare but have classical microscopic appearances that need to be correlated with the biochemical picture. Awareness of these entities amongst general histopathologists is instrumental in guiding the clinician towards the diagnosis.

P119

Intra-Tumoural Expression of CD4 is Predictive of Recurrence in Non-Small Cell Lung Cancer Patients Managed with Surgery and Adjuvant Chemotherapy

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Purpose of the study: Non-small cell lung cancer (NSCLC) represents the majority of newly diagnosed lung cancers. Evading immune destruction has become one of the hallmarks of cancer and NSCLC has been at the forefront of therapeutic advances with immunotherapy. The immune system is implicated in the efficacy of several conventional chemotherapeutics. We wished to explore whether the adaptive immune response as reflected by immune biomarker expression could predict recurrence in patients with non-small cell lung cancer, and if so, whether the relationship was modulated by management with adjuvant chemotherapy.
Methods: Surgical resections of NSCLC from 220 patients were assessed in tissue microarray format for six immune biomarkers CD3, CD4, CD45RO, CD8, FOXP3 and ICOS. All biomarkers were assessed by digital image analysis using open source software (QuPath). Biomarker densities were dichotomised using ROC curves for survival analysis. All statistical analysis was performed using R.
Summary of results: No difference in immune biomarker expression was observed between patients who were managed with surgery and chemotherapy compared to surgery alone ($p>0.05$). Low CD4 expression was associated with disease recurrence ($p=0.05$). When stratified by treatment, low expression of CD4 in was found to be predictive of risk of recurrence in patients treated with adjuvant chemotherapy ($p<0.001$); patients with low intra-tumoural CD4 expression prior to treatment with adjuvant Cisplatin/Vinorelbine were more likely to recur than those with high CD4 expression.
Conclusions: Preliminary findings suggest that CD4 expression is predictive of recurrence in NSCLC patients managed with surgery and adjuvant chemotherapy using Cisplatin/Vinorelbine, but not surgery alone. Intra-tumoural CD4 expression could potentially be used as a biomarker to predict patients at high-risk of relapse when treated with adjuvant Cisplatin/Vinorelbine. Further work is required to validate these findings.

P120

On-Demand EGFR Mutation Testing in Lung Cancer: Results in Three Hours

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Lung carcinoma is the most common cancer in the UK (excluding non-melanoma skin cancer) and the survival for these patients remains low. The majority present with non-small cell lung cancer (NSCLC) and around 16% of tumours have tyrosine kinase inhibitor sensitising mutations in the EGFR gene. These patients generally are often very sick and management decisions need to be made urgently. Pathologists are often under pressure to get results out in time for MDTs or clinics but not all centres have access to molecular diagnostics with fast turnaround times. Consequently, reflex testing of all patients is often employed, and this potentially wastes time and money. The Idylla™ EGFR Mutation Test offers rapid, on-demand results within three hours – from pathologist request to reportable result. The Idylla platform has shown promise in other mutation targets, including BRAF, KRAS and NRAS, and the potential for rapid results in lung cancer is attractive. This study aimed to assess the concordance of Idylla™ EGFR Mutation Test results with current standard tests. Forty formalin-fixed, paraffin-embedded NSCLC tumour cases (20 EGFR mutant and EGFR 20 wild type by standard testing) were retrospectively analysed by the Idylla™ EGFR Mutation Test (CE-IVD) and compared with PCR and NGS methodologies. The overall concordance between Idylla™ and standard testing was 92.5% (95% CI 80.14% to 97.42%) and the specificity of Idylla™ was 100% (95% CI 83.89% to 100%). The sensitivity was affected by loss of tumour content in tissue blocks in a small number of NGS cases; however, comparing Idylla™ with PCR alone, there was 100% concordance (95% CI 89.85% to 100%). The Idylla™ EGFR Mutation Test shows comparative accuracy to routine PCR testing for the most common EGFR mutations in NSCLC. The Idylla™ also offers very significantly reduced turn-around times compared with existing modalities and therefore could save money with eliminating the need for reflex testing.

P121

Storage Cardiomyopathy: A Case of Sudden Death Due to Danon's Cardiomyopathy with Inflammation

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Purpose of the study: Cardiac disease is important in lysosomal glycogen storage diseases (Pompe and Danon disease), mucopolysaccharidoses and glycosphingolipidoses (Anderson-Fabry disease). The phenotype can vary to include hypertrophic and dilated cardiomyopathy, coronary artery disease and valvular disease. Danon's disease is a lysosomal storage disorder caused by an X-linked germline mutation in the LAMP2 gene resulting in cardioskeletal myopathy. Case of a 20-year-old female, previously diagnosed with Danon's disease with positive LAMP2 gene mutation and dilated cardiomyopathy who suffered a sudden cardiac death. We present the results of the heart examination.
Methods: The heart was examined using specific protocol with histology, selected immunohistochemistry and special stains.
Summary of results: There was cardiomegaly with biventricular dilatation and hypertrophy. On histology, hypertrophy and vacuolar degeneration of myocytes with patchy replacement fibrosis was identified. PAS staining failed to identify glycogen-containing lysosomes. Large epicardial areas contained a prominent CD3+ lymphocytic inflammation with myocyte necrosis.
Conclusions: Myocardial hypertrophy associated with vacuolar degeneration of myocytes and patchy fibrosis are hallmarks of Danon's cardiomyopathy. The absence of demonstrable intracytoplasmic glycogen should not discourage the diagnosis in the context of LAMP2 mutation. Genetics is important in the study of cardiomyopathies and LAMP2 mutations have an especially poor prognosis. The presence of an infiltrate of CD3+ lymphocytes has not been reported in Danon's cardiomyopathy before, and provides a potential explanation for her sudden death.

P122

Chaperone-Mediated Autophagy Markers LAMP2A and HSC70 are Independent Adverse Prognostic Markers in Primary Resected Squamous Cell Carcinomas of the Lung

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Purpose of the study: LAMP2A and HSC70 are crucial players in chaperone-associated autophagy (CMA), a process of specific, targeted, lysosome-dependent degradation of proteins. CMA is crucial to maintain cell homeostasis and is frequently upregulated in cancer. Blockage of CMA may be therapeutically exploited. We aimed to evaluate the expression patterns and any prognostic significance of LAMP2A and HSC70 in pulmonary squamous cell carcinomas (pSQCC).
Methods: LAMP2A and HSC70 were analysed by immunohistochemistry in a consecutive cohort of 336 primary resected pulmonary squamous cell carcinomas using tissue microarrays (4 TMA cores from 2 different TMA blocks). Expression levels were determined by an immunoreactivity score (IRS) generated from the staining intensity and the percentage of positive tumour cells.
Summary of results: There was no significant intratumoural staining heterogeneity across the TMA cores. Moreover, no significant correlation between the two markers was seen. There was no association of marker expression with pathological parameters (pT category, pN category, TNM staging, grading). However, high LAMP2A and high HSC70 expression levels, defined as IRS levels above the 4th quartile, were associated with worse outcome, including overall survival (p=0.012 and p=0.001) and disease free survival (p=0.049 and p=0.036). Both markers were also independent adverse prognostic factors in multivariate analysis for overall survival (LAMP2A: HR=1.772; 95%CI 0.121-2.595; p=0.003; HSC70: HR=1.955; 95%CI 1.351-2.830; p<0.001) and disease free survival (LAMP2A: HR=1.528; 95%CI 1.066-2.191; p=0.021; HSC70: HR=1.482; 95%CI 1.047-2.098; p=0.027).
Conclusions: The CMA markers LAMP2A and HSC70 are variably expressed in pSQCC, and could be evaluated as predictive biomarkers for CMA-inhibiting therapy. High expression in untreated pSQCC presents an independent adverse prognostic factor.

P123

Evaluation of the Diagnostic Utility of BAP1 Immunohistochemistry in Malignant Mesothelioma

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Diagnosis of malignant mesothelioma (MM) can be problematic as it can be difficult histologically to distinguish benign reactive mesothelial hyperplasia from malignant mesothelial proliferations. Immunohistochemistry (IHC) is used to help distinguish between benign and malignant mesothelial proliferations, however, sometimes the results are equivocal and samples are sent away to a tertiary centre for further tests including p16 deletion molecular test which increases costs and turnaround times. Loss of BRCA1 associated protein 1 (BAP1), due to mutation, has been implicated in up to 90% of cases of MM and studies suggest IHC to be a useful tool to identify loss of BAP1. IHC for BAP1 was performed in 33 cases of confirmed MM and 33 cases of non-mesothelioma pleural tissue using two tissue microarrays. Loss of nuclear staining, indicating a BAP1 mutation, was seen in 1/33 (3%) of the benign mesothelial samples and 12/23 (52%) of the malignant mesothelial samples. These results suggest the addition of BAP1 to the IHC antibody panel would support and improve the diagnosis of MM in house and reduce the number of cases sent to tertiary centres for molecular testing.

P124

Pulmonary Glomus Tumour

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A 33 year old woman presented with a chest infection to her GP. An abnormal chest x-ray prompted a chest CT which revealed a 20mm peripherally-located lesion in the left lower lobe. Following VATS wedge resection, frozen section and histology, a diagnosis of a low grade mesenchymal neoplasm with muscle differentiation favouring glomus tumour was made.
Introduction: Glomus tumours are uncommon tumours, predominately found in the dermis or subcutis of the upper and lower limbs. Although glomus tumours have been reported throughout the body, pulmonary glomus tumours are considered to be rare.
Case: The patient was referred to our Trust for a second opinion. MDT discussion led to VATS wedge resection and frozen section +/– proceed to lobectomy. It was impossible to provide a specific diagnosis on the frozen section and although tumour was confirmed, formal typing was deferred to paraffin sections. Completion lobectomy was not performed.
Histology: Macroscopically the lung wedge contained a 19mm firm tumour nodule with a greyish cut-surface. Microscopically the tumour was circumscribed and composed of cohesive rounded cells in nests and lobules. The cells were arranged around thin-walled sinusoidal vessels; had grooved and reniform nuclei, with occasional nuclear inclusions; moderate amounts of eosinophilic cytoplasm, which also showed clearing and micro-vacuolation; and prominent cell membranes. There was limited nuclear pleomorphism, no necrosis and no mitotic figures in 50hpf. Several rounds of immunohistochemistry were undertaken to identify the tumour, which showed positive staining with SMA, Caldesmon and Vimentin. All epithelial markers, neuroendocrine markers and vascular markers were negative, as was HMB45.
Conclusion: This is an example of a rare pulmonary tumour which posed an interesting challenge at frozen section and on histology due to the rarity of the lesion and its wide differential diagnosis.

P125

IgG/IgG4 Staining Suggests Some Pulmonary Hyalinising Granulomas are Associated with IgG4-Related Sclerosing Disease

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Pulmonary hyalinising granuloma (PHG) is a rare condition of unknown aetiology, comprising circumscribed single or multiple nodules of thick hyalinised collagen bundles with chronic inflammation. PHG has been associated with immune disorders with one case report suggesting association with IgG4-related sclerosing disease. We therefore reviewed a cohort of PHGs, undertaking IgG and IgG4 immunohistochemistry, in order to assess whether there were features of IgG4-related disease (3 high-power-fields (HPF) were scored for absolute number of IgG4+ cells and IgG4+/IgG+ ratio). 8 cases of PHG (7 surgical and 1 core biopsy; 6 solitary and 2 multiples nodules) in 5 females and 3 males with an average age at diagnosis of 52 years (range 34-70) were assessed. All showed characteristic thick collagen bundles and a lymphoplasmacytic infiltrate. The highest determination of absolute numbers of IgG4-positive plasma cells/HPF in each case was: 37; 40; 49; 34; 155; 22; 83; 89 respectively (range 13-155). The average in IgG4/IgG ratio was 41% (64, 22, 46, 28, 49, 27, 44, 49 respectively). Thus, 3/8 (37.5%) of cases showed >20 IgG4-positive plasma cells and IgG4/IgG ratio greater than 40%, fulfilling criteria for IgG4-related sclerosing disease, this being 5/8 (62.4%) if using an IgG4/IgG ratio >40% only. Furthermore, these 5 cases had additional features adjacent to the PHG nodule comprising moderate to severe lymphoplasmacytic infiltrate with follicular hyperplasia. One case with high IgG4 scores showed coexistent non-necrotizing granulomas consistent with sarcoidosis. 3 cases had concomitant diseases as: rheumatoid arthritis, retroperitoneal fibrosis, pelvic sarcoma. However, only the first two showed >40% IgG4+/ IgG+ plasma cells but none greater than 50 IgG4 positive cells/HPF. These data suggest coexistent IgG4-related sclerosing disease in some cases of PHG. Staining for IgG4 and IgG, as well as the serum IgG4 levels, should be considered in all cases.

P126

Impact of Provision of In-House *EGFR* Mutation Testing for NSCLC on Report Turnaround Times

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Purpose of the study: Our audit aimed to evaluate the impact of the introduction of internal *EGFR* mutation testing on report turnaround times, assess compliance with relevant international standards, and to compare local rates of *EGFR* mutations in non-small cell lung cancer (NSCLC) with national and international rates. Audit targets were set at 90% of *EGFR* tests to be reported within 7 days of request.
Methods: Data was collected retrospectively, with retrieval of records from the pathology electronic request system for all external *EGFR* test requests from July 2016 to December 2017, and for all internal *EGFR* requests from its launch in July 2017 to September 2018. Reporting dates and mutational status were collected from corresponding pathology reports. Timestamps on records were used to calculate turnaround times in working days. Data was analysed using chi-squared testing via Excel.
Summary of results: Internal *EGFR* testing resulted in a mean turnaround time of 5.8 days (±4, n= 179), as compared to 10.7 (±3.6, n=101) for externally performed *EGFR* tests. 83.2% of results for cases tested internally were reported within 7 working days, as opposed to only 5.0% of cases tested externally (p=<0.00001). 96.1% of internal *EGFR* test results versus 58.4% of external *EGFR* test results were available within 10 working days of request (p=<0.00001). The overall rate of NSCLCs harbouring *EGFR* mutations was 17.5%. Of those mutations, L858R was the most common (35.8%), followed by exon 19 deletions (24.5%). *EGFR* test failure rates were 4%.
Conclusions: In-house *EGFR* mutation testing for NSCLC samples has led to significantly reduced turnaround times compared to testing via external laboratories, but areas for improvement of service delivery have been highlighted. Our action plan includes a formal rota to ensure same day reporting and optimisation of our requesting program to flag overdue cases. Local mutation rates are in line with national and European reported rates.

P127

A Filamentous Mimic of Lung Cancer

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Introduction: Actinomyces is a gram positive anaerobic filamentous bacterium, present in soil. There are 47 subtypes, 25 of which are found in humans. actinomyces israelii is a normal commensal of oropharynx, gastrointestinal tract, and urogenital tract.
Case: a 67 year old man with a history of bronchiectasis, presented with worsening symptoms. Base line blood tests were normal. A CT chest showed right lower lobe bronchiectasis, consolidation and a calcified right hilar lymph node. Bronchoscopy showed a right lower lobe endobronchial tumour and subsequent biopsy showed endobronchial actinomyces, associated with acute inflammation.
Discussion: Actinomycosis is usually non-pathogenic unless there is mucosal damage. Thoracic actinomycosis accounts for 15–20% of all the cases. Endobronchial actinomycosis is exceedingly rare. Since 1882, around 200 cases have been reported in the published literature. There are various clinical associations, such as broncholithiasis and intrapulmonary disease. Interestingly, it can also mimic TB and primary lung carcinoma resulting in misdiagnosis, which can occur in up to 25% of cases. Investigative radiological and endoscopic findings are usually non-specific but histology plays a vital role in an area of diagnostic difficulty due to its characteristic microscopic appearance and pattern of staining. Treatment includes prolonged course of beta lactam antibiotics or occasionally surgery. Despite its protracted course, the therapy is highly effective with a good prognosis and clinical outcome.
Conclusion: Our patient is currently on intravenous benzyl penicillin with symptomatic improvement. This case is a useful reminder to consider rare causes of endobronchial lesion.

P128

An Unusual Dual Diagnosis of Rosai-Dorfman Disease Presenting Concurrently with IgG4 Related Disease

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Rosai-Dorfman disease (RDD) is a self-limiting, rare disease also known as sinus histiocytosis with massive lymphadenopathy. Currently classified under histiocytic disorders, the diagnostic histological hallmark of this disease is infiltration by large histiocytes (that express S100 and show emperipolesis) with plasmacytosis. The precise aetiology is not known. Recently, there have been some case reports that describe association of IgG4 related disease (IgG4RD) with RDD. We describe an unusual case of RDD presenting as a right atrial mass in a 52 year-old woman, which was excised by the cardiothoracic surgical team. Histology from the mass showed features typical of RDD – S100+/CD163+ histiocytes associated with emperipolesis with accompanying lymphocytes and plasma cells – along with an additional component: the presence of prominent fibrosis. The fibrosis replaced the myocardium and extended into the pericardium and endocardium. There was no necrosis. Immunohistochemical analysis showed one third of IgG-positive plasma cells also expressed IgG4. Thus, the overall morphology and immunophenotype was in keeping with a dual diagnosis of both RDD and IgG4RD. IgG4RD is typically characterised by a dense lymphoplasmacytic infiltrate rich in IgG4-positive plasma cells associated with 'storiform' fibrosis. Cases of RDD demonstrating a local increase in IgG4 positive plasma cells are described in the literature, however, cardiac involvement by RDD is rare and the course is variable. Consensus guidelines on investigation and management of RDD have been published recently (Blood 2018: blood-2018-03-839753). The precise aetiopathogenetic link between RDD and IgG4RD is unclear. Due to the rarity of this disease, no case series exist till date. Further collaborative work is needed to investigate the co-existence of these diseases further.

P129

Thymic Pathology: 20-Years' Experience at a Regional Cardiothoracic Centre

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Purpose of the study: Thymic tumours account for <1% of all neoplasms. These tumours have complex biology along with a relatively poorly characterised aetio-pathogenesis, making the study of these lesions important. This comprehensive review looks at all histologically confirmed thymic tumours reported at a regional cardiothoracic centre over a 20-year period.
Methods: All confirmed thymic pathology reports issued between 1997 and 2017 were retrospectively reviewed. Data was collected on the type of specimen received, the histological diagnosis and the concordance between biopsy and resection specimens.
Summary of results: 391 [100%] cases of histologically confirmed thymic pathology (resection specimens [71.9%], core needle and open biopsies [27.6%]) were identified during this period. Of these, 9.2% of cases were external referrals. 74.4% of the cases were neoplastic of which 78.0% were thymic epithelial neoplasms (thymoma, thymic carcinoma and neuroendocrine tumours). A resection specimen was received following a biopsy in 35.1% of cases. There was full concordance between biopsy and resection diagnoses in non-thymoma cases. In cases diagnosed as thymoma on the biopsy specimen, 50.0% showed full subtype concordance and 18.8% showed either a major or minor discordance in subtype correlation. In 31.3% of cases, correlation was not feasible.
Conclusion: This retrospective review highlights the wide spectrum of thymic pathology. The presentation emphasises importance of a good clinico-pathological correlation and an appreciation of the morphology of lesions at this site to appropriately triage often limited diagnostic biopsy material. Primary thymic neoplasms should always be considered within the differential diagnosis of a mediastinal lesion, including cases showing squamous or adenocarcinoma morphology.

P130

Primary Pleural Epithelioid Haemangioendothelioma: A Case Report of a Rare Pleural Tumour

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Introduction: Epithelioid haemangioendothelioma is a rare malignant vascular neoplasm which can arise in soft tissue, bone, liver and lung but is extremely uncommon as a primary pleural tumour with less than 35 cases reported within the literature.
Case presentation: A 60 year old man presented with chest and shoulder pain and was found to have a unilateral pleural effusion, thought to be trauma related. He was otherwise fit and well, had never smoked and had previously worked in the armed forces. A follow up CT showed a hydropneumothorax with pleural thickening. A video-assisted procedure (VATS) was performed to drain the effusion and obtain a pleural biopsy. The biopsy showed thickened strips of pleura, diffusely infiltrated by cords and clusters of epithelioid and spindled cells. Focally these showed attempts at vasoformation with intracytoplasmic lumina, some of which were filled with red blood cells. Immunohistochemistry showed positive staining for Thrombomodulin, D2-40, CD31 and focally for CD34 and Pancytokeratin. They were negative for CK5, Calretinin and WT-1. RT-PCR showed the presence of a WWTR-CAMTA1 fusion, confirming the diagnosis of a primary pleural epithelioid haemangioendothelioma.
Discussion: Primary pleural epithelioid haemangioendothelioma is a rare, aggressive and chemotherapy insensitive tumour with a poor prognosis. It often presents with non-specific symptoms such as dyspnoea, chest pain and cough. Imaging often shows a pleural effusion with pleural thickening. It is important to be aware of this entity to avoid misdiagnosis as the radiological and histological appearances can mimic mesothelioma. Molecular analysis is a useful adjunct to diagnosing these tumours through demonstration of WWTR-CAMTA1 or YAP-TFE3 gene fusions.

P131

Exploration of the Benefits of a Regional Digital Pathology Network for Referrals to a Tertiary Centre Specialist Melanoma Multidisciplinary Team Meeting

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Purpose of the study: To assess the existing pathway for referral of cases to a tertiary centre specialist melanoma multidisciplinary team (MDT) meeting for review of histology, evaluate timings of case movement and compare time differences between internal and external cases.
Methods: In order to assess the potential impact to a department as a whole and identify the cancer pathway, which could derive the most benefit from digitisation, volumes of outbound case referrals were obtained from four cellular pathology departments within the region, which referred cases into a central tertiary centre. Timings of events in the patient diagnostic and management pathway were obtained from electronic patient records and the cellular pathology department laboratory information systems and used to calculate the overall pathway lengths for internal and external patients. Time differences for handovers between different processes within a hospital trust and for interprovider transfers were calculated.
Summary of results: Cases from external sites referred to the tertiary centre specialist melanoma MDT meeting passed through a number of additional steps between the initial histological diagnosis and the MDT review resulting in a lengthened pathway. For example, it took on average 6.6 days between the referral being recorded and the slides being received for review and 9.5 days between recording of the referral and the MDT date. In comparison, for internal cases the interval between recording of the referral and the MDT date was 5.5 days.
Conclusions: The use of whole slide imaging at the point of the initial diagnosis and development of a digital pathology network can reduce the time required for interprovider transfers for cancer cases. As preparation for an MDT often requires a fixed cut-off date for receipt of external cases, even a 6 day reduction in number of days in the pathway prior to the MDT (the delay from slide transportation) can potentially have marked impact for patients.

P132

Post-Radiotherapy Cutaneous Mastocytosis: Once Seen, Would You Forget it?

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Cutaneous mastocytosis presenting during adulthood appears to endure for the duration of life. A large series has proposed that in all, and demonstrated that in most, of these cases patients have systemic mastocytosis with cutaneous involvement. The difference between prognosis in cutaneous mastocytosis and systemic mastocytosis renders the distinction in diagnosis and the choice of intervention important. We present the case of a 50-year-old female patient with a localised petechial rash in the left chest wall skin. The patient has a past history of breast cancer and had undergone left mastectomy and chest wall radiotherapy 15 months previously. A punch biopsy was performed. Microscopically the epidermis showed increased basal pigmentation, and an interstitial and perivascular loosely distributed infiltrate of mast cells and small numbers of eosinophils. The mast cells stain positively with C-kit. The appearance is of cutaneous mastocytosis. The patient has an elevated serum tryptase of 21.7ng/ml, suggesting systemic mastocytosis, although she has no systemic symptoms. Tissue was sent for KIT mutation testing, but unfortunately was incomplete as the percentage of nuclei estimated within the area of DNA extraction was below the limit of detection for Sanger sequencing. As a result a bone marrow biopsy is planned. A literature search has revealed only six cases of mastocytosis within irradiated areas, all of which occurred in the context of adjuvant radiotherapy for breast cancer. One of these cases is described as systemic mastocytosis. We explore clinical, histological and molecular genetic findings of this particular case, comparing them to those found within literature. We explore and consider the possibility of a Koebner effect secondary to radiotherapy, a neoplastic phenomenon or the possibility of these being cases of prior undiagnosed systemic mastocytosis.

P133

Improving Turnaround Times and Cost-Effectiveness of BRAF Mutation Testing for Malignant Melanoma Using Immunohistochemistry and a Rapid PCR Platform

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Background: BRAF inhibitor drugs (1) are an important line of treatment for melanoma and require rapid testing of melanoma for BRAF mutations. We have investigated the feasibility and validity of using newly acquired immunohistochemistry (2) and a rapid PCR platform (Idylla) in detecting BRAF V600 mutations (3) within a large NHS hospital. Previously, the trust sent formalin fixed, paraffin-embedded melanoma samples to an external laboratory using Cobas 4800 PCR platform (gold-standard) for BRAF testing (1) with an average of a 7-day turnaround which dermatology found too long.
Method: The study included 37 melanoma samples tested by Cobas which we then tested with immunohistochemistry using the BRAF V600E antibody. It was feasible to also test 17 of these with Idylla which requires more tumour cells than Cobas.
Summary of results: Cobas PCR results were compared with results obtained from Idylla rapid PCR and immunohistochemistry. Immunohistochemistry picked up all but 2 of the positive results from Cobas and there were no false positives. All the results from Idylla matched with Cobas results. Turnaround time for immunohistochemistry is 24 hours whereby for Idylla is 2–3 hours although the latter performed 2–3 times per week. Immunohistochemistry is much cheaper (£26) compared to Cobas (£131) and Idylla (£112).
Conclusion: Immunohistochemistry and Idylla are more cost and time effective than sending to an external laboratory and a strategic algorithm has been designed for BRAF testing using immunohistochemistry followed by Idylla for immunohistochemistry-negative cases to pick up the remaining 5% false negatives (3).
References: (1) Martin-Algarra S, Labiano T, Ignacio Echeveste J, Gomez N, Mercedes M. Use of Cobas 4800 BRAF mutation test for the analysis of BRAF V600 mutations in cytological samples (CS) from metastatic melanoma (MM). | Journal of Clinical Oncology. (2) Schadendorf D, van Akkooi A, Berking C, Griewank K, Gutzmer R, Hauschild A et al. Melanoma. The Lancet. 2018;392(10151):971-984. (3) Melchior L, Grauslund M, Bellosillo B, Montagut C, Torres E, Moragón E et al. Multi-center evaluation of the novel fully-automated PCR-based Idylla™ BRAF Mutation Test on formalin-fixed paraffin-embedded tissue of malignant melanoma. Experimental and Molecular Pathology. 2015;99(3):485-491.

P134

Angiomatoid Fibrous Histiocytoma: Report of a Rare Tumour at an Unusual Site and Review of the *EWSR1-CREB1* and *EWSR1-ATF1* Translocation Associated Neoplasms

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Purpose of the study: Angiomatoid Fibrous Histiocytoma (AFH) is a rare mesenchymal tumour arising in the subcutis of the extremities in children and young adults. AFHs harbour characteristic gene fusions involving the *EWSR1* gene (*EWSR1-CREB1*, *EWSR1-ATF1*) and *FUS-ATF1*, and belong to a family of translocation associated neoplasms. We present a case of AFH, highlighting diagnostic difficulties given the patient's age and the dermal location, and review the recent literature on this associated tumour family.
Methods: We retrospectively reviewed the histology, molecular features, and clinical records of a patient diagnosed with AFH at our institution, and undertook a review of recent literature on *EWSR1-CREB1* and *EWSR1-ATF1* associated tumours.
Summary of results: A 60 year old male presented with a one year history of a lump on the left ring finger. Histology demonstrated a dermal spindle cell tumour with a nodular and storiform pattern, with prominent pseudovascular spaces. Mitoses were readily apparent and cytological atypia with 'monster cells' was observed. The differential diagnosis included an aneurysmal fibrous histiocytoma vs AFH. Tumour cells expressed CD99, EMA and ALK. Break-apart FISH demonstrated a translocation involving *EWSR1* at 22q12. RT-PCR demonstrated an *EWSR1-CREB1* [t(2;22)(q34;q12)] gene fusion, confirming a diagnosis of AFH. This translocation is also found in tumours of both mesenchymal and epithelial origin.
Conclusion: This case of atypically presenting AFH highlights the value of molecular studies in establishing the correct diagnosis. An important pitfall is that many translocations are not tumour specific and are present in range of neoplasms with different therapeutic and prognostic implications. Correlation between the histology and molecular studies remains imperative. The mechanism by which common gene fusions can result in such a heterogeneous group of tumours is unclear, and warrants further study for potential therapeutic applications.

P135

A Case of Multiple Storiform Collagenomas in a Patient with Cowden Syndrome

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Introduction: Storiform collagenoma, also known as sclerotic fibroma, is a rare benign neoplasm and is one of the lesser known cutaneous manifestations of Cowden Syndrome (CS). CS is due to a germline mutation in the PTEN gene and is characterised by an increased propensity to numerous benign and malignant tumours including hamartomatous gastrointestinal polyps, breast carcinoma and thyroid carcinoma. We present a case of multiple storiform collagenomas in a patient with CS.
Case History: A 62 year old male presented with a three week history of a flesh coloured, painless papule on the thigh. Past medical history included CS and clear cell renal cell carcinoma. On examination there were multiple firm lesions on the hands, thigh and face. Histology of each lesion revealed a well-circumscribed, unencapsulated dermal nodule composed of spindle cells admixed with hyalinised eosinophilic fibres arranged in a storiform architecture. The spindle cells were CD34(+), S100(–) and EMA(–). The clinical differential diagnosis included metastatic renal cell carcinoma. The histological differential diagnosis included storiform collagenoma, perineuroma, dermatofibroma and Pacinian neurofibroma. A diagnosis of storiform collagenoma was made and excision was curative.
Discussion: This case highlights the importance of clinicopathological correlation, being aware of the association of storiform collagenoma with CS and the potential wider implication of a diagnosis of storiform collagenoma. The previous history of renal cell carcinoma and extensive family history of malignancies at a young age should prompt one to consider a hereditary cancer syndrome. Although in this case the patient had a known diagnosis of CS, a skin manifestation is one of the commonest first signs of the condition and therefore you can potentially make a huge impact to the patient and their family by raising the possibility of Cowden syndrome.

P136

A Case of Cutaneous Epithelioid Haemangioendothelioma Masquerading as a Sebaceous Cyst

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A 46 year old gentleman presented with a 2–3 year history of a lump on his scalp above the left ear. On examination this was a 2x2cm firm and tender lesion showing overlying erythema. Clinically the impression was of an inflamed sebaceous cyst which was subsequently excised. Macroscopically the specimen comprised an ellipse of skin with no obvious lesion seen on the surface. No cyst was identified and the cut surface showed a firm, pale appearance. On microscopy a well circumscribed lesion was present within the mid dermis extending into the subcutis. This was composed of sheets and cords of epithelioid cells with abundant pale eosinophilic cytoplasm, many of which showed an intracytoplasmic lumina containing a red blood cell. There was moderate cellular atypia with nucleolation, very occasional mitotic figures and a few apoptotic cells. The centre of the lesion showed a hyaline-myxoid matrix. On immunohistochemistry the cells were strongly and diffusely positive for CD31 with patchy staining for CD34. SMA and EMA were negative with very weak but diffuse staining for cytokeratin MNF116. The MIB1 proliferation index was very low. Overall the morphological and immunohistochemical features were consistent with a lesion of endothelial cell origin and given the lack of lobular architecture and inflammation but presence of moderate atypia was diagnosed as an epithelioid haemangioendothelioma. The patient is undergoing further radiological investigation to assess for multisystem involvement. Epithelioid haemangioendothelioma is a vascular neoplasm with behaviour lying between an epithelioid haemangioma and angiosarcoma. The majority arise within soft tissues and visceral organs such as the lung and liver. Solely cutaneous presentations are rare, usually found on the extremities and can be mistaken for benign entities clinically. Although deemed low grade these tumours have a potential for metastatic behaviour, however where resectable have a better clinical prognosis.

P137

HP1BP3 Expression and its Significance as a Prognostic Marker in Human Primary and Metastatic Malignant Melanoma

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Melanoma is the deadliest form of skin cancer, accounting for 80% of all skin cancer related deaths. As such, research into novel proteins as potential prognostic indicators is vital. Despite the necessity, clinical relevance of these so far has been debatable. Epigenetic regulators, such as proteins involved in maintaining chromatin structure or regulating the cell cycle have been of particular interest. HP1BP3, a member of the H1 linker histone family, is one example of these. This study was conducted to investigate the association between HP1BP3 expression and survival of primary and metastatic melanoma patients. To determine HP1BP3 expression in patient melanoma samples, immunohistochemistry (IHC) was used to stain TMA slides, consisting of primary (n=451) and metastatic (n=448) cases. Tumour cores were scored using the H-score method, accounting for percentage positivity and intensity of staining in the nuclei of melanoma cells. High expression of HP1BP3 was associated with higher recurrence-free survival rate in primary melanoma patients(p= 0.04*) which was confirmed by multivariate analysis. High HP1BP3 expression in primary melanoma cases was also associated with lower mitotic rate (p= 0.04*) and focal tumour infiltrating lymphocytes (TILs), (p= 0.04*). Trends were seen between high HP1BP3 expression and overall survival (p= 0.08) and absence of a BRAF mutation (p= 0.05) in the primary cohort. No other significant associations were found. These findings suggest that high HP1BP3 expression is an independent prognostic factor, predicting longer recurrence-free survival in primary melanoma patients. This result, and the associations between HP1BP3 and known prognostic factors, necessitate further research into the role of HP1BP3 in melanoma.

P138

Detection of the BRAF V600E Mutation in Primary and Metastatic Malignant Melanoma: An Evaluation of the Immunohistochemical Approach

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The BRAF V600E mutation is found in over half of melanomas, making it an attractive therapeutic target; this led to the development of anti-BRAF drugs. Detection of this mutation is currently performed using pyrosequencing. A monoclonal antibody specific for the BRAF V600E mutant protein has been developed, thus immunohistochemistry (IHC) can be used for detection. The primary aim is to evaluate IHC as a method of BRAF V600E detection compared to pyrosequencing. Secondary aims include analysing clinicopathological and survival data for primary melanoma cases with comparison to their BRAF V600E mutation status. After collecting the pyrosequencing data from patient records, 5 primary and 4 metastatic melanoma tissue microarrays were stained using the locked down protocol installed on the Roche Ventana Benchmark Ultra platform. The anti-BRAF V600E (VE1) mouse monoclonal antibody (Roche) was used. Informed consent from patients was obtained for their tissue to be used and stored under the Nottingham Health Science Biobank. Slides were scored using the H-scoring method and verified by a pathologist. SPSS v24.0 was used for statistical analysis. Sensitivity and specificity of the antibody compared to pyrosequencing were deduced for the primary cohort (65.91% and 91.94% respectively) and the metastatic cohort (86.59% and 98.35% respectively). BRAF mutation status was significantly associated with age and site (p<0.001), ulceration (p=0.027), mitosis (p=0.011), tumour-infiltrating lymphocytes (p=0.042), and histological subtype (p=0.013). There were no significant associations between survival and BRAF V600E status. The clinicopathological prognostic factors provide evidence for the BRAF V600E mutation as a positive prognostic indicator. IHC could be used to screen for BRAF V600E mutations, with negative cases being referred for molecular testing like pyrosequencing. This would ensure the detection of other BRAF mutations that could benefit from anti-BRAF drugs.

P139

Does Melanocortin-1 Receptor (MC-1R) Play a role in Acne Vulgaris? An Immunohistochemical Study of Egyptian Patient

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Purpose: Acne vulgaris (AV) is a prevalent chronic inflammatory disease of the pilosebaceous units. The exact pathogenesis of AV remains to be clarified, although several pathogenetic components like sebaceous gland hyperplasia with increased sebum production and inflammation and psychoemotional stress have been palmed. Most cutaneous cell types express melanocortin receptors (MC-Rs) and synthesize melanocortin (MCs), both of which regulate melanogenesis and affect non-pigmentary processes, such as inflammation, apoptosis and sebogenesis. This study aimed to investigate immunohistochemical expression of MC-1R in acne lesions (scarring and non-scarring) versus acne- free control subjects.
Methods: This prospective case-control study was conducted on 90 subjects; Group A: Including 30 subjects with non scarring acne, Group B: Including 30 subjects with scarring acne and 60 patients with acne vulgaris (AV) and Group C: Including the 30 healthy controls. Immunohistochemical expression of MC-1R was evaluated in dermis and epidermis of skin biopsies and results were correlated with available clinical data.
Results: Compared to control group, all dermal elements of non scarring acne patients; sebaceous glands (P=0.001), hair follicles (P=0.003), sweat glands (P=0.01), fibroblasts(P=0.001), inflammatory cells (P=0.02) showed significant increase in intensity of MC-1R expression. Similarly, the intensity of MC-1R expression in hair follicles (P=0.007), sweat glands (P=0.004) and inflammatory cells(P=0.006) was significantly stronger in scarring acne patients than controls.
Conclusion: MC-1R may play an active role in early pathogenesis of AV and may contribute to the severity outcome of the lesions and scarring development which open the door for new therapeutic strategies for this disease.

P140

An Unusual Case of Hyperkeratosis

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Hyperkeratosis lenticularis perstans (Flegel's Disease) is a rare, hyperkeratotic skin disorder which typically presents on the lower extremities of Caucasian middle-aged patients. Most cases are sporadic although familial cases with an autosomal dominant mode of inheritance have been reported. Clinically the condition mimics many other hyperkeratotic and inflammatory disorders and the diagnosis is only confirmed on histopathological and clinical correlation. The condition manifests with asymptomatic keratotic/scaly red/brown papules which histomorphologically show lamellar hyperkeratosis with abrupt peripheral basket-weave orthokeratosis, irregular acanthosis and underlying lichenoid lymphocytic infiltrate. The pathogenesis is unknown although ultraviolet light and cell-mediated cytotoxicity against epidermal cells have been implicated. Herein we describe an unusual case of hyperkeratosis in a 44 year old female who was referred with numerous light brown papules on both her upper and lower extremities and neck. A punch biopsy revealed skin containing a central area with abrupt compact hyperkeratosis and focal parakeratosis, epidermal thinning, interface dermatitis with occasional colloid bodies and underlying lichenoid chronic inflammation. The clinical and histomorphological appearances were in keeping with hyperkeratosis lenticularis perstans. There was no evidence of dysplasia or malignancy.

P141

Audit of Radical Lymph Node Dissection Involvement by Metastatic Melanoma in Patients with a Positive Sentinel Lymph Node

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Introduction: The Scottish Intercollegiate Guidelines Network (SIGN) guideline for melanoma advises that radical lymph node dissection (RLND) is indicated in patients with metastatic melanoma in a sentinel lymph node (SLN). RLND carries a risk of significant morbidity and anecdotal evidence suggests that most patients with a positive SLN have a negative RLND. This audit analysed whether there are characteristics of the primary melanoma, the SLN, or the patient that are associated with having a negative RLND.
Methods: A retrospective analysis was performed of anonymised data from all patients who had melanoma, a positive SLN, and subsequent RLND between 2006 and 2018 (n = 38) in our centre. Information regarding the primary melanoma and the RLND was extracted from reports. The slides of the SLNs were reviewed by a consultant dermatopathologist and data regarding tumour burden (1mm or less vs greater than 1mm, subcapsular vs parenchymal) were recorded. The data were analysed using SPSS Statistics.
Results: 66% of patients with a positive SLN had a negative RLND. 88% of patients with subcapsular metastatic melanoma only in their SLN and 79% of patients with metastatic melanoma measuring 1mm or less in their SLN had a negative RLND. The mean Breslow thickness of the primary melanoma in patients with a negative RLND was slightly less than that for patients with a positive RLND (3.11mm vs 4.99mm, $p = 0.04$). The mean age of patients with a negative RLND was significantly lower than that of patients with a positive RLND (47 vs 63, $p < 0.002$).
Conclusion: Factors such as SLN tumour burden, pT stage of the primary melanoma or the patient's demographic might help predict which patients are likely to have a negative RLND and could therefore spare them the associated morbidity. This would also reduce the medical, surgical and pathological resources required in managing these patients.

P142

Benign Hidradenoma Metastasising to Regional Lymph Node: A Case Report of a Rare Entity

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Nodular hidradenoma is a benign adnexal tumour of sweat gland origin that usually presents as a solitary solid or cystic nodule in the skin. The course is usually benign with no loco regional recurrence or distant metastasis. However rare cases of hidradenoma with benign histology but showing vascular invasion have been reported in literature. Even rarer are benign hidradenoma with regional lymph node metastasis which to the best of our knowledge, only three cases have been reported so far. We hereby report a forty nine year old gentleman who presented clinically with a 1.5cm intact cyst in the right shin. On excisions biopsy, this was a dermal based tumour with ductal differentiation exhibiting histological features of a benign nodular hidradenoma with no atypical features. However an excision biopsy of a right groin lymph node that was simultaneously submitted contained a deposit of nodular hidradenoma. This again did not show any atypical features other than it was a "metastasis" in the lymph node draining the dermal benign hidradenoma. This was later confirmed to be a "benign metastasis" of nodular hidradenoma after expert consultation. "Benign" metastasis of nodular hidradenoma to the lymph nodes is a challenging entity that needs to be recognized by the pathologists. It is probably prudent to pursue a long term follow up in these patients to confirm that the course is benign. The few similar cases that have been reported so far in the literature have shown an uneventful long term follow up. In our case, the lesion in the skin was completely excised and long-term clinical follow up was recommended.

P143

Audit of Dysplastic Naevi in a Tertiary Referral Centre

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Dysplastic naevi (DN) are defined as atypical clinically and/or histologically by architectural and cytological atypia. They were first reported in 1987 by Clark, Lynch and Elder as histologically defined lesions in melanoma-prone families. Despite the multiple consensus conferences and studies with regard to their diagnosis, guidelines for diagnosis and management including wider excision remain unclear. In this audit, we reviewed the diagnosis of DN in a tertiary referral centre over a one year period with a 6 year follow up to look at the burden of DN within our service and the excisional status of these naevi. All DN diagnosed in 2012 were retrieved from the pathology files at our institution with a 6 year follow up. DN accounted for 19.3% (425/2205) of melanocytic lesions diagnosed with 79.1% of those diagnosed as mildly DN. Moderately DN accounted for 14.6% and 5.2% were severely DN. Of these, 407 cases were classified as excisions while the remaining 18 were diagnostic biopsies. Of 407 excisions, 302 were documented as being completely excised while a further 99 had positive or close margins (defined as ≤1mm to margin). Of these 99 cases, only 17 (17.2%) had a re-excision, 7/17 (41.2%) of these being mildly dysplastic, 4/17 (23.5%) moderately dysplastic and 6/17 (35.3%) severely dysplastic. Of those cases that did not have a re-excision, the majority (55/76, 72.4%) were mildly DN with 3 severely DN. No recurrence or new naevi in the vicinity were documented during the 6 year follow up in 69/76 cases while the remaining 7 had a DN diagnosed in the vicinity. DN account for a significant proportion of melanocytic lesions submitted for histopathological examination, the majority of which are mildly dysplastic. Despite only 17.2% of cases with positive or close margins on initial excision having a re-excision, no definite recurrence of a dysplastic naevus was reported in our series suggesting that wider excisions of dysplastic naevi are not always necessary.

P144

The Role of FOS Expression in the Diagnosis of Bone-Forming Tumours: Osteoid Osteoma / Osteoblastoma and Osteosarcoma

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Purpose of the study: Osteoblastoma and osteoid osteoma are together the most frequent benign bone-forming tumour, arbitrarily separated by size: the former being more than 2cm in greater dimension. Although in most clinical scenarios these are straight forward diagnoses, in some instances, it can be very difficult to differentiate osteoblastoma from osteosarcoma, in particular the osteoblastoma-like variant. Following our group description of FOS gene rearrangement in these tumours, the aim of this study is to evaluate the value of immunohistochemistry in osteoid osteoma, osteoblastoma and osteosarcoma for diagnostic purposes.
Methods: Spinal and non-spinal osteoblastomas (n=83), osteoid osteomas (n=33) and sequential biopsies of osteosarcomas (n=215) were retrieved from the files of the Royal National Orthopaedic Hospital and Basel University Hospital. A total of 332 cases were tested with antibodies against cFOS.
Results: 83% of osteoblastomas and 73% of osteoid osteoma showed significant expression of cFOS in the osteoblastic cell tumour component. Of the 19 cases negative for cFOS expression, 4 showed FOS gene rearrangement by FISH and 12 were non-informative for FOS and FOSB FISH. No additional cases of FOSB rearranged were identified. Of the osteosarcomas, 10% showed cFOS expression, usually focal and in highly atypical areas. 4% of the cases showed more conspicuous expression.
Conclusions: FOS rearrangement is the genetic abnormality underpinning osteoblastomas and osteoid osteoma. cFOS immunohistochemistry is positive in the vast majority of such cases. Expression, usually focal or patchy, is seen in up to 14% of osteosarcoma biopsies. In most osteosarcomas the expression is seen in highly atypical, non-osteoblastic areas, which would not be included in histological differential diagnosis. Our findings highlight the importance of undertaking assessment of expression patterns of antibodies in the light of morphological, clinical and radiological patterns.

P145

Exploration of the Cancer Immune Cell Infiltrate in Sarcomas for Clinical Utility

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Background: There is emerging evidence that cancer cell intrinsic properties such as neo-antigen burden are suitable biomarkers for immune therapy stratification. Most sarcomas generally do not respond well to immunotherapies with the exception of genomically complex subtypes such as undifferentiated pleomorphic sarcoma (UPS). There is therefore an unmet need to understand what biological mechanisms contribute to responses and the lack thereof. There is also a need to identify new biomarkers for immunotherapy clinical trials.
Aims: (1) To describe the immune landscape in soft tissue sarcoma. (2) Determine the intrinsic genomic cancer cell features associated with the immune microenvironment phenotypes.
Methods: Analysis was performed on a cohort of 206 sarcomas from The Cancer Genome Atlas and 59 UPS where RNA-seq, methylation arrays and whole genome/exome sequencing was available. Immune cell decomposition was performed using bio-informatic tools on RNA and methylation data. These were validated using immunohistochemistry and correlated with genomic features such as tumour mutational burden, neoantigen repertoire and HLA typing, mutation and loss of heterozygosity.
Results: We found that methylation data was more robust than gene expression for informatic estimates of immune cell composition. The dominant immune cell type were macrophages - predominantly of the M2 type. We also found an enrichment of the antigen presentation machinery, IFN γ response and an increase in the CD8 T cells in the samples showing a hypermutator phenotype.
Conclusions: Aneuploidy is a marker of immune evasion in sarcomas. Immune "hot" patterns are underpinned by high tumour mutational burden which may be a useful biomarker for patient selection for immune checkpoint therapies in sarcomas.

P146

The 100,000 Genome Project: Our Experience as a Specialist Sarcoma Centre

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Purpose of the study: As a specialist centre for bone and soft tissue sarcoma, we participated in the 100,000 Genomes Project, now complete, delivering samples from 609 patients with sarcoma. From August 2019, whole genome sequencing (WGS) for all sarcomas becomes available as standard of care for the NHS. Lessons learnt from our experience can ensure we make the most of this initiative and deliver the best care for patients.
Methods: Analysis of the patient recruitment and sample collection pathway using data from our laboratory information systems.
Summary of results: Of 975 recruited participants, tumour DNA was not sent for WGS from 359 patients (37%). Of these, frozen tissue was not available from 40% (n=142) of patients. Records showed that tumours were often considered too small to freeze (n=44/142, 31%), and 39/142 specimens (27%) were placed in formalin either in error or when operations occurred out of hours. Samples were frozen from the remaining 217/ 359 patients (60%) and were cut for DNA extraction but yielded insufficient DNA for WGS. These tumours were either of low cellularity, many of which were low grade, or were extensively necrotic, accounted for by the nature of the disease, or use of neoadjuvant chemo or radiotherapy (n=40). 208 biopsies were frozen, of which 98 (47%) were submitted for WGS. Insufficient yield of DNA or biopsies thawed to provide a diagnosis explained the greater part of the 110 (53%) samples not sent. DNA from a subsequent frozen resection was available in some cases. Frozen tissue was never available when patients were biopsied offsite.
Conclusion: The number of samples sent for WGS can be increased by simple measures such as those introduced in our centre, which included removing formalin from theatres, storing specimens at 40C at night and weekends and educating staff about freezing small tumours and biopsies. Engagement with radiologists to increase numbers of biopsy cores may help to overcome the impact of neoadjuvant therapies.
Supported by The Pathological Society and Cancer Research UK.

P147

Expression of Hormone Receptors and PARP-1 in Aggressive Fibromatosis

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Purpose of the study: Aggressive Fibromatosis or desmoid tumour, is a benign, but locally invasive entity. State-of-the-art treatment is mostly radical excision. However, incomplete surgery leads to a high risk of recurrence. Hormone modifying therapies were reported to be successful in several cases but need additional evaluation.
The DNA-repairing enzyme Poly ADP Ribose Polymerase-1 (PARP-1) might contain therapeutic potential, suggested by successful trials of PARP-inhibition in other malignancies, especially in carcinomas and selected sarcomas.
Methods: In this study, we retrospectively investigated the expression of the hormone receptors: estrogen receptors (ER) α and β, progesterone receptor (PR) and androgen receptor (AR), as well as PARP-1 by immunohistochemistry and quantitative RT-PCR in tissue samples of Aggressive Fibromatosis (n=69). Immunoreactivity scores were employed to quantify staining status. PCR-results were numerically analysed as well as using explorative cutoffs. The obtained expression patterns were correlated with clinical-pathological parameters in order to detect prognostic factors.
Summary of results: The analysed hormone receptors showed mostly no reactivity to immunohistochemical staining. PARP-1 on the other hand exposed variable nuclear positivity in all stained samples. Univariate survival analysis portrayed higher ERα expression to be a negative prognostic factor (p=0.005). Multivariate analysis demonstrated that higher PARP-1 expression is associated with earlier relapse (p=0.003). In general, survival analyses underlined that recurrent tumours relapse faster than primary tumours (p<0.001).
Conclusions: According to this study, PARP-1 expression is associated with poorer prognosis, i.e. faster tumour recurrence. PARP-1 expression could therefore be an interesting target for a new and rather personalized treatment. Hormone receptor status has limited prognostic value in our study.

P148

Proximal-Type Epithelioid Sarcoma: Case Report of a Rare Aggressive Tumour.

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Proximal-type epithelioid sarcoma is a rare aggressive tumour. In contrast to the classic-type, it occurs in an older age group and at more proximal sites. It is associated with frequent recurrences, early metastasis and a high mortality. Definitive diagnosis relies on histological examination. Early detection, complete surgical excision +/– adjuvant therapy and close follow-up remain the mainstay of treatment. We report a case of a 49 year old male presenting with a lump in the left groin. An initial diagnostic biopsy suggested a high grade malignancy with possible origin from the urinary bladder in view of the GATA3 positivity. However clinically there was no bladder lesion. Imaging confirmed a 106mm soft tissue mass within the lower abdominal wall. The excision specimen showed a 90mm tumour in the deep soft tissues. Histology revealed epithelioid cells with rhabdoid morphology which led us to consider a wide range of differential diagnosis including metastatic poorly differentiated carcinoma, melanoma, leiomyosarcoma, rhabdomyosarcoma, angiosarcoma and anaplastic large cell lymphoma. A wide panel of immunohistochemical markers showed positivity for AE1/AE3, EMA, Vimentin, GATA3 and loss of SMARCB1 (INI1) in the tumour. A diagnosis of proximal-type epithelioid sarcoma was made. Material sent for cytogenetics showed bi-allelic inactivation of SMARCB1 (INI1) consistent with epithelioid sarcoma. The explanation for GATA3 positivity remained unclear. The patient on follow-up had recurrent disease with widespread systemic and nodal metastasis, ultimately resulting in patient's death. We report this rare sarcoma with a wide clinical and histological differential diagnoses. We want to highlight the pitfalls caused by GATA3 positivity which initially resulted in unwarranted investigations for a bladder primary. Pathological diagnosis is vital for early recognition and timely management with close follow-up in view of its poor prognosis.

P149

Intestinal Secretory Leukocyte Protease Inhibitor (SLPI) is Induced by Repetitive Microbial Contact and is Increased in a Subgroup of Inflammatory Bowel Disease (IBD) Patients

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IBD patients are a heterogeneous group with varying therapy responsiveness. One cause for this variation may be the dual role for innate immune defects. Patients may have loss of control of the innate immune system, causing hyperactive immune responses to harmless bacteria. Alternatively, patients may have hyporesponsive immune function, resulting in failure of microbial eradication. Strikingly, methods identifying these patient groups are lacking as these immune processes are not fully elucidated. Recently, we have observed that intestinal SLPI expression is driven by repetitive microbial interaction. SLPI inhibits NF-κB activation and reduces host responses to harmless bacteria. Therefore, we hypothesized that high intestinal SLPI expression identifies IBD patients with insufficient anti-microbial activity. We analysed 84 biopsies from therapy-naïve paediatric IBD patients by quantitative PCR and observed a 10-100 fold increased SLPI mRNA expression in macroscopically inflamed ileac and colonic biopsies compared to non-inflamed biopsies. Next, we analysed SLPI protein expression by immunohistochemistry in 57 biopsies from paediatric IBD patients. Ileac SLPI protein expression was low, but colonic SLPI was increased in approximately half of the IBD patients. To assess whether high SLPI correlates with insufficient anti-microbial activity, we compared biopsies from a chronic granulomatous disease (CGD) patient (defective microbial killing) to an interleukin-10 receptor alpha (IL10RA) deficiency patient (hyperactive immune system) and found high SLPI protein expression in the CGD patient but nearly absent expression in the IL10RA deficiency patient. Our data suggests that high intestinal SLPI expression identifies a subtype of IBD possibly reflecting insufficient anti-microbial activity. To demonstrate this, we are currently investigating how intestinal SLPI expression relates to antimicrobial antibody levels and therapy responses in a larger therapy-naïve IBD cohort.

P150

Combining RNAscope and IHC results to Identify a C-MET Aberrant High-Risk Colorectal Cancer Patient Subgroup with a High RNA and Low Protein Expression Profile

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Colorectal cancer (CRC) is Europe-wide the second most common cause of cancer death and represents with several different phenotypes. c-MET overexpression is associated with poor outcome and c-MET inhibitors were suggested to be applicable to resistant tumours. Despite promising results in clinical phase 1 and 2 trials, phase 3 trials tend to be unsuccessful. A reason for the latter could be the lack of an officially regulated and well-established scoring system for c-MET. The aim of this study was to analyse c-MET DNA amplification as well as to score RNA and protein levels, correlate these results, evaluate them statistically and identify the prognostic significance of aberrations.
The present study includes 241 FFPE tissue microarrays (TMAs) of CRC patients that were stained during double-DNA in-situ hybridization (DDISH), RNAscope processing and immunohistochemistry (IHC). The samples were evaluated by digital pathology methods. We show for the first time that a high-risk c-MET aberrant subgroup must be identified by a combination of RNA and protein evaluating methods. Furthermore, our results suggest that the high-risk patients (HR = 2.1, 95% CI = 1.2 - 3.67) that present with a 35 % lower 5-year survival, display a phenotype comprised of high RNA levels and low protein expression. This finding contrasts with earlier studies that correlate protein overexpression with poor prognosis but are in line with recent study findings from 2016. It is crucial to identify the biological mechanism behind this phenotype to establish appropriate treatment options. Furthermore, we suggest to conduct a more detailed analysis concentrating on focal and invasive edge expression of c-MET.

P151

The Molecular Aspects of Aberrant Negative P53 Immunohistochemistry in Barrett's Oesophagus: A Pilot Study

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Purpose: p53 immunohistochemistry is an adjunct in the histological diagnosis of dysplasia in Barrett's oesophagus (BO). Previously p53 immunohistochemistry assessment was binary with recognition of aberrant positive or normal staining. Currently an aberrant negative/null staining pattern is also recognised, which results in total loss of staining compared to background wild-type/normal staining and may carry a greater risk of progression to adenocarcinoma. This pilot study aims to determine the molecular biology of aberrant negative p53 BO-related dysplasia using Next Generation sequencing (NGS) in endoscopic biopsies and assess if mutations can also be detected in non-dysplastic BO.
Methods: Biopsies showing BO-related dysplasia with aberrant negative p53 immunostaining were identified from archives and reviewed for suitability. Tissue was macrodissected from annotated sections, with separate dissection of dysplastic and non-dysplastic tissue. DNA was extracted and analysed using NGS (Illumina MiSeq) and Sanger sequencing.
Results: 12 cases were identified for this pilot. 3 cases provided separate samples of dysplastic and non-dysplastic areas. 44 TP53 mutations were detected in 9/12 dysplastic samples and 3/3 non-dysplastic samples. The most common mutations seen were missense (61%) with exon 4 most frequently affected (61%). Sanger sequencing on 4 cases did not detect any mutations.
Conclusions: NGS has characterised TP53 mutations in biopsies showing BO-related dysplasia associated with aberrant negative p53 immunostaining and may be more sensitive than Sanger sequencing. No single defining mutation was identified but missense mutations in exon 4 were most frequent. NGS also identified mutations in separate non-dysplastic BO. Further studies can be expanded to include cases BO-related dysplasia associated with wild type and aberrant positive p53 staining and non-dysplastic BO. Such studies may help to establish a role for NGS in future surveillance strategies.

P152

Cten Ability to Induce Migration is Independent of its Nuclear Localization

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Cten (Tns4) is a member of the tensin family and it is not expressed in normal colorectal tissue. However, in Colorectal cancer it is upregulated where it acts as an oncogene and enhances EMT, migration and invasion but not proliferation. The structure of Cten has a predicted Nuclear localization signal (NLS).NLS tags Cten protein to be imported to the nucleus. In abnormal situations like cancer for example, many proteins translocate to the nucleus and enhance migration, invasion and proliferation. In colorectal cancer, Cten is mainly expressed in the cytoplasm, however, it has been reported in that it translocates to the nucleus in several colorectal cancer cell lines and patient tissues. The translocation to the nucleus usually requires Nuclear Localization Signal (NLS) within the protein sequence. These amino acid sequence direct proteins to be translocated to the nucleus through binding to Importin. There is speculation about the role of Cten in the nucleus in colorectal cancer, however the exact biological function of Cten localization in the nucleus is to be investigated. We have used NLSMapper website to predict the nuclear localization signal of Cten. The predicted NLS was then deleted from the wild-type Cten plasmid using Site-directed mutagenesis. We then transfected HCT116 with Empty vector, wild-type Cten, and Cten(Δ NLS). This was followed by western blot, proliferation and Migration assay. Western blot and immunofluorescence data shows that deleting the predicted NLS prevented Cten protein from translocating to the nucleus. Cten induces migration but not proliferation in HCT116. The ability of Cten to induce migration was retained when we prevented the nuclear localization of Cten.
Conclusion: Cten translocation into nucleus was prevented by deleting NLS.
The ability of Cten to induce migration in colorectal cancer is independent of its nuclear localisation.

P153

Validating a Tissue Microarray Linked Colorectal Cancer Cohort

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Introduction: Tissue microarrays (TMA) offer an important way of testing the predictive and prognostic power of novel biomarkers. In order to be a useful biomarker assay, the TMA should be linked to a cohort of diagnostic and follow-up data. These data should be valid – one way of ensuring this is to demonstrate concordance with other published cohorts so that the conclusions are generally applicable. Here we test the validity of our new colorectal cancer cohort.
Methods: We identified 1000 eligible cases of colorectal cancer from our local pathology database, including full pathology reports of surgical biopsies and resections (initial diagnosis from 2008 to 2014). Patient demographics and follow-up data were gathered from the hospital patient information system. We investigated a range of known risk factors and their effects on overall survival, time to recurrence and cancer-related mortality.
Results: The average age was 68.8 (SD: 11.37) and 56.8% of patients were male. Mean follow-up time was 54 months (SD: 29). 88.6% were moderately differentiated (2% well-differentiated, 9.3% poor); the overall stage distribution was 1 – 16%, 2 – 40.1%, 3 – 32%, 4 – 11.9%. A range of established clinical (eg. overall stage, metastases) and pathological (eg. vascular/perineural invasion, T3/T4 stage, high lymph node stage) risk factors were associated with adverse prognosis (overall survival, cancer-related mortality, recurrence). We confirmed that for high tumour stage cancers, lymph node status has minimal further impact on prognosis.
Conclusion: We have constructed a valid colorectal cancer cohort which underlies our new tissue microarray. This will be an invaluable resource for investigating novel colorectal cancer biomarkers.

P154

Investigating the Role of TNS4 in the Colorectal Tumour Microenvironment Using 3D Spheroid Models of Invasion

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TNS4 (Tensin 4 or Cten) has been identified as a putative oncogene in colorectal cancer (CRC) contributing to the dynamics of cell adhesion, motility, invasion and epithelial to mesenchymal transition, as shown by vitro 2D-based assays. Our objective was to assess the role of TNS4 in 3D spheroid proliferation and invasion into the extracellular matrix. In order to mimic the 3D tumour microenvironment in vitro we used a spheroid model combining cancer-associated fibroblasts (CAFs) and TdTomato transduced colorectal cancer cell lines where TNS4 was stably knocked down via lentiviral transduction with PLKO.1 shTNS4. Cell growth was measured by spheroid volumetry, TdTomato fluorescence and 3D and quantification of DQ-BSA green degradation marker in the extracellular matrix was used as a measure of invasion. For the invasion assay, spheroids were embedded in basal membrane extract containing DQ-BSA green and overlaid by media containing EGF (40ng/mL) as a chemoattractant. Spheroids were imaged by confocal microscopy Z-stacks after 96h of invasion and the volume of emitted DQ-BSA green and TdTomato fluorescence was measured. TNS4 knockdown increased cell proliferation slightly, but significantly in cell lines producing compact spheroids, and also reduced adhesivity between CRC cells and CAFs or collagen type I. In general the addition of CAFs in spheroids supports proliferation of CRC cells, whereas CAFs themselves do not proliferate in low adherence conditions. The 3D invasion assay has shown a reduction of invasion with TNS4 knockdown, whereas addition of CAFs to spheroids increased the extracellular matrix degradation. In a 3D spheroid model, TNS4 seems to have an effect in modulating the ability of CRC cells to invade the extracellular matrix and their proliferation. These results confirm previous research using 2D-based assays and support the role of TNS4 as an oncogene in CRC, suggesting its value as an actionable therapeutic target to prevent early metastasis.

P155

Preliminary Report of TNS4 Overexpression in the ApcMin Mouse Model of Colorectal Cancer

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ApcMin mice constitute a gold standard experimental model of colorectal cancer, mimicking the familial adenomatosis polyposis (FAP) disease in humans. Tns4 is part of the tensin family, localized in focal adhesions and is considered a putative oncogene in colorectal cancer. Tns4 is overexpressed in inflammatory bowel disease and human colorectal tumours, from the earlier stages of adenoma development. Our objective was to assess expression of Tns4 in early stage adenomas developed in ApcMin mice. Samples were collected to RNA later from polyps and adjacent normal intestine of sacrificed ApcMin mice, at approximately 120 days old showing initial signs of anaemia. Swiss roll preparations of the intestine were used to detect Tns4 by immunohistochemistry. Each animal presented fewer than 12 polyps in the whole intestine, and in 50% of these polyps expression of Tns4 could be detected by immunohistochemistry, whereas normal areas were negative in the small bowel, but positive in the colonic crypts. Tissue obtained from Apc wild-type mice confirmed positive Tns4 expression in colonic crypts, but negative expression in the small bowel. RT-qPCR analysis showed approximately 3-fold upregulation of Tns4 in the polyps compared to adjacent normal tissue. Tns4 overexpression in intestinal polyps of ApcMin mice confirms previous results obtained in cases of human FAP patients and validates the suitability of this mouse model for further investigation of the role of Tns4 in colorectal carcinogenesis.

P156

CD26 Affects Metastatic Potential and Chemosensitivity Across Multiple Colorectal Cancer Molecular Subtypes

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CD26 is a transmembrane glycoprotein expressed on several cell types and has recently been identified to play a role in tumour biology. CD26 expression in colorectal cancer has been correlated to invasion, metastasis and poor clinical outcome. The functional mechanism of CD26 is not yet fully understood, therefore the aim of this study was to investigate the functional role of CD26 in generating the metastatic phenotype in more aggressive consensus molecular subtypes (CMS), specifically CMS3 and CMS4. CD26 was transiently knocked down by siRNA transfection in CRC CMS3 cell lines, HT29 and CL-34, and CMS4 cell line, HCT-116. The effect of CD26 knockdown on cell viability, migration and invasion were assessed, as well as the expression of epithelial mesenchymal transition (EMT) markers. Immunohistochemistry was piloted in a tissue microarray (n=84) to analyse expression of CD26 and associations with clinicopathological characteristics in primary colorectal cancer. Knockdown of CD26 in each cell line lead to a decrease in sensitivity to 5-FU. The migratory and invasive capabilities of each cell line were reduced when CD26 was knocked down ($p<0.05$) alongside changes in EMT-related markers. Expression of CD26 in tumour nuclei had a weak negative correlation to resection margin status and KRAS status ($p=0.05$ and $p=0.049$). The results suggest that CD26 plays a role in tumour progression and metastasis in both. CMS3 and CMS4 CRCs. However, knocking down CD26 may reduce chemosensitivity to 5-FU and hence further studies are necessary to elucidate its functional role in CRC.
This study was supported by a CDF from the Pathology Society.

P157

The Role of CD24/P-Selectin Complex in the Progression of Colorectal Cancer

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Introduction: Colorectal cancer (CRC) is a malignant neoplasm developed from the epithelium of the colon and rectum and metastasise to distant organs. CD24 is highly expressed in CRC cells and it has been shown to have a vital role in inducing cell proliferation, invasion and migration. It also acts as a ligand for P-selectin that is expressed in vascular endothelium.
Methodology: Two CRC cell lines, HCT116 (CD24-ve) and SW620 (CD24+ve cell) were used in this study. HCT116 was transfected with CD24-plasmid and SW620 was transfected with siRNA for CD24. The two cell lines were thereafter treated separately with P-selectin. The effect of P-selectin on CD24 expression and its downstream signalling molecules (Notch1, Cten, FAK, and ILK) was assessed by Western blotting. Functional assays on invasion and migration were also performed. Additionally, adhesion and trans-endothelial migration assays were carried out using endothelial cells (HUVEC).
Results: CD24 expression was increased in SW620 and HCT116-transfected with CD24 plasmid after stimulation with P-selectin. Along with the overexpression of CD24 in the presence of P-selectin, the downstream signalling molecules were also activated. Increased migration and invasion in these cells was observed upon treatment with the P-selectin. In addition, silencing either CD24 or P-selectin in HUVEC decreased the number of CRCs adhesion to HUVEC and similarly reduced their migration through endothelial cells.
Conclusion: P-selectin/CD24 signalling axis is responsible for the aggressiveness in the CRC cells and may facilitate metastasis of CRC cells through the vascular barrier.

P158

CD24 Modulates Angiogenesis in Colorectal Cancer

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Introduction: Colorectal cancer (CRC) is defined as a cancerous growth initiated by unregulated growth of cells or tissues in the internal lining of colon and rectum. The metastatic process requires the establishment of an appropriate local microenvironment, including angiogenesis, to support tumour growth. CD24 gene is highly expressed in CRC cells and has been shown to have a vital role to induce many cancer-like characteristics, such as invasion and migration.
Methodology: Two techniques of tube formation assay were performed to study the effect of CD24 on angiogenesis. Firstly, using conditioned media retrieved from CRC cells (HCT116 and SW620) that had been transfected with overexpressed CD24 plasmid or siRNA knockdown respectively. Secondly, performing an indirect co-culture where the CRC cells were on top of the Transwells insert and the HUVECs were at the bottom well coated with Matrigel. The migration of HUVECs was also studied using Transwells migration assay.
Results: An increase in the endothelial tubular formation was observed by in the presence of CD24 in either HCT116 overexpressing CD24 or SW620 siRNA control. However, in the absence of CD24, a significant inhibition in tube formation was determined. This data was confirmed by using indirect co-culture, which showed similar results. In addition, the number of migrated endothelial cells was increased when the CD24 positive cells were used as a chemoattractant compared with control.
Conclusion: CD24 expression induced endothelial tube formation and migration, which suggests it may have a major role in colorectal cancer progression by stimulating angiogenesis.

P159

TSC22D4 and the Metastatic Phenotype in Colorectal Cancer

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TSC22D4, a gene encoding a leucine zipper protein, has been hypothesized as being a transcription factor. Recent integrated profiling data from the Cancer Genome Atlas Network suggests that TSC22D4 may be a novel gene involved in vascular invasion and metastasis in colorectal cancer. This study investigated functional effects and clinicopathological associations of TSC22D4 in colorectal cancer. TSC22D4 was transiently knocked down with siRNA in CRC cell lines HCT116 and DLD-1. Proliferation, invasion, migration and wound healing assays were performed to determine functional activity. Survival of TSC22D4 knock-down cells treated with 5-FU were investigated by cell viability assay. Western blot was used to determine the expression of TGF-β and VEGF-C proteins. Immunohistochemistry (IHC) was performed on a pilot CRC tissue microarray to assess correlation with clinicopathological variables. TSC22D4 knock-down decreased the rate of migration, more significantly in DLD-1 than HCT116 cells. In DLD-1, knock-down increased TGF-β expression. No significant changes in VEGF-C expression were seen in either cell lines. Knock-down of TSC22D4 has a transient trend of decreased drug sensitivity (24 hours), which becomes abrogated at 72 hours. No significant clinicopathological variables were associated with expression on IHC. TSC22D4 seems to have effects on migration but not on invasion in some CRC cell lines. The lack of clinicopathological and functional correlations, suggests a passenger rather than driver effect and demonstrates the difficulties in translating metadata findings to clinically relevant functional studies.
This study was supported by a CDF from the Pathology Society.

P160

Cten is Overexpressed Under Hypoxic Conditions in Metastatic Colorectal Cancer Cell Line

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Introduction: Hypoxia is a condition characterized by low oxygen levels within tissue and is common in malignant tumours. Hypoxia stimulates a multitude of different signaling pathways in cancer cells and can specifically enhance epithelial-mesenchymal transition (EMT) leading to increased cell motility and metastasis. C-terminal tensin-like protein (Cten) is an adaptor protein that has been shown to induce EMT and migration in different cancer types including colorectal cancer (CRC). The goal of our study is to investigate the expression, regulation and role of Cten under hypoxic conditions in CRC.
Methods: The CRC cell lines SW480 and SW620 were incubated under hypoxic (1%O2) and normoxic conditions. Protein was extracted at 24, 48 and 72 hours. Western blot was used to assess Cten protein levels under both conditions. To see if Cten is regulated by HIF-1α, we silenced HIF-1α in SW620 using HIF-1α siRNA. To investigate the impact of Cten on HIF-1α we silenced Cten using Cten siRNA.
Results: Cten protein was upregulated under hypoxic conditions in SW620 but not SW480. Silencing HIF-1α in SW620 seems to reduce the expression of Cten under hypoxic conditions. Silencing Cten in SW620 seems to reduce the expression of HIF-1α under hypoxic conditions.
Conclusion: The upregulation of Cten in CRC under hypoxic conditions is cell line dependent. SW480(primary) and SW620 (metastatic) are derived from the same patients, therefore, it can be possible that Cten is upregulated under hypoxic condition in metastatic tissue only. Cten is regulated by HIF-1α under hypoxic conditions, moreover, the reduction of Cten expression in SW620 result in reduced Hif-1αexpression under hypoxic condition. These data suggest that Cten might be a contributor to hypoxia.

P161

The Value of Modified Davidsons Fixative (MDF) in Lymph Node Retrieval in Gastro-Intestinal (GI) Cancers

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Purpose of study: The identification of lymph node involvement in GI cancer specimens is important for prognosis and treatment. Our on-going study is examining the use of a proven lymph node retrieval solution MDF to establish an accurate and reproducible staging for GI cancers whilst minimally impacting an established diagnostic pathway.
Methods: The histological data was compared in a range of GI cancer specimens with and without MDF treatment. The final impact on diagnosis and the overall effects on integrating MDF into an established diagnostic pathway were examined.
Summary of results: Preliminary results have shown that on average a further 10 lymph nodes per case were found after treatment with MDF (n=16). The majority of these nodes were less than 3mm in size and difficult to see prior to treatment. Furthermore, the majority of these cases would not have had the minimum lymph node yield defined by RCPath guidelines before treatment. The use of MDF has been shown to be of no detriment to immunohistochemistry or molecular tests. The use of MDF has had minimal impact on cost and the established diagnostic pathway.
Conclusions: This preliminary study has shown that MDF has the potential to be a reproducible and accurate method for staging GI cancers whilst minimally impacting an established diagnostic pathway or further prognostic assays. Further work is continuing to establish its benefits in the upstaging in a range of GI cancers.

P162

Challenges and Temporal Trends in the Pathological Diagnosis of Indeterminate Colitis and Inflammatory Bowel Disease Unclassified: A Tertiary Centre Experience

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Arriving at a firm diagnosis of ulcerative colitis (UC) or Crohn's disease (CD) has important treatment implications; however, 5–10% of inflammatory bowel disease (IBD) cases defy subtyping and per RCPath guidelines should be classified as indeterminate colitis (IC) or inflammatory bowel disease unclassified (IBDU) at resection or biopsy, respectively. The aims of this study were to: 1) evaluate histopathologists' adherence to IBD classification guidelines; 2) identify the main challenges in subtyping IBD; 3) identify the broader multi-parameter and temporal diagnostic contexts of IC and IBDU. Electronic histopathological, endoscopic, radiographic, and clinical records were reviewed retrospectively for patients at a tertiary institution, diagnosed as IC or IBDU between 2008 and 2018 (mean duration of follow-up: ~7 years). There were 16 IC and 110 IBDU cases. IC and IBDU were appropriately labelled in 94% and 96% of cases, respectively. Occasional mislabelling resulted from the use of nonstandard terminology in pathology reports. The main factor that precluded IBD subtyping on colectomy specimens was overlapping histopathological features of UC and CD (88%). The main factors for IBDU diagnosis on biopsy samples were lack of specific histopathological features (43%) and conflicting findings on different modes of investigations (31%). A final diagnosis of IC was preceded by a consistent long-term history of UC in 31% and CD in 13% of cases. The majority of IBDU cases either had an initial diagnosis persisting as IBDU (45%) or were subsequently reclassified (31%) as UC or CD. Strict adherence to standard diagnostic terms is recommended to prevent mislabelling of unclassifiable IBD cases for both clinical and research benefit. Difficulties in subtyping arise even when a multidisciplinary approach is used. Emerging computational and imaging analysis techniques may help process this complex data and improve categorisation of difficult IBD cases.

P163

Increasing Patient Access to MSI Testing with Automated PCR

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Microsatellite Instability (MSI) is now a recognised feature of many common cancers and is the hallmark of Lynch syndrome. The MSI status of many tumours is increasingly sought urgently by oncologists for the MDT to help guide treatment and prognosis. In colorectal cancer (CRC) specifically, the use of MSI testing is well established and NICE guidelines now mandates screening all patients for MSI/Lynch syndrome. Despite this demand and the current national reorganisation of molecular diagnostics, there are difficulties in many centres in accessing testing. The Idylla™ MSI Mutation Assay (research use only, pending CE-IVD approval) offers an easy to use and automated platform that can be placed in any histopathology department and offers rapid, on-demand testing. The platform has shown promising results in other key targets for patients with CRC (BRAF, KRAS, NRAS) and together with new MSI assay the platform can offer complete molecular diagnostics and Lynch screening for patients with CRC. This study aimed to assess the concordance of this novel test with current standard methods. Thirty formalin-fixed, paraffin-embedded CRC tumour cases (15 MSI-High and 15 microsatellite-stable tumours by routine testing) were retrospectively analysed with the Idylla™ MSI Mutation Assay. Twenty seven of the cases had also undergone mismatch repair (MMR) immunohistochemistry (IHC) at the time of initial diagnosis. The concordance of Idylla with routine MSI testing (Promega) was 100% (95% CI 88.65% to 100.00%) and the concordance with IHC was 96.3% (95% CI 81.72% to 99.34%; the concordance of IHC and MSI testing at initial diagnosis was the same). The results show that the Idylla™ MSI Mutation Assay is a potentially accurate alternative to existing methodologies and together with the other assays available on the platform, could easily widen access to routine molecular testing for patients with CRC across the UK.

P164

Infiltrative Tumour Growth Pattern Correlates with Poor Outcome in Esophageal Cancer

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Esophageal cancer (EC) is an aggressive malignancy with a 5-year survival rate of 50% for localized tumours. Accurate prognostic markers which could guide treatment decisions in routine practice are urgently needed. Tumour growth pattern (TGP) has been shown to reflect tumour aggressiveness in a variety of tumours. However, limited data are available on the significance of TGP in EC. We performed a retrospective assessment of TGP in a group of patients with adenocarcinoma or squamous cell carcinoma. We searched for patients who had undergone surgery for EC from 2005 to 2017 at Institut Jules Bordet or Hôpital Erasme. Patients with haematoxylin and eosin stained slides from surgical specimens with a minimum of 10% of residual tumoural area over total tissue area were included. TGP was classified as either pushing (PP) if solid sheets of tumour cells present a well-demarcated tumour-stromal interface, or infiltrative (IP) if cords of tumour cells infiltrate the surrounding stroma in a spray-like pattern. Kaplan-Meier curves, Cox's proportional hazards model and log-rank tests were used with p values considered statistically significant at the bilateral <0.05 level. 101 patients were included. 35 had undergone surgery alone while 66 had received a neoadjuvant treatment (chemoradiotherapy in 23 cases ; chemotherapy in 43 cases). TGP was IP in 61 patients and PP in 40 patients. Patients with tumours with a PP had a significantly better OS (mOS 7,64 years vs 2,42 years) than those with tumours with IP (HR 0,56 [95% CI 0,33-0,93], p=0.03). After adjusting for prognostic variables including histology and disease stage in multivariate analyses, the association between TGP and OS remained unchanged (HR 0,5 [95% CI 0,28-0,9], p=0.02). This study shows that TGP is an independent prognostic factor in EC patients who undergo surgical resection of the primary tumour. Further studies are required to elucidate the molecular mechanisms underlying TGP and to characterize their dynamics.

P165

The Immune Landscape in Esophageal Cancer

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Esophageal Cancer (EC) is an aggressive cancer with an increasing worldwide incidence. With a 5-year survival rate of 50% for localized tumours, immunotherapy research holds the promise of new treatment options. Hence, characterizing the EC immune microenvironment is indispensible. We included 115 patients with adenocarcinoma (ADC) or squamous cell carcinoma (SCC). Surgical specimens were stained for CD3/CD20 or CD4/CD8 and scored for percentage of tumoural surface occupation, either in the intratumoural compartment (IC) or in the migration front (MF), by an experienced pathologist. Tertiary lymphoid structures (TLS) were counted. In case of (quasi-) complete response (pCR) to neoadjuvant treatment, the assessment was performed on the persistent scar. 15 negative proximal margins of gastric cancer were used as control. Mann-Whitney tests were used with p values considered statistically significant at the bilateral <0.05 level. Among 44 SSC, 8 patients received chemotherapy (CT), 19 radiochemotherapy (RCT), while 17 had no preoperative treatment. Among 71 ADC, 38 received CT, 15 RCT and 18 had surgery alone. 78,6% of the pCR cases received RCT, 21,4% received CT. Tumour samples without any preoperative treatment showed stronger infiltration of CD20, CD4 and CD8 than controls (p<0,0001). Treatment naïve SSC tumours were more infiltrated in CD4 than ADC (p=0,02) at the MF and showed stronger decrease in CD4 upon RCT (p<0,0001) than ADC. Remarkably, CD20 was decreased in the RCT group, in ESCC and ADC, in IC and MF (p≤0,0003). TLS were also less in the RCT group compared to patients with surgery alone (ESCC: p=0,004; ADC: p<0,0001). Concomitantly, pCR tumours were less infiltrated in CD20, CD4 and TLS in IC or MF. No association was found between CD8 and different types of histology or treatment received. While current research focuses on CD4 and CD8 cells for immunotherapy development, these preliminary results pinpoint a preponderant role of CD20+ B cells in EC.

P166

A Prognostic Signature of Brown Fat-Associated Proteins in Colorectal Cancer

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Colorectal cancer is a common type of malignancy with a relatively poor outcome and is one of the main contributors to cancer related deaths. Brown fat phenotype/proteins have been implicated in tumour growth and metastasis. Therefore, the aim of this study was to characterise the expression of brown fat-associated proteins cell-death-inducing DNA fragment factor 45-like effector A (CIDEA), elongation of very long fatty acids 3 and 5 (ELOVL3 and 5) and uncoupling protein 1 (UCP1) in colorectal cancer. Monoclonal antibodies to these protein targets were developed with short peptide immunogens which were selected using a range of bioinformatic tools. To select each peptide, the structural and physico-chemical properties of each protein were analysed. The antibodies were used to profile the expression of proteins by immunohistochemistry in a discovery cohort (274 primary colorectal cancers) and in a validation cohort (549 primary colorectal cancers). Unsupervised hierarchical cluster analysis was used to examine the overall relationship of proteins expression with overall survival and based on this identify a protein signature associated with prognosis. Cluster analysis of all proteins identified a cluster that was significantly associated with patient survival in the discovery cohort (HR=1.574, 95%CI=1.037-2.390, χ^2=4.658, p=0.031). Cluster analysis of the validation cohort also showed that the pattern of expression was significantly associated with patient survival (HR=1.691, 95%CI=1.284-2.228, χ^2=14.405, p<0.001). Multi-variate analysis confirmed that the cluster group was prognostically independent of clinically-established prognostic parameters (p=0.03). This study showed that novel targets CIDEA, ELOVL3, ELOVL5 and UCP1 are overexpressed in colorectal cancer. A prognostic signature of these proteins has been identified in colorectal cancer.

P167

Audit of Lymph Node Yield from Colorectal Cancer Resections 2005–2018

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Introduction: The Royal College of Pathologists (RCPath) dataset for reporting of colorectal cancer (CRC) highlights that there is variability between pathologists in the detection of lymph node (LN) involvement. A proportion of cases with low LN yield will be understaged because positive LNs may not have been processed. The dataset advises that the median number of LN examined should be 12 or more. Furthermore, the Scottish CRC Quality Performance Indicator (QPI) states that at least 90% of resection specimens should yield 12 LNs or more. This audit compared LN yield in CRC resections in our centre with the standards set by the RCPath and Scottish QPI.
Methods: A retrospective analysis was performed of anonymised data collected from all CRC resections reported by our centre (a regional cancer centre) from 2005–2018 inclusive.
Results: A total of 3614 cases were analysed. The mean age of patients was 69 and 55% were male. 27% of cases were rectal tumours (median LN yield = 18), 43% in the proximal colon (median LN yield = 19), and 30% in the distal colon (median LN yield = 17). 20% of cases had received neoadjuvant therapy, of which 94% were rectal tumours. Neoadjuvant therapy had little effect on the median LN yield (17 with therapy vs. 18 without). The Dukes stage also had little effect on LN yield (Dukes A = 16, Dukes B = 19, Dukes C = 18). Median LN yield ranged from 14 to 23, with a general trend of increasing LN yield over the period analysed. A median yield of at least 21 LNs was achieved since 2013. A median LN yield of 12 or more has been achieved in at least 90% of cases consistently since 2011.
Conclusions: The data demonstrate that our centre has either met or exceeded the RCPath standard for the past 14 years. Furthermore, the Scottish QPI target has been exceeded consistently since its introduction. The data demonstrate that these targets can be attained consistently in routine practice.

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This abstract has been withdrawn

P169

Audit of Colorectal Cancer Resection Reporting from January to September 2018

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Purpose of the study: To retrospectively review the histopathology reports of non-neoadjuvant colorectal cancer resections from January to September 2018, comparing the data reported with the recommended minimum standards set out by the Royal College of Pathologists (RCPath).
Methods: All cases, SNOMED coded as 'colorectal cancer' reported at this institute from January to September 2018 were identified and reviewed for the reporting of the recommended RCPath core items. The findings were then compared to the standards set by the RCPath. The RCPath stipulate that three standards be assessed on at least 50 non-neoadjuvant resection specimens for symptomatic, non-screening detected cancers. 1)The median number of lymph nodes examined should be greater than 12. 2)The frequency of serosal involvement should be at least 20% for colonic cancers and 10% for rectal cancers. 3)The frequency of venous invasion, including intramural (submucosal and intramuscular) and extramural, should be at least 30%. The key performance indices set by the RCPath state that proforma reports must contain at least 95% of the structured data.
Summary of results: On exclusion of 27 neoadjuvant cases, the remaining 58 colorectal cancer specimens from a 9 month period were audited. The median number of lymph nodes examined was 18, the frequency of serosal involvement overall was 24% and the frequency of venous invasion was 28%. Of note, only 2 cases (3%) reported the apical lymph node even though this remains one of the RCPath core items. Cases audited overall contained 94% of structured data. All cases were discussed at the gastrointestinal cancer MDT meeting.
Conclusion: This study revealed that while the majority of parameters audited were within the range expected as per the RCPath guidelines, some parameters fell just short of recommended standards. Further analysis of the parameters not meeting RCPath recommendation will be carried out in order to improve standardised reporting in the future

P170

An Unusual Presentation of Hepatoid Gastric Carcinoma

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The authors present an unusual presentation of an uncommon tumour in a female born in 1975. A presentation CT scan had shown widespread lymph node and skin metastases and an assumed diagnosis of a germ cell tumour was made clinically due to very high AFP levels. However, a biopsy of a groin lymph node and a gastric ulcer biopsy taken due to an upper gastrointestinal bleed showed evidence of a hepatoid carcinoma, presumed to originate from the stomach. The authors briefly review hepatoid carcinomas, discuss the role of immunohistochemistry in the diagnosis and outline their behaviour.

P171

Retroperitoneal Liposarcoma Mimicking a Primary Colonic Tumour: A Case Report

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Retroperitoneal liposarcoma is usually an incidental finding or patients may have non-specific symptoms such as increasing abdominal girth, abdominal and back pain. Some of the very unusual presentation include colonic intussusception with occasional cases reported in literature. We hereby report a 62 year old gentleman who presented with diarrhoea. He was noticed to have a large 15cm intussuscepting polyp in the transverse colon in Computed Tomography. There were several enlarged mesenteric lymph nodes with inflammatory changes in the mesentery. In addition two separate metastatic deposits were also noticed in the psoas muscle. A primary colonic tumour was radiologically considered and the patient underwent a right hemicolectomy. Histological examination revealed a largely ulcerated mesenchymal lesion involving mainly the submucosa and closely associated with the colon. This was composed of malignant pleomorphic spindle cells admixed with a large numbers of inflammatory cells including eosinophils. The spindle cells were positive for S100, Desmin, Vimentin and negative for AE1/AE3, HMB45, Melan A, SOX10, H-Caldesmon and ALK. The case was referred to centralised soft tissue pathological service for further opinion. Expert consultation confirmed well differentiated liposarcoma in the mesenteric fat with a dedifferentiated component mainly seen in the submucosa of the transverse colon that had mimicked a primary colonic intussuscepting polyp. Further retroperitoneal resection revealed extensive well differentiated liposarcoma in the retroperitoneum extending into the right renal hilum and encircling the adrenal gland. This case emphasizes the need for considering dedifferentiated liposarcoma in the differential diagnosis of malignant spindle cell lesions in the gastrointestinal tract and for careful examination of mesentery. This case was unusual as it had presented predominantly as an endoluminal polyp and radiologically also mimicked a primary colonic tumour.

P172

The Two Week Weight: The Increasing Burden of Targets in Histopathology

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Purpose and method of study: Increasing numbers of requests requiring prioritisation are being received by histopathology departments. We have analysed endoscopic specimens received as urgent (U), two week wait (2WW), Bowel Cancer Screening (BCSP) and routine (R) in our department from 2018 and a 'snapshot' week in March 2019 to assess this workload and to consider whether our service is being used appropriately.
Results: Jan–Jun 2018 Total endoscopy requests 7025. 2WW requests 1113 (20.9%). BCSP requests 1096 (15.5%) Jul–Dec 2018 Total endoscopy requests 7398. 2WW requests 1845 (25.4%). BCSP 1013 (13.7%). Week commencing 11 March 2019. Total endoscopic requests 320 (total biopsy sites 357) U=16.2%. 2WW=19.6%, BCSP=22.7%. R=41.5%. Using published guidelines for the appropriate use of histopathology for investigation of the upper and lower GI tract (UGI and LGI), an assessment was made for each specimen (excluding BCSP) in the study week and potentially inappropriate biopsies were recorded.
Potentially inappropriate requests UGI. U 32.0%. 2WW 36.9%. R 29.7%
Potentially inappropriate requests LGI. U 13.3%. 2WW 40.0%. R29.4%
Discussion: The results confirm the high proportion of a large GI workload that requires prioritisation. Prioritisation is disruptive in terms of workflow and impacts on the performance of scientific staff, medical staff and office staff to the detriment of specimens regarded as routine. There are frequent disruptions by specimen trackers and the responses take up additional time for all staff groups. The typing delay for routine specimens was 5 working days during the study week and has sometimes been as high as 10 working days. Additional resources are unlikely to be forthcoming in the current funding climate but the rational of use of histopathology in the investigation of the GI tract could reduce demand. Local discussions with gastroenterologists may be more beneficial than attempts to increase funding.

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Nicorandil Induced Enteropathy Clinically Mimicking Neuroendocrine Tumour (NET) of the Small Intestine: A Case Report

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A 66 year old man with a history of high blood pressure, angina, resected diverticular disease and incisional hernia, presented with weight loss, nausea, abdominal cramping and loose stools. A CT scan indicated subacute small bowel obstruction suspicious of a slow growing tumour. Octreotide scan showed local increased uptake suggestive of neuroendocrine tumour (NET). Increased serum Chromogranin A of 8.4nmol/L was also found and the patient underwent small intestinal resection under the cancer pathway. Macroscopic dissection showed a localised stricture with fibrosis, extramural abscess formation and diffuse longitudinal linear punctate ulcers throughout the small bowel. Histology revealed a patchy active chronic enteropathy with multifocal demarcated ulcers with little inflammation. The stricture showed a transmural, undermining ulcer with surrounding fibrosis. No malignancy, microorganisms or granulomas were found. Drug-induced enteropathy due to Nicorandil was concluded following close clinical-pathological correlation. The patient discontinued Nicorandil, with advice to the patient's GP to comprehensively review the patient's current medication regimen. He has remained well, with no further angina attacks. Nicorandil is a commonly used drug in the symptomatic treatment of angina. Rare, but known, complications of Nicorandil use include gastrointestinal ulceration, which may progress to perforation/abscess or fistulation. This case highlights that gastrointestinal side effects of Nicorandil can rarely clinically impose as a tumour. Increased awareness of potential Nicorandil induced enteropathy may help to recognise early lesions that may still respond to drug cessation. Nicorandil induced ulceration/perforation enters the differential diagnosis when encountering gastrointestinal ulceration of unknown cause at pathological examination.

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The Utilisation of CD31/AE 1/3 Dual Staining to Identify Venous Invasion in Colorectal Cancer

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Objective: To examine whether the use of dual staining (DS) for CD31\AE 1\3 will improve the detection of venous invasion (VI) in colorectal Cancer (CRC).
Background: Extramural venous invasion is a well-established independent prognostic indicator in CRC .Assessing VI is a requirement for TNM8. DS can be used to improve the detection of VI
Methods: A single- centre, retrospective review including the detection of VI using DS in 72 blocks from the first 18 CRC specimens, Stage I to III, reported in2019. Talbot's definition of VI was used subdivided into intramural (IM) and extramural (EM). The dual staining was carried out on a Ventana Bench Mark XT using monoclonal CD31 and AE1/AE3 antibodies. Ventana Ultra view detection kits where used alongside Ventana DAB and Ventana universal AP red chromogen systems.
Results: VI (IM and EM combined) was detected in 66.6% (12/18) on Haematoxylin and Eosin (H&E) preparations. 55.5% (10/18) was EMVI and 11.1% (2/18) IMVI. The use of DS for CD31/AE1/3 improved the detection rate of VI (IM and EM combined) to 88.8% (16/18).IMVI was identified in 38.8% (7/18) and EMVI in 50% (9/18).
Conclusion: The result of the current study introduces a novel approach with increased VI detection rate (88.8%) as compared to the previously published data using elastic staining which offers 56% detection rate. IMVI has been proven in recent meta-analysis to also be of prognostic significance. Standard use of DS amongst non-specialist gastrointestinal pathologists may enhance VI detection rate including IMVI.

P175

Audit to Assess the Impact of the Royal College of Pathologists Colorectal Cancer Dataset in a District General Hospital

Ⓟ D O'Dwyer; SH Heng

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Purpose: Colorectal cancer remains the third most common cancer in the UK and surgical resection the mainstay of treatment. To improve outcomes and standardize pathological assessment the Royal College of Pathologists (RCP) has outlined a colorectal cancer dataset. This audit aimed to assess the impact of this in a district general hospital.
Methods: Pathology reports for colorectal cancer resection specimens generated between two separate time periods (Jan 2014 – Dec 2014 and Apr 2017 – Apr 2018) were quality assessed against RCP dataset guidelines. These two time periods were then compared using the Fisher exact test.
Results: 229 pathology reports were generated between 2013–2014 and 99 for 2017–2018. In both groups the reporting of tumour site, tumour type, the extent of local invasion and lymph node status was 100%. Venous invasion, tumour differentiation and involvement of the longitudinal resection margin were reported in >90% in both groups. In the 2013–2014 group the reporting of certain parameters was below standard: maximum tumour diameter reported in 85.2% of cases, tumour distance to nearest longitudinal margin in 74.7%, tumour perforation in 19.7%, circumferential resection margin involvement in 74.2% and distant metastasis in 77.7%. Conversely these parameters were reported in >90% of the 2017-2018 group cases. When compared this was significant (p=0.0001). Additionally the median lymph node harvest increased from 13 in the earlier group to 17 in 2017–2018. Furthermore the proportion of cases with a lymph node harvest of >12 has increased significantly (p=0.0001).
Conclusions: Regular audit has been shown to improve patient outcomes in many specialties. This audit, undertaken in a district general hospital, has shown that the introduction of RCP guidelines has not only improved the quality of pathology reporting over time but also routine clinical practice.

P176

Comparison of Pathological and Radiological Reporting of Extramural Venous Invasion in Rectal Cancer

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Purpose: Extramural venous invasion (EMVI) is a poor prognostic indicator in colorectal cancer and an important factor in determining management during MDT discussion. EMVI detection is a core data item in the colorectal dataset. A detection rate of 25% is recommended. Evidence suggests that MRI may be more sensitive and provide an earlier opportunity to detect EMVI in rectal cancer compared to histological reporting. Royal College of Radiologists (RCR) guidelines advocate including EMVI in the MRI report. This audit aims to assess and compare the pathological and radiological reporting of EMVI in rectal cancer over time at a district general hospital.
Method: Pathology and MRI reports following rectal cancer resection generated between two separate time periods (Jan 2014 – Dec 2014 and Apr 2017 – Apr 2018) were quality assessed against RCP and RCR guidelines. These two groups were then compared using the Fisher exact test.
Results: During 2013–2014 there were 42 rectal cancer resections with both pathology and MRI reporting available and there were 26 for 2017–2018. Over 2013–2014 EMVI was included in 90.4% of pathology reports. Of those 18.4% were EMVI positive. In the 2017–2018 group EMVI was reported in 98% of pathology reports. Of these EMVI was detected in 24%. The increase in pathological reporting of EMVI between these time periods was proportionally significant (p=0.02). Comparatively EMVI was included in only 4.8% of the 2013–2014 MRI reports and 7.7% of the 2017–2018 reports. There was no significant improvement in MRI reporting rates of EMVI over time (p=0.63).
Conclusions: Clinical audit can help to improve patient outcomes and clinical practice. This audit has shown that pathology reporting of EMVI has improved over time whilst radiological reporting of EMVI has not and is overall poor. Regular inter-specialty audit and discussion should be performed to help improve communication between specialties, highlighting areas for improvement.

P177

Primary Pyloric Adenosquamous Carcinoma: A Rare Case Report

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Introduction: Primary gastric adenosquamous carcinoma (ASC) is an extremely rare malignancy, particularly in Western populations with most cases affecting those of Asian origin. We report a rare case of primary pyloric ASC in a Caucasian.
Case report: A 78-year Caucasian male presented with symptoms of gastric outlet obstruction and weight loss. Endoscopy showed non-erosive gastritis and a pyloric abnormality. The biopsy was not representative of the lesion, however due to clinical and radiological suspicions of malignancy he underwent sub-total gastrectomy.
Histology: Macroscopic examination revealed a polypoid, fungating and obstructing mass in the pylorus measuring 35 mm. Microscopic examination showed poorly differentiated adenosquamous carcinoma (adenocarcinoma 60% and squamous carcinoma 40%). This was confirmed by immunohistochemistry. Lymphovascular invasion was present with a total of 4/11 lymph nodes examined showing metastatic adenosquamous carcinoma.
Discussion: Primary gastric ASC commonly involves the distal stomach and tends to present with more advanced stage disease and have a poorer prognosis than gastric adenocarcinoma. Various hypotheses have been suggested as to the origin of the tumour, with that of the squamous carcinoma arising from adenocarcinoma being favoured at present. There is also some suggestion that the histological nature of the metastatic component either squamous carcinoma or adenocarcinoma may affect overall behaviour and prognosis.

P178

Audit on Comparison of Oesophageal Brush Cytology with Biopsy and Endoscopy in the Detection of Oesophageal Candida

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Purpose of the study: Endoscopic sampling when oesophageal candidiasis is suspected includes brush cytology and biopsy. Although studies have compared the relative yield of brush cytology to biopsy, it is not clear what is best practice for the diagnosis of oesophageal candidiasis. The aim of this audit is to evaluate the yield, sensitivity, and specificity of brush cytology vs. biopsy in the diagnosis of oesophageal candidiasis.
Methods: This is a retrospective review of 141 oesophageal brushings from 01/2014 to 02/2017 at a university hospital. Cytology results were compared to the final histology, where available, and endoscopy to assess the overall sensitivity and specificity.
Summary of results: Of the 141 cases, 68 had all three tests completed. Of the 68 cases, Candida was seen on cytology in 50 (74%) and on both biopsy and cytology in 6 (8.8%) cases. 63 of the 68 cases (92.6%) were identified to suspect Candida on endoscopy. 48 of the 63 cases (76.2%) were found to be positive on cytology. Two of 5 cases not suspected to have Candida on endoscopy had positive cytology. Endoscopy had a sensitivity of 0.96 (CI = 0.85-0.99) and a specificity of 0.16 (CI = 0.04-0.42), cytology had a sensitivity of 1 (CI = 0.91-1) and a specificity of 1 (CI = 0.78-1), biopsy had a sensitivity of 0.12 (CI = 0.05-0.25) and a specificity of 1 (CI = 0.78-1) for the detection of oesophageal Candida.
Conclusions: The sensitivity of brush cytology is significantly higher than biopsy and better than endoscopy for presence of oesophageal Candida. Endoscopy with brush cytology diagnosis appears to be an ideal method for diagnosing oesophageal Candida.

P179

Primary Malignant Melanoma of Oesophagus: A Rare Case Report

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Introduction: Primary Malignant Melanoma of the Oesophagus (PMMO) is a rare and aggressive malignancy, accounting for only 0.1 to 0.2 % of all oesophageal malignancies. Mean age of diagnosis is 60.5 years with male preponderance. We report PMMO in a young female with past medical history of stage V Wilms tumour at the age of 2.5 years with pulmonary metastasis treated with right nephrectomy and chemo-radiotherapy. Rare reports of malignant melanoma have been described following treatment for Wilms tumour.
Case report: A 34-year female presented with 6 months history of intermittent dysphagia, more persistent for last 6 weeks. Endoscopy revealed a 58 mm polypoid mid-oesophageal tumour and biopsy showed malignant melanoma. The tumour was staged as T3N2M0 on PET-CT scan and EUS. Two-stage oesophago-gastrectomy was performed.
Histology and follow up: Histology confirmed an ulcerated malignant melanoma with Breslow thickness of 6.2mm, mitotic count of 30/mm2 and lymph node metastasis. Junctional activity and in-situ melanoma were noted in the adjacent oesophageal squamous mucosa. Molecular testing showed no evidence of BRAF or KIT gene mutation. The tumour was staged as pT3N2M1 (oesophageal carcinoma), pT4bN3M1c (skin melanoma) and pT3N1M1 (upper aero-digestive tract melanoma). Based on morphology and immunohistochemical phenotype, and in the absence of a primary melanoma elsewhere, a diagnosis of PMMO was rendered. Despite palliative treatment with Nivolumab, the patient developed widespread metastatic disease and sadly died within 3-months of diagnosis.
Discussion: PMMO is a highly aggressive tumour with a poor outcome despite radical surgery and chemo-radiotherapy. Due to its rarity, our understanding of tumour biology is limited and there is current lack of consensus on treatment and staging guidelines. Recent molecular studies have shown different mutations in PMMO than seen in melanoma at other sites and this may be helpful in targeted immunotherapy for better survival.

P180

This abstract has been withdrawn

P181

Colon Biopsies: Too Many Pots!

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Maidstone Hospital receives specimens from three hospitals (A, B, C). A discrepancy was anecdotally appreciated between these sites in regards to the numbers of colonic biopsies received, specifically the number of specimens for "normal" colonoscopies. An investigative audit was undertaken to corroborate this observation and assess whether this practice was congruous with current guidelines.

Method: A retrospective Telepath search was performed from 1st Oct 2017 – 31st Dec 2017 using T codes for all lower GI specimens. Resection specimens, biopsies for dysplasia and miscoded cases were excluded. Data was collected for: hospital site, number of pots per case, number of biopsies per pot per case, clinical indication and final diagnosis. The data was analysed using an Excel spread sheet.

Results: A total of 1208 cases were identified for the 3 month period. This translated to 2885 pots and 6784 biopsies in total. The number of cases per site per month were roughly equivalent, however, site A submitted consistently more pots and biopsies per month than sites B and C. Site A had, on average, 4 pots per case compared to sites B and C with 2 pots per case. This difference was demonstrated further when analysed in terms of frequency of pot numbers per case in terms of "low pot numbers" (1–3 pots) and "high pot numbers" (6–9 pots). The distribution was skewed towards low pot numbers for sites B and C whilst site A had a more even distribution causing a flattened profile of the frequency curve. The high pot number cases were sub-analysed. Site A had a total of 95 cases with high pot number (30% of total cases). Sites B and C respectively had a total of 34 and 28 cases (9 and 5% of total cases).The histological diagnostic distribution however, was equivalent across all sites despite biopsy practices varying.

Conclusion: A substantiated discrepancy exists between site A when compared to B and C. This provides scope for rationalisation and standardisation of biopsy practice.

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