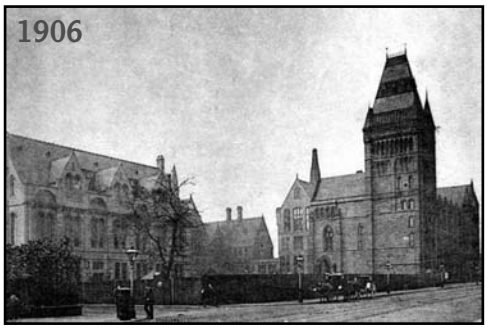
Pathological Society
of Great Britain & Ireland

Understanding disease
CENTENARY YEAR
2006

Centenary Meeting

MANCHESTER
1824

The University of Manchester



The Centenary (190th) Meeting
of the Pathological Society of Great Britain & Ireland

4–7 July 2006

To be held at the Armitage Centre, The University of Manchester

Hosted by the Division of Laboratory and Regenerative Medicine
The University of Manchester

CONTENTS

PROGRAMME ACKNOWLEDGEMENTS

Published by
The Pathological Society of Great Britain & Ireland

COVER PHOTOGRAPH
The cover photographs are reproduced with permission

Programme is typeset in Times and ITC Stone Sans

This programme was designed, produced and printed in England by
Byte & Type Limited · Birmingham · Ⓣ 0121 772 3828 · Ⓔ info@bytetype.co.uk

PROGRAMME SYNPOSIS AND TIMETABLE

(Showing times, sessions and venues)

TUESDAY 4 JULY

07.45	**Registration** *(Armitage Centre Foyer)* and Coffee *(Main Hall Part 2)*
08.30–11.30	Symposium: *Cervical Screening 2010 and Beyond* (Joint Symposium with the British Society for Clinical Cytology) *(Studio A)*
09.00–11.00	Symposium: *Structure and Function of Connective Tissues and Pathology (Conference Room)*
09.00–17.00	Slide Seminar Competition Case Viewing: *Infections of the Gastrointestinal Tract (Win a case of champagne!) (Studio B)*
10.00–10.30	Coffee (Cervical Screening) *(Main Hall Part 2)*
11.00–11.30	Coffee (Connective Tissues) *(Main Hall Part 2)*
11.30–11.45	Welcome Address: Prof AJ Freemont, Manchester *(Main Hall Part 1)*
11.45–13.00	Joint Symposium with the Japanese Society of Pathology: *Lymphoproliferative diseases and skin tumours: Differences of disease patterns between the UK and Japan (Main Hall Part 1)*
13.00–14.00	Lunch *(First Floor Balcony)*
13.00–14.00	Meet the Experts: *Gynaecological Pathology (Conference Room)*
14.00–15.00	Poster Presentations & Rounds *(Abstracts P1–P108)* and Trade Exhibition *(Main Hall Part 2)*
15.00–17.00	Oral Communications: Abstracts O1–O6 *(Conference Room)* Abstracts O7–O12 *(Studio A)*
16.00–16.30	Tea *(Main Hall Part 2)*
17.30–18.30	Public Lecture: *Pathologists Under Fire: Society Divided – Nothing New* Prof M Brazier, Manchester *(Roscoe A – Main Campus)* ***Buses will depart from the Armitage Centre at 17.00***
18.30–20.00	Welcome Reception *(The Manchester Museum)*

WEDNESDAY 5 JULY

08.15	**Registration** *(Armitage Centre Foyer)*
09.00–15.00	Slide Seminar Competition Case Viewing: *Infections of the Gastrointestinal Tract (Win a case of champagne!) (Studio B)*
09.00–10.30	Trainees Programme: *Histopathology Examinations: A Guide for Trainees and Trainers (Studio A)*
09.00–16.00	UK NEQAS: *EQA Scheme for HER2 IHC Testing (Chancellors – Flowers Lecture Theatre)*
10.00–15.30	UK NEQAS for Cellular Pathology Technique – Annual Meeting *(Conference Room)*
10.30–11.00	Coffee *(Main Hall Part 2)*
11.00–13.00	Trainees Programme: *Oral Presentations (Abstracts O13–O20) (Studio A)*
13.00–14.00	Lunch *(First Floor Balcony)*
13.00–14.00	Meet the Experts: *Skin Pathology (Conference Room)*
13.00–14.30	Liver EQA Meeting *(Studio A)*
14.00–15.00	Trade Exhibition *(Main Hall Part 2)*
15.00–17.30	Plenary Oral Session *(Abstracts PL1–PL8) (Main Hall Part 1)*
16.00–16.30	Tea *(Main Hall Part 2)*

17.30–18.15 Pathological Society's 4th Doniach Lecture: *Functional Pathology of Breast Cancer* Prof AM Neville, London and New York *(Main Hall Part 1)*

19.30 for 20.00 Centenary Society Dinner *(Manchester Town Hall)*
Buses will depart from the Armitage Centre at 19.00 and will leave the Town Hall at 23.00

THURSDAY 6 JULY

08.15 **Registration** *(Armitage Centre Foyer)*

09.00–17.00 Association of Clinical Electron Microscopists Companion Meeting *(Chancellors – Marquis Room)*

09.00–12.00 Oral Communications: Abstracts O21–O30 *(Conference Room)*
Abstracts O31–O40 *(Studio A)*

10.30–11.00 Coffee *(Main Hall Part 2)*

12.00–12.45 Pathological Society's 27th CL Oakley Lecture: *Mechanisms underlying chromosomal instability in gastrointestinal cancer* Dr H Grabsch, Leeds *(Main Hall Part 1)*

12.45–14.00 Lunch *(First Floor Balcony)*

13.00–14.00 Meet the Experts: *Breast Pathology (Conference Room)*

13.00–14.00 Molecular Pathology Group Meeting *(Studio A) — All Welcome*

14.00–15.00 Poster Presentations & Rounds *(Abstracts P109–P210)* and Trade Exhibition *(Main Hall Part 2)*

15.00–18.00 UK Renal Pathology Group Meeting, including Renal Pathology EQA and Renal Transplant EQA *(Conference Room)*

15.00–15.30 Tea *(Main Hall Part 2)*

15.30–17.00 Slide Seminar Discussion Session: *Infections of the Gastrointestinal System (Studio A)*

17.00–18.00 Pathological Society – Annual Business Meeting *(Main Hall Part 1)*

18.00–20.30 Centenary Symposium and Reception *(Main Hall Part 1)*
Symposium: *The Past, Present and Future of the Pathological Society and the Journal of Pathology,* **sponsored by John Wiley & Sons**

FRIDAY 7 JULY

08.15 **Registration** *(Armitage Centre Foyer)*

09.00–12.00 Symposium: *The Future of Pathology Practice (Conference Room)*

10.30–11.00 Coffee *(First Floor Balcony)*

SCIENTIFIC SESSIONS INFORMATION

PLENARY ORAL SESSION *(Main Hall Part 1)*

The plenary oral session, in which the 8 highest-ranked submitted oral abstracts will be presented, will be held on **Wednesday 5 July, 15.00–17.30 hrs**.

Prize

A prize for the best presentation, donated by the *Journal of Pathology* will be presented at the Society Dinner.

ORAL COMMUNICATIONS *(Conference Room and Studio A)*

Oral communication sessions will be held as follows:

Main Meeting Tuesday 4 July, 15.00–17.00
Thursday 6 July, 09.00–12.00

Note to presenters: Speakers are reminded that no communication may exceed the time allocated on the programme without the consent of the meeting, obtained through the Chairman.

POSTER VIEWING & ROUNDS *(Main Hall Part 2)*

Posters will be displayed throughout the meeting, but dedicated viewing sessions will be on **Tuesday 4 July** and **Thursday 6 July** (14.00–15.00 both days).

Poster Rounds

Chairmen will review posters during Tuesday 4 July and Thursday 6 July.

Ideally, posters should be in place by **12.00 hrs on Tuesday 4 July** and removed by **17.00 hrs on Thursday 6 July**. At least one of the contributors must be in attendance during the viewing period, as indicated in the programme synopsis.

Prizes

Prizes are awarded for the three best posters: The Sir Alastair Currie Prize, Second and Third prizes will be presented at the **Centenary Reception on Thursday 6 July**.

SYMPOSIA

Five symposia will be held.

Tuesday 4 July

08.30–11.30 *Cervical Screening: 2010 and Beyond*
Joint Symposium with the British Society for Clinical Cytology *(Studio A)*

09.00–11.00 *Structure and Function of Connective Tissues and Pathology (Conference Room)*

11.45–13.00 *Lymphoproliferative diseases and skin tumours: Differences of disease patterns between the UK and Japan*
Joint Symposium with the Japanese Society of Pathology *(Main Hall Part 1)*

Thursday 6 July

18.00–20.30 Centenary Symposium and Reception: *The Past, Present and Future of the Pathological Society and the Journal of Pathology*
Sponsored by John Wiley & Sons. *(Main Hall Part 1)*

Friday 7 July

09.00–12.30 *The Future of Pathology Practice (Main Hall Part 1)*

TRAINEES PROGRAMME

Meet the Experts *(Conference Room)*

Trainees are encouraged to bring along images of problematic cases which will be projected onto the main lecture theatre screen using a multi-headed microscope. Discussion will take place on the diagnostic points relating to such cases. The experts may also provide examples of common diagnostic problems.

Tuesday 4 July

13.00–14.00 **Gynaecological Pathology**
Experts:
Prof M Wells, University of Sheffield
Dr N Wilkinson, St James's University Hospital, Leeds
Dr GE Wilson, Manchester Royal Infirmary

Wednesday 5 July

13.00–14.00 **Skin Pathology**
Experts:
Dr SS Banerjee, Christie Hospital, Manchester
Dr N Kirkham, Royal Victoria Infirmary, Newcastle-upon-Tyne
Dr N Leonard, Royal Liverpool University Hospital

Thursday 6 July

13.00–14.00 **Breast Pathology**
Experts:
Dr WF Knox, South Manchester University Hospital, Wythenshawe, Manchester
Dr SE Pinder, Addenbrooke's Hospital, Cambridge
Prof RA Walker, Leicester Royal Infirmary

Other Trainee Events *(Studio A)*

Wednesday 5 July

09.00–10.30 Histopathology Examinations: A Guide for Trainees and Trainers
11.00–13.00 Trainee Oral Presentations

SPECIAL INTEREST GROUP *(Studio A)*

Thursday 6 July

13.00–14.00 **Molecular Pathology Group** — *All Welcome*

SLIDE SEMINAR COMPETITION – *Infections of the Gastrointestinal System (Studio B)*

There will be a slide seminar competition and digital images of all cases will be available for preview during the course of the meeting. The images will also become available on the Society's website in due course.

The images will be available for viewing in Manchester on Tuesday 4 July, 09.00–17.00 and on Wednesday 5 July, 09.00–15.00.

Prize

A case of champagne will be awarded at the Society Dinner on Wednesday 5 July.

Discussion Session

Thursday 6 July, 15.30–17.00 *(Studio A)*

PUBLIC LECTURE *(Roscoe A – Main Campus)*

Tuesday 4 July, 17.30–18.30

The Pathological Society's 2nd Public Lecture entitled: *Pathologists Under Fire: Society Divided – Nothing New* will be given by Professor M Brazier, School of Law, The University of Manchester.

SOCIETY LECTURES *(Main Hall Part 1)*

Wednesday 5 July, 17.30–18.15
The Pathological Society of Great Britain & Ireland's 4th Doniach Lecture entitled: *Functional Pathology of Breast Cancer* will be given by Prof A Munro Neville, Ludwig Institute for Cancer Research, London and New York.

Thursday 6 July, 12.00–12.45
The Pathological Society of Great Britain & Ireland's 27th CL Oakley Lecture entitled: *Mechanisms underlying chromosomal instability in gastrointestinal cancer* will be given by Dr H Grabsch, Leeds Institute of Molecular Medicine, University of Leeds.

COMPANION MEETINGS

Wednesday 5 July

09.00–16.00 UK NEQAS: EQA Scheme for HER2 IHC Testing *(Chancellors – Flowers Lecture Theatre)*

10.00–15.30 UK NEQAS for Cellular Pathology Technique – Annual Meeting *(Conference Room)*

13.00–14.30 Liver EQA *(Studio A)*

Thursday 6 July

09.00–17.00 Association of Clinical Electron Microscopists *(Chancellors – Marquis Room)*

15.00–18.00 UK Renal Pathology Group Meeting, including Renal Pathology EQA and Renal Transplant EQA *(Conference Room)*

TRADE EXHIBITION *(Main Hall Part 2)*

Delegates are encouraged to visit the **Trade Exhibition** and are requested to support the companies represented there.

Continuing Professional Development (CPD)

This meeting has been approved by the following bodies for the purposes of Continuing Professional Development. Credits can be accrued as follows:

Royal College of Pathologists
For each full day: 7 points
For each half day: 3 points

Institute of Biomedical Sciences (Meeting Code: GM094N06)
For each full day: 8 credits

Certificates
Delegates who are eligible for CPD points should collect their certificates at the Registration Desk before leaving the Meeting.

Enquiries before the meeting
Pathological Society of Great Britain & Ireland
2 Carlton House Terrace, London SW1Y 5AF
Telephone +44 (0)20 7976 1260
Fax +44 (0)20 7976 1267
Email admin@pathsoc.org.uk

General Arrangements

REGISTRATION

Registration is via our website: **www.pathsoc.org.uk**
Follow the *Centenary Meeting Online Registration* link on our home page.

The registration system will issue an automated email acknowledgement. All tickets will be issued on arrival at the Delegate Reception Desk in Manchester.

FEES

Fees include all refreshments and lunch.

- **Society Members:**
  (excluding Trainees, Senior and Honorary Members – see concessions below)

  Up to and including 2 June 2006
  £220 for the whole meeting, **or** £90 per day (or part day)

  After 2 June 2006
  £320 for the whole meeting, **or** £110 per day (or part day)

- **Non-Members:**
  (for concessions see below)

  Up to and including 2 June 2006
  £270 for the whole meeting, **or** £110 per day (or part day)

  After 2 June 2006
  £370 for the whole meeting, or £130.00 per day or part day

- **Concessions:**

  £15 for Society Members per day (or part day)
  £25 for Non-members per day (or part day)

  Qualifying Categories:
  - Honorary & Senior Members of the Society
  - PhD Students, Junior Technicians, Residents*, Trainees*, Biomedical Scientists and Undergraduates*

  * *To qualify for this reduced fee, delegates must submit an identification document signed by your Head of Training, including National Training Numbers where applicable. Trainee Society Members are NOT required to provide this.*
  Please send your identification document by post or via email to: **julie@pathsoc.org.uk**

- **Society Dinner:**

  £50 per ticket

ADVANCE REGISTRATION – CLOSING DATE

Advance registration will close on **Friday 23 June**.
After this deadline registration will only be accepted on-site in Manchester.

CANCELLATIONS

Please note that the Society is **unable** to refund registration fees for cancellations **received after Friday 23 June**.

DELEGATE ENROLMENT (AT THE MEETING)

Enrolment at the Delegate Reception Desk will take place in the **Armitage Centre Foyer** as follows:

Tuesday 4 July	**from 07.45**
Wednesday 5 July	**from 08.15**
Thursday 6 July	**from 08.15**
Friday 7 July	**from 08.15**

General Arrangements *continued*

PRESENTATION CHECKING AND PREVIEW
This will be available in Studio B.

ORAL PRESENTATIONS AND LECTURES
Presentations must be in Microsoft PowerPoint (PC or Mac).

Presentations should be submitted **in advance** of the meeting to arrive **no later than Friday 30 June**.

They may be sent by the following methods:

- emailed to Tony Pendlebury via: **tony@pendleburyav.co.uk**
- sent on a CD by post to the following address:
  Tony Pendlebury, Pendlebury Audio Visual
  68 Border Brook Lane, Worsley, Manchester M28 1XJ

Please **label** the files or CDs with:

- Date and time of your lecture
- Lecture theatre
- Your name

IMPORTANT: Please bring another copy of your presentation with you to the meeting.

MESSAGES
During the Meeting, messages for delegates may be left on **telephone: 0161 224 0404**.

There will also be a message board located beside the Delegate Reception Desk.

REFRESHMENTS
Coffee and tea will be available **all day** in **Main Hall Part 2**.

BADGES
Delegates are requested to wear their badges at **all** times.

SMOKING
Smoking is prohibited at all meetings and social events except in the designated areas .

DISCLAIMER
The Pathological Society of Great Britain & Ireland cannot be held responsible for any injury or loss sustained during the Meeting.

SOCIAL ACTIVITIES
Tuesday 4 July, 18.30–20.00 *(The Manchester Museum)*
Welcome Reception
Tickets are free – to reserve your ticket please tick the relevant box when registering .
Buses will depart from The Armitage Centre at 17.00 for the Public Lecture held immediately prior to the Reception, returning from The Manchester Museum at 20.30.

Wednesday 5 July, 19.30 for 20.00 *(Manchester Town Hall)*
Centenary Society Dinner
Tickets are £50 each. To reserve your ticket please tick the relevant box when registering .
Buses will depart from the Armitage Centre at 19.00 returning from Manchester Town Hall at 23.00.

Thursday 6 July, 18.30–19.30 *(The Armitage Centre, Main Hall Part 1)*
Centenary Reception
Tickets are free – to reserve your ticket please tick the relevant box when registering .

Local Places of Interest
Please refer to the Internet for information: **www.cam.ac.uk/cambarea/tourist.html**

FUTURE MEETINGS

2007	3–5 January	University College, London
	3–6 July	**Glasgow Pathology 2007**: 4th Joint Meeting of the Pathological Society and the British Division of the IAP
2008	7–9 January	Oxford
	1–4 July	Leeds
2009	7–9 January	GKT, London
	2–5 June (tbc)	**Cardiff Pathology 2009**: 5th Joint Meeting of the Pathological Society and the British Division of the IAP
2010	January	Imperial College, London
	July	St Andrews

07.45 **Armitage Centre Foyer**
REGISTRATION

08.30–11.30 **Studio A**
SYMPOSIUM: *Cervical Screening: 2010 and Beyond*
(Joint Symposium with the British Society for Clinical Cytology)
Chair: Dr M Desai, The University of Manchester

08.30–09.20 **[S1]** *New Strategies in Cervical Screening*
Prof HC Kitchener, The University of Manchester
Academic Unit of Obstetrics and Gynaecology

09.20–10.10 **[S2]** *Molecular markers in cytology – scope for the future*
Prof JJ O'Leary, Trinity College, Dublin

10.10–10.40 COFFEE *(Armitage Centre – Main Hall Part 2)*

10.40–11.30 *HPV Vaccine – state of the art*
Dr M Stanley, University of Cambridge

09.00–11.00 **Conference Room**
SYMPOSIUM: *Structure and Function of Connective Tissues and Pathology*
Chair: Prof AJ Freemont, The University of Manchester
Dr JA Hoyland, The University of Manchester

09.00–09.30 **[S3]** *Our shape comes in extracellular modules. Their role in biology and pathology*
Prof JE Scott, The University of Manchester

09.30–10.00 *Evolution of collagen*
Dr R Boot-Handford, The University of Manchester

10.00–10.30 **[S4]** *The connective tissue stem cell: lineage, plasticity and function*
Dr JA Hoyland, The University of Manchester

10.30–11.00 *Tumours/Non-tumour pathology*
Prof AJ Freemont, The University of Manchester

11.00–11.30 COFFEE *(Armitage Centre – Main Hall Part 2)*

09.00–17.00 **Studio B**
SLIDE SEMINAR COMPETITION CASE VIEWING: *Infections of the Gastrointestinal Tract* (Win a Case of Champagne!)

11.30–11.45 **Main Hall Part 1**
OPENING ADDRESS: Prof AJ Freemont, The University of Manchester

11.45–13.00 **Main Hall Part 1**
JOINT SYMPOSIUM – with the Japanese Society of Pathology
Lymphoproliferative diseases and skin tumours: Differences of disease patterns between the UK and Japan
Chair: Prof AJ Freemont, The University of Manchester
Prof R Osamura, Tokai University, School of Medicine

11.45–12.15 *Differences of disease patterns in skin tumours between the UK and J apan*
11.45–12.00 Dr N Kirkham, Royal Victoria Infirmary, Newcastle-upon-Tyne
12.00–12.15 **[S5]** Dr H Sasano, Tohoku University, Japan

11.45–13.00 **Main Hall Part 1** *Continued*

12.15–12.45 *Incidence and environmental factors in the aetiology of lymphomas*
12.15–12.30 Dr N Rooney, Southmead Hospital, Bristol
12.30–12.45 **[S6]** *Pyothorax-associated Lymphoma-A Distincive Type of Lymphoma which is common in Japan but rare in Western countries*
Prof K Aozasa, Osaka University, Japan

12.45–13.00 Discussion

13.00–14.00 LUNCH *(First Floor Balcony)*

13.00–14.00 **Conference Room**
MEET THE EXPERTS: *Gynaecological Pathology*

13.00–15.00 **Main Hall Part 2**
TRADE EXHIBITION

14.00–15.00 **Main Hall Part 2**
POSTER PRESENTATIONS & ROUNDS

CATEGORIES
Cellular/Molecular **[P1-P19]**
Gastrointestinal **[P21-P52]** *(Note: P20 withdrawn)*
Gynaecological **[P53-P69]**
Hepatobiliary/Pancreas **[P70-P78]**
Neuropathology/Ophthalmic **[P79-P83]**
Osteoarticular/Soft Tissue **[P84-P100]**
Technical Advances **[P101-P108]**

15.00–16.00 **Conference Room**
ORAL COMMUNICATIONS
Categories: Gastrointestinal
Chair: Dr RFT McMahon, The University of Manchester
Prof CS Herrington, University of St Andrews

15.00 **[O1]** *Biopsy technique and outcome in long segment Barrett's oesophagus*
JE Abela, **{P}** JJ Going, M McKernan, RC Stuart

15.15 **[O2]** *The Clonal Origin of Human tumours: is Familial Adenomatous Polyposis (FAP) different?*
{P} SJ Leedham, LC Maia, O Seiber, SL Preston, SAC McDonald, IPA Tomlinson, MR Novelli, NA Wright

15.30 **[O3]** *Chromosome 20q Amplification in Colorectal Adenoma to Carcinoma progression*
{P} B Carvalho, C Postma, S Mongera, B Ylstra

15.45 **[O4]** *Characterization of Side Populations of Human Gastrointestinal Cell Lines*
{P} J Burkert, WR Otto, D Davies, K Allen, J Paris, NA Wright

16.00–16.30 TEA *(Armitage Centre – Main Hall Part 2)*

16.30–17.00 **Conference Room**

ORAL COMMUNICATIONS

Categories: Gynaecological; Hepatobiliary/Pancreas

Chair: Dr RFT McMahon, The University of Manchester
Prof CS Herrington, University of St Andrews

16.30 **[O5]** *An audit of management of inadequate smears*
{P} I Babarinsa, J Mathew, A St John, A Oladipo

16.45 **[O6]** *The Heat Sink Effect may cause inadequate tissue ablation: a histological evaluation of microwave, radiofrequency and cryoablation in the rat liver*
{P} N Bhardwaj, A Strickland, L Atanesyan, F Ahmad, K West, D Lloyd

15.00–16.00 **Studio A**

ORAL COMMUNICATIONS

Categories: Cellular/Molecular Pathology; Technical Advances

Chair: Dr RJ Byers, The University of Manchester
Prof M Ilyas, Queen's Medical Centre, Nottingham

15.00 **[O7]** *Functional analysis of disease associated mutations in SEPT9*
{P} PA Hall, S McDade, P Hyland, L Chatham, SEH Russell

15.15 **[O8]** *DC-SIGN association with the Th2 environment of lepromatous leprosy lesions: cause or effect?*
{P} EJ Soilleux, EN Sarno, MO Hernandez, E Moseley, J Horsley, UG Lopes, MJ Goddard, SL Vowler, N Coleman, RJ Shattock, EP Sampaio

15.30 **[O9]** *Laser capture microdissection and bioinformatics profiling to identify tumour vascular markers*
BD Tarlow, LA Strickland, Z Modrusan, TD Wu, **{P}** HK Koeppen

15.45 **[O10]** *Measurement of Lymphoma Gene Signatures by Real-time PCR in Globally Amplified PolyA cDNA from Paraffin Embedded Tissue*
{P} J Goodchild, E Sakhinia, C Glennie, L Menasce, JA Radford, JA Hoyland, RJ Byers

16.00–16.30 TEA *(Armitage Centre – Main Hall Part 2)*

16.30–17.00 **Studio A**

ORAL COMMUNICATIONS

Categories: Osteoarticular/Soft Tissue

Chair: Dr RJ Byers, The University of Manchester
Prof M Ilyas, Queen's Medical Centre, Nottingham

16.30 **[O11]** *Cytokine Involvement in the Pathogenesis of Disc Degeneration: IL-1 or TNF Alpha?*
{P} CL Le Maitre, AJ Freemont, JA Hoyland

16.45 **[O12]** *Accelerated Cellular Senescence in Human Intervertebral Disc Degeneration*
{P} CL Le Maitre, AJ Freemont, JA Hoyland

17.30–18.30 **Roscoe A – Main Campus**

PUBLIC LECTURE: *Pathologists Under Fire: Society Divided – Nothing New*

Prof M Brazier, School of Law, The University of Manchester

Chair: Prof Sir NA Wright, St Bartholomew's and the Royal London Hospital School of Medicine and Dentistry

(Buses will depart from the Armitage Centre at 17.00)

18.30–20.00 **The Manchester Museum**

WELCOME RECEPTION

Welcome Address by Prof Dame Nancy Rothwell, FRS, MRC Prof of Physiology and Vice President for Research, The University of Manchester

08.15 **Armitage Centre Foyer**
REGISTRATION

09.00–15.00 **Studio B**
SLIDE SEMINAR COMPETITION CASE VIEWING
Infections of the Gastrointestinal Tract (WIN A CASE OF CHAMPAGNE!)

09.00–10.30 **Studio A**
TRAINEES PROGRAMME
Histopathology Examinations: A Guide for Trainees and Trainers
Chair: Dr P Hasleton, The University of Manchester
Dr PJ Gallagher, University of Southampton

09.00–09.10 Introduction
Prof AJ Freemont, The University of Manchester

09.10–09.30 **[S7]** *OSPE Assessment and Year 1 Run-through Training*
Dr AC Bateman, Southampton University Hospitals NHS Trust

09.30–09.50 **[S8]** *MRCPath in Histopathology Part 1: Why the Change*
Dr EW Benbow, The University of Manchester

09.50–10.10 **[S9]** *Part 2 MRCPath Examination*
Dr RFT McMahon, The University of Manchester

10.10–10.30 Questions & Answers

10.30–11.00 COFFEE *(ARMITAGE CENTRE – MAIN HALL PART 2)*

11.00–13.00 **Studio A**
TRAINEES PROGRAMME
ORAL COMUNICATIONS
Chair: Co-Chairs of the Pathological Society Trainees Sub-Committee:
Dr M Deheragoda, University College London,
Dr KE Robertson, University of Dundee

11.00 **[O13]** *Diagnostic Accuracy of Conventional Smears and ThinPrep Samples in Cervical Screening*
{P} B Shah, D Rana, J Marshall, M Desai

11.15 **[O14]** *Stromal Nodules and Vessel Wall Thickening in TURP Specimens are Associated with Failure of Alpha-Blocker Treatment*
{P} M-A Tran-Dang, RD Smith, B Khoubehi, A Patel, MM Walker

11.30 **[O15]** *Hadfield's Procedure – Pathology Review and Appropriateness of Surgical Treatment*
{P} A Naveed, B Benatar, A Benatar, S Ellenbogen

11.45 **[O16]** *The area of involved lymph nodes in colorectal carcinoma. Lessons for the radiologist and pathologist*
{P} ML Baxandall, F Lewis, G Casali, P Guillou, H Thorpe, J Walker, D Jayne, AMH Smith, RM Heath, JM Brown, P Quirke

12.00 **[O17]** *Cervical Stromal Sarcoma: A Rare Neoplasm*
{P} P Jaiswal, A Samarrai

11.00–13.00 **Studio A** *Continued*

12.15 **[O18]** *Application of a Modified Histological Activity Index in Primary Biliary Cirrhosis*
{P} A Awasthi, A Smith, TW Warnes, RFT McMahon

12.30 **[O19]** *A Comparison of Cervical Screening Histories in Women Developing Intraepithelial Neoplasia and Invasive Carcinoma*
{P} I Morrison, A Deery

12.45 **[O20]** *Development of an in vitro Model of Prostate Carcinoma Metastasis to Bone*
{P} J Roulson, MD Brown, TD Allen, JH Shanks, NW Clarke

13.00–14.00 LUNCH *(First Floor Balcony)*

13.00–14.00 **Conference Room**
MEET THE EXPERTS: *Skin Pathology*

13.00–14.30 **Studio A**
LIVER EQA MEETING

14.00–15.00 **Main Hall Part 2**
TRADE EXHIBITION

15.00–16.00 **Main Hall Part 1**
PLENARY ORAL SESSION
Chair: Prof M Pignatelli, University of Bristol,
Prof AJ Freemont, The University of Manchester

15.00 **[PL1]** *Global Histone Modifications in Breast Cancer and their Prognostic Significance*
{P} S El Sheikh, EA Rakha, EC Paish, DM Heery, AR Green, IO Ellis

15.15 **[PL2]** *Microarray analysis of mesenchymal tumours identifies brachyury, a novel biomarker for chordomas*
{P} S Vujovic, R Tiraboco, S Henderson, C Boschoff, AM Flanagan

15.30 **[PL3]** *Bone Marrow Contributes to Functionally Active Cells in Tumour Stroma*
{P} NC Direkze, R Jeffery, K Hodivala-Dilke, F Burke, FT Hunt, R Poulsom, F Balkwill, NA Wright, MR Alison

15.45 **[PL4]** *Array comparative genomic hybridization profiling of early invasion in colorectal adenomas*
{P} AJ Watkins, G Poulogiannis, K Daly, N Johnson, M-Q Du, AEK Ibrahim, AH Wyllie, MJ Arends

16.00–16.30 TEA *(Armitage Centre – Main Hall Part 2)*

16.30–17.30 **Main Hall Part 1**
PLENARY ORAL SESSION
Chair: Prof M Pignatelli, University of Bristol,
Prof AJ Freemont, The University of Manchester

16.30 **[PL5]** *Recurrent ovarian cancer cDNA signatures*
{P} A Laios, SA O'Toole, BL Sheppard, N Gleeson, T D'Arcy, EPJ McGuinness, M Ring, P Smyth, O Sheils, JJ O'Leary

16.45 **[PL6]** *Limited Expression of the Coxsackie and Adenovirus Receptor in Pancreatic Cancer May Reduce Suitability of Adenoviral Gene Therapy*
{P} CS Verbeke, S Hamdan, J Booth, HS Pandha, GE Blair

17.00 **[PL7]** *Novel FISH probes designed to detect IGK-MYC and IGL-MYC rearrangements in B-cell lineage malignancy reveal a new breakpoint cluster region designated BVR2*
{P} A Dogan, RR Einerson, ME Law, PJ Kurtin, ED Remstein

17.15 **[PL8]** *Matrix Metalloproteinase production is inhibited in degenerate intervertebral discs by IL-1Ra gene transfer*
{P} CL Le Maitre, AJ Freemont, JA Hoyland

17.30–18.15 **Main Hall Part 1**
PATHOLOGICAL SOCIETY'S 4TH DONIACH LECTURE
[S10] *Functional Pathology of Breast Cancer*
Prof AM Neville, London and New York
Chair: Prof Sir NA Wright, St Bartholomew's and The Royal London Hospital School of Medicine and Dentistry

19.30 **Manchester Town Hall**
for 20.00 **CENTENARY SOCIETY DINNER**
(Buses will depart from the Armitage Centre at 19.00 and will return from the Town Hall at 23.00)

09.00–16.30 **Chancellors – Flowers Lecture Theatre** *(Open to all delegates)*
UK NEQAS: EQA SCHEME FOR HER2 IHC TESTING
Special Slides Workshop/Seminar
on EQA, Interpretation & Management of HER2 IHC Results
Sponsored by Roche UK

09.00–13.00 *HER2 IHC Slides Viewing Session: Evaluation of typical case results by participants using an anonymised questionnaire*

10.30–11.00 COFFEE *(Armitage Centre – Main Hall Part 2)*

13.00–14.00 LUNCH *(Armitage Centre – First Floor Balcony)*

14.00–16.00 SEMINAR & OPEN DISCUSSION SESSION

- 14.00–14.45 *Herceptin Treatment of Breast Cancer: Overview*
  Dr I Greenfield, Senior Scientific Advisor, Roche UK
- 14.45–15.10 *UK NEQAS: Scheme for Assessment of Technical Aspects of HER Testing*
  Dr M Ibrahim, Scheme Manager, UK NEQAS
- 15.10–15.30 *Clinical Management of HER2 Testing and Interpretation of the Results*
  Prof B Jasani, Breast Cancer Module Leader, UKNEQAS
- 15.30–16.00 Feedback from Slides and Open Discussion

16.00–16.30 TEA *(Armitage Centre – Main Hall Part 2)*

10.00–15.30 **Conference Room**
UK NEQAS FOR CELLULAR PATHOLOGY TECHNIQUE ANNUAL MEETING

10.00–10.20 Introduction and Annual Report
D Evans, Scheme Organiser

10.20–10.40 *Rapid Biopsy Processing – Leaner and Fitter?*
F Sim, Lincoln County Hospital

10.40-11.00 *Xylene-Free Processing – the Newcastle Experience*
D Evans, Royal Victoria Infirmary, Newcastle-upon-Tyne

11.00–11.30 COFFEE *(Armitage Centre – Main Hall Part 2)*

11.30–12.00 *The HTA and its Implications*
I Sturdgess, Bedford Hospital NHS Trust

12.00–12.30 *From the Cut-up Room to the Reporting Room – the Way Forward for Biomedical Scientists?*
S Dicken, Walsall Manor Hospital

12.30–13.30 LUNCH *(First Floor Balcony)*

13.30–13.50 *The Renal Biopsy Programme*
R Hughes, Nottingham City Hospital

13.50–14.10 *Feedback via the Website*
G Thompson and J Elsam, Nottingham City Hospital & HTEQA Services Ltd

14.10–14.40 *The H&E from a Quality Viewpoint*
F Tajbhai, St Helier Hospital, Carshalton

14.40–15.30 Workshop – *Revisiting the Assessment Criteria: Your opportunity to help redesign H&E assessment*

08.15 **Armitage Centre Foyer**
REGISTRATION

09.00–10.30 **Conference Room**
ORAL COMMUNICATIONS
Categories: Breast
Chair: Dr JH Shanks, Manchester
Prof LB Jones, St Bartholomew's Hospital, London

09.00 **[O21]** *Analysis of MMP Single Nucleotide Polymorphisms in relation to breast cancer progression*
{P} S Hughes, D Holliday, J Shaw, S Duffy, I Hart, L Jones

09.15 **[O22]** *Breast Cancer Profiling: A Minimised Panel of Predictive Markers for Survival*
{P} N Thompson, S Leigh-Bramwell, DG Powe, G Ball, E Rakha, J Williams, EC Paish, IO Ellis

09.30 **[O23]** *EGFR amplification and lack of activating mutations in metaplastic breast carcinomas*
{P} JS Reis Filho, C Pinheiro, MBK Lambros, F Milanezi, S Carvalho, K Savage, PT Simpson, C Jones, S Swift, A Mackay, RM Reis, JL Hornick, EM Pereira, F Baltazar, CDM Fletcher, A Ashworth, SR Lakhani, FC Schmitt

09.45 **[O24]** *Comprehensive molecular genetic analysis reveals FGFR1 as a potential therapeutic target for lobular breast carcinomas*
{P} JS Reis Filho, PT Simpson, N Turner, MBK Lambros, C Jones, A Mackay, A Grigoriadis, D Sarrio, K Savage, T Dexter, M Iravani, K Fenwick, B Weber, D Hardisson, FC Schmitt, J Palacios, SR Lakhani, A Ashworth

10.00 **[O25]** *BRCA1 gene promoter methylation in metaplastic breast carcinomas*
{P} JS Reis Filho, N Turner, FC Schmitt, A Tutt, A Ashworth

10.15 **[O26]** *Cyclin D1 protein overexpression and gene amplification in breast carcinomas: an immunohistochemical and CISH analysis*
{P} JS Reis Filho, K Savage, MBK Lambros, M James, D Steele, RL Jones, M Dowsett

10.30–11.00 COFFEE *(Armitage Centre – Main Hall Part 2)*

11.00–12.00 **Conference Room**
ORAL COMMUNICATIONS
Categories: Genitourinary/Renal
Chair: Dr JH Shanks, Manchester
Prof LB Jones, St Bartholomew's Hospital, London

11.00 **[O27]** *Bone Marrow Contributes to Podocyte Regeneration and Amelioration of Renal Disease in Alport's Syndrome*
{P} E I Prodromidi, R Poulsom, R Jeffery, C A Roufosse, T Hunt, C D Pusey, HT Cook

11.15 **[O28]** *TOP2A Amplification in Prostate Cancer is associated with HER-2 Amplification, Androgen Resistance & Decreased Survival*
{P} AJ Murphy, CA Hughes, C Barrett, H Magee, B Loftus, J O'Leary, O Sheils

11.30 **[O29]** *Urine Cytology Screening for Polyoma Virus Nephropathy following Renal Transplantation: Review of a Working Service*
TP Thamboo, K Jeffery, P Friend, **{P}** ISD Roberts

11.45 **[O30]** *Assessment of the Cardiff nephrectomy cut-up protocol with total block ing of the renal sinus: Impact on Tumour Staging and Practical Issues*
{P} EJ Soilleux, ISD Roberts

09.00–10.30 **Studio A**
ORAL COMMUNICATIONS
Categories: Lymphoreticular; Skin; Head & Neck; Autopsy & Forensic; Neonatal/Paediatric
Chair: Dr EW Benbow, The University of Manchester
Dr S Deen, Queen's Medical Centre, Nottingham

09.00 **[O31]** *YY1 Expression Predicts Survival in Follicular and Diffuse Large B-cell Lymphoma*
E Sakhinia, C Glennie, JA Hoyland, L Menasce, JA Radford, **{P}** RJ Byers

09.15 **[O32]** *Different Expression Pattern of Vascular Endothelial Growth Factor-A (VEGF-A) in Diffuse Large B-Cell Lymphoma (DLBCL) and Hodgkin's Lymphoma (HL)*
{P} L A Sanders, C Burton, S Chalasani, G Frantz, A Jack, H Koeppen

09.30 **[O33]** *A Novel Picropolychrome Technique for the Selective Staining of Basement Membranes*
{P} K Asaad, DT Sharpe, S Huq, S Stevenson, A Adekunle, IL Naylor

09.45 **[O34]** *Molecular genetic analysis suggests a developmental origin for malignant myoepithelial carcinomas of the salivary gland from pleomorphic adenomas*
{P} MB Lambros, D Tan, E Arriola, K Fenwick, N Tamber, A Mackay, K Savage, A Ashworth, S Di Palma, JS Reis Filho

10.00 **[O35]** *Implementation of the 2005 Coroner's Rules Amendments – A survey of practice in England and Wales*
{P} RJ Delaney, ISD Roberts

10.15 **[O36]** *Immunohistochemical Nuclear Positivity for WT1 in Acute Myeloid Leukaemia*
{P} M Al-Adnani, S Williams, J Anderson, M Ashworth, M Malone, NJ Sebire

10.30–11.00 COFFEE *(Armitage Centre – Main Hall Part 2)*

11.00–12.00 **Studio A**
ORAL COMMUNICATIONS
Categories: Education & Audit
Chair: Dr EW Benbow, The University of Manchester
Dr S Deen, Queen's Medical Centre, Nottingham

11.00 **[O37]** *National Histopathology Training Schools (NHTS) – Recruitment Experiences, Issues and Challenges!*
S Cossins, **{P}** A McGregor, K West, J Appleyard, D Bailey, J Brinklow, T Collins, PJ Gallagher, C Gore, NR Griffin, S Hill, R Liebmann, M Young

11.15 **[O38]** *Routine examination of multiple levels from gastro-intestinal biopsies: a resource management approach*
C Chambers, A McGregor, **{P}** KP West

11.30 **[O39]** *Positive Predictive Value of Cervical Smears Reported as Mild Dyskaryosis*
{P} B Shah, J Marshall, D Rana, M Desai

11.45 **[O40]** *Direct Observation of Practical Skills in Early Histopathology Training: A Pilot Study of Competency Assessment*
{P} KP West

12.00–12.45 **Main Hall Part 1**
PATHOLOGICAL SOCIETY'S 27TH CL OAKLEY LECTURE
[S11] *Mechanisms underlying chromosomal instability in gastrointestinal cancer*
Dr H Grabsch, University of Leeds
Chair: Prof M Pignatelli, University of Bristol

12.45–14.00 LUNCH *(First Floor Balcony)*

13.00–14.00 **Conference Room**
MEET THE EXPERTS: *Breast Pathology*

13.00–14.00 **Studio A**
SPECIAL INTEREST GROUP
Molecular Pathology Group *— All Welcome*
*Please see our website (www.pathsoc.org.uk) and click **Molecular Pathology** in the main menu for further information.*
Chair: Dr RJ Byers, The University of Manchester
Provisional Agenda:
1 Welcome, Prof PA Hall, General Secretary, Pathological Society
2 Background
3 Purpose of the Molecular Pathology Group
4 Membership
5 Meeting frequency
6 Workshops
7 Any other business

14.00–15.00 **Main Hall Part 2**
TRADE EXHIBITION

14.00–15.00 **Main Hall Part 2**
POSTER PRESENTATIONS & ROUNDS
CATEGORIES
Autopsy & Forensic **[P109–P115]**
Breast **[P116–P141]**
Cardiovascular/Pulmonary **[P142–P146]**
Education & Audit **[P148–P163]** *(Note: P147 withdrawn)*
Endocrine **[P164–P168]**
Experimental Tumour Pathology **[P169–P171]**
Genitourinary/Renal **[P172–P193]**
Head & Neck **[P194–P197]**
Lymphoreticular **[P198–P204]**
Neonatal/Paediatric **[P205–P208]**
Skin **[P209–P210]**

15.00–18.00 **Conference Room**
UK RENAL PATHOLOGY GROUP MEETING
including Renal Pathology EQA and Renal Transplant EQA

15.00–15.30 TEA *(Armitage Centre – Main Hall Part 2)*

15.30–17.00 **Studio A**
SLIDE SEMINAR DISCUSSION SESSION
Infections of the Gastrointestinal System
Contributors:
Dr EW Benbow, Manchester Royal Infirmary
Dr A Curry, Manchester Royal Infirmary
Dr H Denley, Manchester Royal Infirmary
Dr RFT McMahon, Manchester Royal Infirmary
Dr K Skordilis, Manchester Royal Infirmary

17.00–18.00 **Main Hall Part 1**
PATHOLOGICAL SOCIETY – ANNUAL BUSINESS MEETING

18.00–19.30 **Main Hall Part 1**
CENTENARY SYMPOSIUM
The Past, Present and Future of the Pathological Society and the Journal of Pathology
Sponsored by John Wiley & Sons
Chair: Prof PA Hall, Queen's University Belfast

18.00–18.20 *Bequest, Request and Quest: An overview of the Past of the Pathological Society*
Prof F Walker, Edinburgh, Ex-Chairman & General Secretary

18.20–18.40 *The Pathological Society: Present*
Prof Sir NA Wright, London, President

18.40–19.00 *The Pathological Society: Future*
[S12] Prof DA Levison, Dundee, President-Elect

19.00–19.20 *The Journal of Pathology (and Bacteriology): Past, Present and Future*
Prof CS Herrington, St Andrews, Editor-in-Chief, Journal of Pathology

19.30–20.30 **Main Hall Part 1**
CENTENARY RECEPTION

09.40–17.00 **Chancellors – Marquis Room**
ASSOCIATION OF CLINICAL ELECTRON MICROSCOPISTS COMPANION MEETING

09.40–10.30 *Human DNA Viruses: Life cycle, Negative stained, and Thin section*
Mr B Wagner, Northern General Hospital, Sheffield
Mr B Dowsett, Porton Down, Salisbury
Mr B Cooley, Veterinary Laboratories Agency, Addlestone
Chair: Dr R Fleck, National Institute for Biological Standards and Control, London

10.30–11.00 COFFEE *(Armitage Centre – Main Hall Part 2)*

11.00–11.50 *Human RNA Viruses: Life cycle, Negative stained, and Thin section*
Mr B Wagner, Northern General Hospital, Sheffield
Mr B Dowsett, Porton Down, Salisbury
Mr B Cooley, Veterinary Laboratories Agency, Addlestone
Chair: Dr R Fleck, National Institute for Biological Standards and Control, London

12.00–12.50 *Use of EM in bone disease – diagnosis and therap y*
Dr J Cassella, Staffordshire University, Stoke on Trent
Chair: Mr B Wagner, Northern General Hospital, Sheffield

13.00–14.00 LUNCH *(Armitage Centre – First Floor Balcony)*

14.00–14.50 *Ultrastructure of erythroblasts in health and disease*
Prof S Wickramasinghe, St Mary's Hospital, London
Chair: Dr J Moss, Charing Cross Hospital, London

14.55–15.30 *Case Presentations*
Mr I Scott, Queens Medical Centre, Nottingham
Chair: Mrs N Costin-Kelly, Children's Hospital, Birmingham

15.30–16.00 TEA *(Armitage Centre – Main Hall Part 2)*

16.00–17.00 ASSOCIATION OF CLINICAL ELECTRON MICROSCOPISTS – AGM
(Open to ACEM members & non-members)
Chair: Dr T Ryder, Charing Cross Hospital, London

08.15 **Armitage Centre Foyer**
REGISTRATION

09.00–12.30 **Conference Room**
SYMPOSIUM
The Future of Pathology Practice
Chair: Prof DA Levison, University of Dundee

09.00–09.30 *Future of pathology practice in the NHS*
Dr JP Sheffield, United Bristol Healthcare Trust

09.30–10.00 **[S13]** *The Future of Pathology – Tissue Banking*
Dr C Womack, AstraZeneca

10.00–10.30 *Commerce – interactions with pathologists*

10.00–10.15 *Challenge of image-based atlases of pathology*
Dr A Warford, Atlas of Gene Expression Project, Wellcome Trust Sanger Institute

10.15–10.30 *The Advances in Digital Imaging and Telepathology*
Mr S Denham, Nikon UK Ltd

10.30–11.00 COFFEE *(Armitage Centre – Main Hall Part 2)*

11.00–11.30 **[S14]** *Molecular Pathology*
Dr FA Lewis, Leeds Teaching Hospitals NHS Trust

11.30–12.00 *Driving change*
Prof AJ Freemont, The University of Manchester

12.00–12.30 Final discussion

12.30 LUNCH *(a packed lunch will be available)*

The Pathological Society of Great Britain & Ireland

wishes to acknowledge

the support of the following companies

participating in the Trade Exhibition

APERIO TECHNOLOGIES, USA

APPLIED IMAGING INTERNATIONAL LTD

ECACC (EUROPEAN COLLECTION OF CELL CULTURES)

HAMAMATSU PHOTONICS UK LTD

LAUNCH DIAGNOSTICS LTD

NIKON UK LTD

SLIDEPATH

providing the slide digitising service
for the "Meet the Experts" session

VOICEPOWER

JOHN WILEY & SONS LTD

WISEPRESS ON-LINE BOOKSHOP

(as at the time of going to press)

Abstracts

Plenary

PL1

Global Histone Modifications In Breast Cancer And Their Prognostic Significance

{P} S El Sheikh, EA Rakha, EC Paish, DM Heery, AR Green, IO Ellis
University of Nottingham, Nottingham, Nottinghamshire, United Kingdom

Global changes in histone modification show association with patient outcome in prostate cancer. However, the clinical significance of these modifications in breast cancer is unknown. Therefore, global histone modification in a well-characterised series of breast carcinomas (n=880) with long term follow-up was assessed using immunohistochemistry and tissue microarray. Specific antibodies were used to detect acetylation of H3 (Lys9 and Lys18) and H4 (Lys12), and dimethylation of histone H4 (Arg3) and H3 (Lys4). The presence of these chromatin 'marks' was correlated with clinicopathological variables and patients' outcome.
Reduced levels of histone acetylation/dimethylation were observed in medullary-like carcinomas, whereas they were readily detected in lobular and tubular carcinomas. Reduced global histone acetylation/dimethylation was significantly associated with established poor prognostic variables; larger tumour size, higher stage, recurrence, distant metastases and higher mortality rate. Survival analyses showed low detection of the histone modifications, with the exception of acetylated H3K9, was associated with shorter overall survival and shorter disease free interval.
Our results show, for the first time, that global changes in specific histone modifications patterns may play an important role in breast cancer development and progression and their reduced expression is associated with poor prognosis and shorter survival.

PL2

Microarray analysis of mesenchymal tumours identifies brachyury, a novel biomarker for chordomas

{P} S Vujovic, R Tiraboco, S Henderson, C Boschoff, AM Flanagan
UCL, London, United Kingdom

Microarray analysis of connective tissue tumours was performed with the aim of assessing the relationship between different tumour types and identifying unique molecular fingerprints, that could be used to aid diagnosis. We found that differentiated tumours cluster in groups reflecting common tissue types: neurofibromas/schwannomas, alveolar/embryonal rhabdomyosarcomas, and well-differentiated liposarcomas/lipomas. Similarities were found between the expressional profiles of chondrosarcomas and chordomas. The diversity of gene expression was greatest in osteosarcomas, leiomyosarcomas and pleomorphic sarcomas. Many of the genes identified as the molecular fingerprints of a particular tumour type were related to the metabolism or function of its tissue of origin,: adipocytic tumours were distinguished by the expression of perilipin and lipoprotein lipase, genes involved in lipid metabolism. The brachyury transcription factor, known to be involved in notochordal development, was uniquely expressed in chordomas. All 53 chordomas analysed and an embryonic notochord showed nuclear positivity with a brachyury antibody and was negative in over 300 other neoplasms including 163 chondroid neoplasms, making it a specific marker for the notochord and tumours showing notochordal differentiation. Brachyury expression was detected in one tibial neoplasm with morphology and immunohistochemistry (CK19, S100, CEA, EMA, HMBE1+) consistent with a chordoma, making this an extra-axial chordoma.

PL3

Bone Marrow Contributes To Functionally Active Cells In Tumour Stroma

{P} N.C. Direkze, R. Jeffery, K. Hodivala-Dilke, F. Burke, F.T. Hunt, R. Poulsom, F. Balkwill, N.A. Wright, M.R. Alison
Cancer Research UK, London, United Kingdom

BACKGROUND Bone marrow contributes to myofibroblast populations in multiple organs and also tumour stroma. However, there has been a call to show that these cells are functionally active. Here we show that bone marrow-derived cells in murine insulinomas are producing appropriate mRNA indicating functionality. In addition we demonstrate the bone marrow contribution to myofibroblast populations in a second murine tumour model.

METHODS Female transgenic mice that develop pancreatic insulinomas were transplanted with male bone marrow. After tumour harvest bone marrow-derived myofibroblasts were demonstrated by combining *in situ* hybridisation (ISH) for the Y chromosome with immunohistochemistry for α-smooth muscle actin (αSMA). To show the functionality of these cells a new method to combine the techniques of ISH for DNA and mRNA with immunohistochemistry for markers myofibroblast morphology (αSMA) was developed and used to assess the tumour tissue.

RESULTS Bone marrow-derived cells in tumour stroma express mRNA for pro-collagen1α.

CONCLUSION Bone marrow contributes collagen-producing cells to tumour stroma. These functionally active cells provide further evidence that the bone marrow provides part of an important axis in the process of fibrosis which may ultimately lead to the development of new methods of targeting therapy.

PL4

Array comparative genomic hybridization profiling of early invasion in colorectal adenomas

{P} AJ Watkins, G Poulogiannis, K Daly, N Johnson, M-Q Du, AEK Ibrahim, AH Wyllie, MJ Arends
Department of Molecular Histopathology, Addenbrooke's Hospital, University of Cambridge, Cambridge, United Kingdom

The frequencies of major genetic alterations in colorectal neoplasia have been well characterised, but those at the transition from severe dysplasia to early invasive carcinoma are the least studied, with predominantly *p53* changes on chromosome 17p being implicated. We investigated the adenoma/carcinoma transition using paraffin sections of 22 cases of severely dysplastic adenomas with early stalk invasion. Samples of both the invasive carcinoma component and the adjacent adenoma were retrieved by microdissection. The DNA extracted was subjected to whole genome amplification. DNA copy number changes were analysed by array comparative genomic hybridization at 1 Mb resolution, relative to a mixed control sample of normal DNA. We found a characteristic pattern of changes frequently occurring at the transition between adenoma and carcinoma with DNA copy number losses on chromosomes 1p, 17 and 18q and gains on chromosomes 13q and 20q, often affecting only small regions of the chromosomes. In conclusion, molecular profiling by array CGH of paired adenoma and carcinoma samples has identified a small panel of changes in DNA copy number that occur at the adenoma-carcinoma transition.

PL5

Recurrent ovarian cancer cDNA signatures

{P} A Laios[1], SA O'Toole[1], BL Sheppard[1], N Gleeson[1], T D'Arcy[1], EPJ McGuinness[1], M Ring[2], P Smyth[2], O Sheils[2], JJ O'Leary[2]
[1]. Department of Obstetrics and Gynaecology, Trinity College Dublin, Dublin, Dublin8, Ireland, [2]. Department of Histopathology, Trinity College Dublin, St James Hospital and Coombe Womens Hospital, Dublin, Dublin8, Ireland

The majority of epithelial ovarian carcinomas (EOC) are of serous subtype, with most women presenting at an advanced stage. Approximately 80% respond to initial chemotherapy but eventually relapse. We aimed to investigate whether primary and recurrent ovarian cancers express a distinct gene profile, using gene expression arrays and to identify markers of recurrence.
Tumour tissues from primary serous papillary EOC, FIGO S3, G3 were profiled against recurrent of the same grade using the Applied Biosystems 1700 microarrays. R and Panther were used for statistical analysis.
A 187-gene signature was identified as differentially expressed between primary and recurrent tumours (FDR 0.1). Quantitative RT-PCR was used to validate the microarray data on 21 targets yielding a high concordance ($r=0.874$, $P<0.01$).
Our results suggest a multifactorial mechanism for recurrence; the elevation of Claudin16, suggesting the enhancement of adhesive properties of tumour cells, the upregulation of EGFR ligands such as BTC and activation of the EGF pathway and intracellular signalling via a calcium binding protein S100B. These genes, thought to be correlated to the mode of action of chemotherapeutic agents, may provide evidence that mechanisms involved in recurrence are specific to the drugs used.

PL6

Limited Expression of the Coxsackie and Adenovirus Receptor in Pancreatic Cancer May Reduce Suitability of Adenoviral Gene Therapy

{P} CS Verbeke[1], S Hamdan[2], J Booth[1], HS Pandha[3], GE Blair[2]
[1]. Dpt of Histopathology, St James's University Hospital Leeds, Leeds, United Kingdom, [2]. School of Biochemistry and Molecular Biology, University of Leeds, Leeds, United Kingdom, [3]. Postgraduate Medical School, University of Surrey, Surrey, United Kingdom

Background: The response of pancreatic cancer to existing therapies is limited and gene therapy may offer a new approach. Efficient adenovirus infection of target cells depends on the presence of the Coxackie and Adenovirus Receptor CAR.
Aim: To evaluate the potential efficacy of adenoviral therapy in pancreatic cancer, we evaluated CAR expression in pancreatic cancer cell lines and archival normal and cancer tissue.
Method: CAR expression in 10 human pancreatic cancer cell lines was analysed by flow cytometry and correlated with transduction efficiency of recombinant Ad5CMVEGFP virus. Using the same antibody, immunostaining was performed on tissue microarrays containing 188 pancreatic ductal adenocarcinomas and 68 controls.
Results: The level of CAR expression correlated with the susceptibility to adenoviral transduction of the pancreatic cancer cell lines. Immunostaining for CAR was absent in 103 (55%) of adenocarcinomas, while moderate to strong staining was observed in 85 (45%) cases. Absence of CAR immunolabeling correlated with poor histological tumour differentiation. In normal tissue, immunolabeling was seen in islet cells and inter- and intralobular duct epithelium.
Conclusion: Absence of CAR expression in a considerable proportion of pancreatic cancers and constitutive CAR expression in normal tissue may reduce the suitability of adenoviral gene therapy for pancreatic cancer.

PL7

Novel FISH probes designed to detect *IGK-MYC* and *IGL-MYC* rearrangements in B-cell lineage malignancy reveal a new breakpoint cluster region designated BVR2

{P} A Dogan, RR Einerson, ME Law, PJ Kurtin, ED Remstein
Mayo Clinc, Rochester, MN, United States

Detection of translocations involving *MYC* at 8q24.1 in B-cell malignancies (BCL) is important for diagnostic and prognostic purposes. However, routine detection of *MYC* translocations is often hampered by the wide variation in breakpoint location within the *MYC* region, particularly when a gene other than *IGH*, such as *IGK* or *IGL*, is involved. To address this issue, we developed and validated 4 FISH probes: 2 break-apart probes (BAP) and 2 dual color, dual fusion (D-FISH) probes to detect *IGK-MYC* and *IGL-MYC* translocations. *MYC* rearrangements (4 *IGK-MYC*, 12 *IGL-MYC*, and 4 unknown partner gene-*MYC*) were correctly identified in 20 of 20 archival BCL specimens known to have *MYC* rearrangements not involving *IGH*. Seven specimens, all of which lacked *MYC* rearrangements using a commercial *IGH/MYC* D-FISH probe, were found to have 8q24 breakpoints within a cluster region >350-645 kb 3' from *MYC*, provisionally designated as Burkitt rearrangement region-2 (BVR2). In light of the discovery of the distally-located BVR2 breakpoint cluster region, it is likely that the frequency of *MYC* translocations in BCL has been significantly underestimated and it will be important to use *MYC* FISH probes that cover a breakpoint region at least 1.0 Mb 3' of *MYC* to identify *MYC* rearrangements comprehensively.

PL8

Matrix Metalloproteinase production is inhibited in degenerate intervertebral discs by IL-1Ra gene transfer

{P} CL Le Maitre, AJ Freemont, JA Hoyland
University of Manchester, Manchester, United Kingdom

Conventional therapies for Low back pain (LBP) do not target the cause, which is often disc degeneration (DD). During DD an increase in IL-1 with related increases in matrix metalloproteinase (MMP) activity is thought to be involved in the pathogenesis of DD. We have previously shown that IL-1Ra can be delivered successfully to degenerate discs. This study investigates the ability of IL-1Ra gene transfer to inhibit MMP production in degenerate discs.
Degenerate human IVD tissue injected with Il-1Ra infected/or non-infected IVD cells or control tissue was used for the immunohistochemical analysis of MMPs 1,3,7 and 13.
MMPs were expressed in human degenerate tissue. The number of immunopositive cells for MMPs 1,3 and 7 but not 13 was significantly decreased following injection of uninfected normal disc cells. However, in explants injected with Ad-IL-1Ra infected cells a greater decrease was seen. Furthermore the number of MMP 13 immunopositive cells was significantly decreased following injection with Ad-IL-1Ra infected cells compared to uninjected control tissue ($P<0.05$).
This study suggests that an *ex vivo* method of IL-1Ra gene transfer could be a feasible approach to reduce the production of MMPs within the degenerate disc and hence prevent the further degeneration of the disc in LBP.

Abstracts

Posters

P1

KiRas4A promotes growth in renal cell carcinoma

{P} S Fury, L Christie, S fleming
University of Dundee, Dundee, United Kingdom

We investigated the expression of the minor 4A isoform of KiRas in renal cell carcinoma (RCC) cell lines during siRNA transfection. KiRas is part of the growth signal transduction cascade in many cell types. It exists as two alternative isoforms, the ubiquitously expressed KiRas4B and the alternative KiRas4A that has a more restricted pattern of expression. Kidney is one of the small number of tissues in which the 4A isoform is expressed. Experimental data suggests that it may be important in the mineralocorticoid dependent growth response.
We have examined the expression of the KiRas isoforms in Renal carcinoma cell lines: RCC4 cells that are VHL (Von Hippel Lindau) deficient and RCC4 plus VHL cells have been transfected with pcDNA3-VHL that conveys neomycin resistance and encodes a normal VHL tumour suppressor gene product as a control.
We designed siRNAs specific to KiRas4A using Reynold's Algorithm. After having transfected the cells, RNA was extracted at 24, 48 and 72 hours post transfection and was then subjected to reverse transcription PCR in order to identify the specific 4A isoform. PCR products were then run on a 1.5% agarose gel and bands of the appropriate size were detected. The PCR product was directly sequenced and found to match the published sequence of KiRas4A. These studies showed that RCC4 cell lines express KiRas 4A and that following siRNA transfection KiRas4A knock down was achieved. Furthermore at each time point, the number of transfected cells was less than non-transfected cells. On average RCC4 VHL cells showed a 40% reduction in cell number when transfected with KiRas4A siRNA and VHL deficient cells showed a 73% reduction in cell number post siRNA transfection.We hypothesise that KiRas4A has a growth-promoting role in renal cell carcinoma and in renal epithelial.

P2

Diagnostic Utility Of Alpha-Methylacyl-CoA Racemase And Basal Cell Marker 34 betae12 In The Assessment Of Prostate Needle Biopsy Specimens

{P} A NAVEED, N DALAL
[1] *Tameside General Hospital, Manchester, United Kingdom,* [2] *Tameside General Hospital, Manchester, United Kingdom*

Prostatic adenocarcinoma is one of the major health problems in the male population. The diagnosis of prostatic carcinoma in core biopsies and small suspicious foci of glands could sometimes be a very challenging task for the Histopathologist. Immunosatins can sometimes be helpful in the diagnosis. Immunohistochemical markers such as basal cell markers and the new stain alpha-methylacyl-CoA racemase can be useful in such situations.
The objective of this project was to see the diagnostic utility of the immunostains 34betaE12 and P504S when used in combination. A literature search was also performed. The panel of racemase and 34betae12 was performed on 30 prostatic core biopsies of adenocarcinoma and PIN. The results showed that the panel was useful in small foci (10% of the cases) of the tumour and the two stains were complementary to each other in all cases. The sensitivity and specificity of P504S is high when used in combination with a basal cell marker.
This combination can be of diagnostic utility when limited amount of tissue is present.

P3

Stem Cell Kinetics, Carcinogenesis And The Red Cell Paradox

{P} J A Morris, Priya Mairembam
University Hospitals of Morecambe Bay, Lancaster, United Kingdom

From published literature we have obtained estimates of the total number of cells produced in a human lifetime in major organs and systems. The total number is 9×10^{15}, of which 5×10^{15} are red cells, 2.5×10^{15} are neutrophils, and only 1×10^{15} are epithelial cells. We found no relationship between the total number of cells in a system and the risk of malignancy in that system. This is contrary to conventional concepts of carcinogenesis. The conventional view is that carcinogenesis is a stochastic process in which mutations accumulate in asynchronously dividing stem cells; cancer arises in a stem cell that has, by chance, acquired a set of *n* specific deleterious mutations. But if this idea were correct erythroleukaemia would be one of the commonest forms of malignant disease. If by contrast malignancy arises in fully differentiated cells then the red cell paradox would be explained since mature red cells shed their nucleus. This explanation, however, requires a revision of our theories of stem cell kinetics. A previously published model, based on a stem cell hierarchy, is described. This model is consistent with a differentiated cell origin of cancer. The number of cell generations is $\log_2 N$, where N is the total number of cells produced in a human lifetime ($\log_2 N < 60$). If the hierarchy is strictly maintained malignancy would be rare, but if the hierarchy is disturbed and differentiated cells become established as asynchronously dividing stem cells for a localised anatomical unit then malignancy can arise. It is shown that this model solves the man/mouse paradox, Peto's paradox as well as the red cell paradox; it also explains Wright's enigma

P4

Array CGH & Expression Profiling of Fibro-epithelial Lesions of the Breast

A Jones[1], A.M. Hanby[2], C.E. Gillett[3], R. Walker[4], D. Rowlands[5], A. Colclough[6], S. Hales[7], C. Smith[8], PH Tan[9], I.P.M. Tomlinson[1], {P} E.J. Sawyer[1]
[1]. *Cancer Research UK, London, United Kingdom,* [2]. *Leeds General Infirmary, Leeds, United Kingdom,* [3]. *Guys Hospital, London, United Kingdom,* [4]. *University of Leicester, Leicester, United Kingdom,* [5]. *New Cross Hospital, Wolverhampton, United Kingdom,* [6]. *Colchester General Hospital, Colchester, United Kingdom,* [7]. *Countess of Chester Hospital, Chester, United Kingdom,* [8]. *Frimley Park Hospital, Frimley, United Kingdom,* [9]. *Singapore General Hospital, Singapore, United States*

We have used 1Mb array CGH to investigate copy number changes in 7 fibroadenomas (FA), 16 benign and 10 borderline/malignant phyllodes tumours (PT). For this analysis we have only considered large scale changes involving a whole or part (greater than a third) of a chromosome arm.
Malignant/borderline PTs showed more chromosomal-scale changes than benign tumours and FAs. (Mean number of chromosomal-scale changes per tumour was 4.8 (range 0-26), 1.8 (0-7) and 0.42 (0-2) for each group respectively). Losses were more common than gains. 53% of the changes in the malignant/borderline group were loss, 80 % in the benign group and 100% in the FAs. The commonest changes were: partial loss of 1p (15 cases, 11 benign, 3 borderline/malignant, 1 FA); loss of part or whole of 17p (9 cases, 3 benign, 5 borderline/malignant, 1FA) and gain of 1q (6 cases, 2 benign, 4 borderline/malignant). Loss of 17p was associated with the malignant/borderline phenotype (Fishers exact test- $p = 0.017$). The region of loss includes the p53 gene. We have performed SSCP and sequenced exons 4-8 of the p53 gene and have not found any evidence of mutation. We are continuing to analyse the p53 gene and other genes in this area. We have also found that partial loss of 1p is a common finding in fibro-epithelial lesions and are currently analysing genes within this region.
We have performed expression array analysis on these tumours and this data will be presented. Preliminary analysis using hierarchical clustering suggests that it is possible to identify a benign and malignant expression signature. We hope that this may help predict how borderline tumours are going to behave clinically.

P5

Her-2 testing in breast cancer: observer effects and associations with histopathology

{P} C Dunn, EA Mallon, LS Murray, F Duthie, JJ Going
University of Glasgow, Glasgow, United Kingdom

Background. Use of Herceptin in breast cancer requires accurate Her2 testing by immunohistochemistry (e.g. 'HercepTest') and FISH. This audit of 'HercepTest' results and FISH testing in '2+' carcinomas looks for any observer effect in 'HercepTest' reading. *Methods.* EAM and JJG independently read 'HercepTest' staining on 2981 (EAM) and 1590 (JJG) breast cancers (total 4571; all stained in one laboratory) during 01/01/03-31/12/05. Allowable scores were 'negative' (0, 1+); 2+; and 3+ (positive). FISH for *HER2* amplification was performed on all 2+ carcinomas. Complete histopathology data for 771 breast carcinomas treated in North Glasgow included size, type, grade, node involvement, steroid receptor status and vascular invasion. *Results.* Distribution of 3+, 2+ and negative carcinomas was 18%, 14% and 68% for EAM and 10%, 16% and 74% for JJG. The difference (EAM-JJG) in the percentage scored 3+ had 95% CI 6.0 to 10.0%. Of 2+ cases scored by EAM, 54/351 (15.4%) were *HER2* amplified while 55/239 (23.0%) of 2+ cases scored by JJG were *HER2* amplified. This difference (EAM-JJG) had 95% CI -7.6 to -14.1. Positive Her2 status was associated with higher grade, ductal type, lymph node positivity and ER negativity. *Conclusions.* Immunohistochemical Her2 testing remains subject to inter-observer differences, even when scored by experienced breast pathologists adhering strictly to published guidelines.

P6

BRAF (T1799A) MUTATIONS ARE NOT EXPRESSED IN NON-THYROID PAPILLARY TUMOURS

{P} RJ Flavin, P Smyth, SP Finn, S Cahill, EM O'Regan, M Ring, J O'Leary, O Sheils
Histopathology Research,Trinity College Dublin, Dublin, Ireland

BACKGROUND: The Ras/Raf/MEK/ERK signalling pathway has been implicated in a variety of human neoplasms, with BRAF mutations having been detected in tumours such as melanoma, low-grade serous papillary carcinomas of the ovary and papillary carcinoma of the thyroid (PTC). We investigated the hypothesis that BRAF (T1799A) mutations might be expressed in non-thyroid papillary tumours. DESIGN: Papillary tumours from FFPE tissue (N=57) comprised 20 serous carcinoma of the ovary, 10 renal cell carcinoma, 10 urothelial cell carcinoma, 9 serous peritoneal carcinoma, and 8 endometrial serous carcinoma. Tumour was laser captured microdissected, DNA extracted, and analyzed for expression of mutated BRAF T1799A by Taqman™ SNP detection. RESULTS: All 57 tumours were homozygous for wild type BRAF. CONCLUSION: BRAF (T1796A) mutations are restricted to certain subgroups of papillary tumours (PTC, low grade serous carcinoma of the ovary) and do not serve as a molecular link between tumours with a papillary phenotype.

P7

Identification Of Cell Cycle Regulatory Molecules As Biomarkers In Cervical Cancer By Gene Expression Profiling

{P} K Astbury[1], C Martin[2], M Ring[1], O Sheils[2], JJ O'Leary[2]
[1]. Coombe Women's Hospital, Dublin, Ireland, [2]. Trinity College Dublin, Dublin, Ireland

Cervical cancer is the second most common female malignancy worldwide. Human papillomavirus (HPV) is strongly implicated in its pathogenesis. HPV viral oncoproteins, E6 and E7, interfere with cell cycle regulatory pathways involving p53 and retinoblastoma protein (pRb). HPV E7 interaction with pRb causes release of transcriptionally active E2F, which then stimulates increased production of other cell cycle regulatory molecules.

CaSki, C33A, SiHa and HeLa cervical cancer cell lines were cultured. Total RNA was extracted from harvested cells. RNA quality was confirmed by gel electrophoresis. 5μg total RNA from 3 passages of each cell line and 5μg total normal cervix RNA in triplicate (BioChain) was labeled with digoxigenin using a 2 step RT-IVT approach and hybridised to Applied Biosystems human genome survey microarrays.

Data analysis was performed using Spotfire software. Samples were normalised and p values calculated using a t test. Comparing malignant to benign datasets, 6979 genes were differentially expressed, with 2286 up-regulated and 4693 down-regulated. Up-regulation of 14 of these genes, including a number of E2F target genes, was confirmed in all 4 cell lines by RT and quantitative real time PCR using ABI Gene Expression TaqMan assays. These may represent potential cervical cancer biomarkers

P8

Expression Of $\alpha_5\beta_1$ Integrin In Paraffin-Embedded Human Normal And Degenerate Disc Tissue

{P} J FRAIN, CL LE MAITRE, AJ FREEMONT, JA HOYLAND
University of Manchester, Manchester, United Kingdom

Few studies have investigated the expression of integrins in the human intervertebral disc (IVD). Integrin $\alpha_5\beta_1$ has been implicated as an important component of cellular mechanotransduction pathways in articular cartilage. As the IVD is also a loaded tissue and abnormal pressure has shown to be a significant causative agent in IVD degeneration it seemed pertinent to analyse integrin $\alpha_5\beta_1$ expression in normal and degenerate IVD tissue to investigate whether differences in protein expression were exhibited.

Immunohistochemistry was performed on 24 archival paraffin-embedded sections of human IVD ranging from normal to severely degenerate IVD. A mouse monoclonal anti-human $\alpha_5\beta_1$ integrin antibody was used and positive/negative controls included. The percentage immunopositive cells was calculated for nucleus pulposus (NP), inner (IAF) and outer annulus fibrosus (OAF).

As expected with a transmembrane protein, staining was localised to cells, not the surrounding matrix. Immunopositivity was predominantly observed in the NP and IAF with very little expression observed in the OAF. There was no significant difference between the NP and IAF. Results showed no correlation between percentage $\alpha_5\beta_1$ expression and grade of degeneration.

These results suggest that $\alpha_5\beta_1$ expression varies in the different regions of the IVD but expression does not appear to change with degeneration.

P9

Investigating the Effects of Hydrostatic Pressure on Nucleus Pulposus Cell Gene Expression in Short-term Alginate Culture

{P} J FRAIN, CL LE MAITRE, AP FOTHERINGHAM, AJ FREEMONT, JA HOYLAND
University of Manchester, Manchester, United Kingdom

There are limited studies on the effects of hydrostatic pressure on human intervertebral disc (IVD) nucleus pulposus (NP) cells in alginate culture. In this study the effect of load on normal and degenerate NP cell gene expression was investigated.

Primary NP cells from 1 human normal and 1 degenerate IVD sample were cultured in monolayer prior to seeding in 1.2% alginate constructs at 1 million cells/ml. After 48 hours in alginate culture a loading regime of 0.8-1.7 MPa at 0.5Hz. was applied for 2 hours. RNA extraction and cell viability assessment were performed 1 hour after loading ceased. Relative real-time PCR was performed for several target genes known to be within the human IVD using 18S as a housekeeping control.

Cell viability ranged between 80-100% and was not affected by loading. Relative to the unloaded control samples real-time PCR data showed no change in aggrecan, type II collagen or c-fos gene expression for loaded normal samples yet MMP-3 was downregulated. In degenerate samples a decrease was observed in all gene expression investigated.

These results show that normal and degenerate NP cells regulate gene expression differently in response to hydrostatic loading suggesting an altered mechanotransduction mechanism with disease.

P10

miRNA Expression Signatures in Cervical Cancer

{P} CM Martin, K Astbury, O Sheils, JJ O'Leary
Department of Pathology, Coombe Women's Hospital,& Trinity College Dublin, Dublin 8, Ireland

Cervical cancer, a potentially preventable disease, remains the second most common female malignancy worldwide. Human papilloma virus (HPV) has been implicated in the development of virtually all cases of cervical cancer. MicroRNAs (miRNAs) are small non-protein-coding RNAs known to negatively regulate expression of protein coding genes. Studies have indicated differential expression patterns of miRNAs in cancer and normal tissues and interactions between miRNAs and specific oncogenes.

In this study we examine expression of 180 miRNAs in cervical cancer cell lines; C33A (HPV negative) and CaSki (HPV 16 and 18) and normal cervical tissue using the Applied Biosystems TaqMan MicroRNA Assays Human Panel - Early Access Kit.

C33A and CaSki cells demonstrated distinct miRNA expression signatures in comparison with normal cervical tissue. In addition, differential miRNA expression patterns were observed between HPV positive and negative cell lines, with the predicted function of differentially expressed targets coinciding with a number of cell cycle regulatory molecules, including CDKN2a, CDK 6 and 8, Cyclin D2, Cdc14, E2F and Rb.

These findings highlight the potential importance of miRNA in the complex pathobiology of cervical pre-cancer and cancer. These miRNAs may serve as potential biomarkers of pre-invasive cervical disease and potential therapeutic targets.

P11

Profiling Markers Of Prognosis In Colorectal Cancer

{P} M S Lyall, S R Dundas, G I Murray
University of Aberdeen, Aberdeen, United Kingdom

Colorectal cancer is one of the most common forms of cancer in developed nations and the incidence of this disease is increasing. There is a need to further stratify prognostically distinct groups of colorectal cancer and the purpose of this study was to identify prognostically significant immunohistochemical marker profiles in colorectal cancer.

A range of markers [pRb, p16, p21, p27, p53, pCNA, Cyclin D1, bcl-2, EGFR, C-erb-B2, topoisomerase-I, liver fatty acid-binding protein, matrix metalloproteinases (MMP) 1-3,7,9 and 13, MT1-MMP, MT2-MMP and tissue inhibitors of MMP 1-3] of putative prognostic significance have been investigated by immunohistochemistry on formalin-fixed, wax-embedded sections in a series (n = 90) of Dukes C colorectal cancers. An immunohistochemical score based on the intensity of immunoreactivity and, where relevant, the proportion of immunoreactive cells was established for each marker.

Unsupervised two-dimensional hierarchical cluster analysis identified three distinct cluster groups (designated 1-3) with different marker profiles. There were significant survival differences between groups 1 and 2 (log rank = 11.48; p = 0.0007) and between groups 1 and 3 (log rank = 8.32; p = 0.0039). Multivariate analysis showed that the complete marker profile was independently the most significant prognostic factor (hazard ratio, 2.27; 95% confidence interval, 1.15 - 4.48; p = 0.004).

This study has identified an immunohistochemical marker profile of colorectal cancer and showed that it is an independent indicator of prognosis in this type of cancer.

P12

Malignant Melanoma; Using Tissue Microarrays to Understand a Killer

{P} S Hegarty, E Murray, D Ryan, M Rafferty, WM Gallagher, PA Dervan
Pathology, UCD School of Medicine and Medical Science, Dublin, Ireland

Malignant melanoma is one of the most aggressive and lethal forms of cancer. Unfortunately, incidence rates are increasing rapidly each year, with little improvement in treatment for this invasive disease, which generally fails to respond to conventional chemotherapy. The current level of understanding of tumour progression in malignant melanoma is poor, and the aggressive nature of an individual tumour is inferred from the histological appearance. Given the mortality associated with metastases, it would be useful to have a reliable panel of immunohistochemical markers that would allow the pathologist to gauge the invasive potential of the tumour, and advise the clinician with regard to follow up and surgery.

However, the small size of most primary tumours is problematic when evaluating large numbers of immunohistochemical markers, as the tumour blocks are quickly depleted. We present work from a study evaluating the usefulness of tissue microarray technology in sampling a large cohort of cutaneous malignant melanomas, and subjecting them to standard immunohistochemical methods. We have developed methods to overcome technical difficulties in working with cores of malignant melanoma, and our results indicate that tissue microarrays are a reliable alternative to full sections of malignant melanoma in the research environment.

P13

Comparative Genomic Hybridisation study on a Cohort of Melanoma Cell Lines

{P} D Ryan, E McSherry, S Hegarty, M Rafferty, P.A. Dervan, W.M. Gallagher
Conway Institute,UCD, Dublin, Ireland

In the Western world, the incidence and mortality rates of malignant melanoma are rising. The lack of accurate prognostic indicators and effective therapies emphasises the need for a better understanding of genetic and phenotypic changes in melanoma formation and progression.
In this project, our aim was to investigate chromosomal aberrations in melanoma using an array-based comparative genomic hybridisation (CGH) on a panel of melanoma cell lines.
TheVysis Genosensor CGH system was used to examine DNA from ten melanoma cell lines, representing different stages of melanoma progression. The most frequent genetic changes observed with the melanoma cell line model were amplifications on chromosomes 7p and 20q.
To examine if similar genetic abnormalities occur in melanoma tumours, we are planning to perform fluorescence *in situ* hybridisation (FISH) on a melanoma tissue microarray (TMA). This TMA contains a histopathologically identified cohort of 250 cases of primary cutaneous malignant melanoma from 1993-2003.The resulting data will contribute to increased understanding of melanoma progression and potentially lead to improved clinical management.
1. Gallagher WM *et al.* (2005). Multiple markers for melanoma progression regulated by DNA methylation: insights from transcriptomic studies. Carcinogenesis. 2005 Nov; 26(11): 1856-67.

P14

Pool of Only 3 HPV-types Enough as a Screening Method and for Measuring Effectiveness of the Prophylactic HPV Vaccine?

{P} S Hovland[1], F Karlsen[1], AK Lie[2], B Risberg[2], P Snijders[3], C Meijer[3], B Borge[2], W Ryd[4], H Skomedal[1], T Kadima[5], L Batumbola[5], L Kyembwa[5], D Mukwege[6], E Bilayi[5], M Mvula[7], E Berle[8], R Chirimwami[7], JP Kashongwe[7], M Arbyn[9]
[1]. NorChip, Klokkarstua, Norway, [2]. Dep of Pathology, Norwegian Radium Hospital, Oslo, Norway, [3]. Dep of Pathology, VU University Medical Center, Amsterdam, Netherlands, [4]. Cyt. Dept. Sahlgrenska University Hospital, Gøteborg, Sweden, [5]. Centre Hospitalier CELPA, Bukavu, Congo, DR, [6]. Panzi Hospital CEPAC, Bukavu, Congo, DR, [7]. General Hospital, Bukavu, Congo, DR, [8]. Gynecology Dep., Ullevål University Hospital, Oslo, Norway, [9]. Scientific Institute of Public Health, Brussels, Belgium

OBJECTIVE Define which HPV types and number in the pool should be included in nucleic acid tests to detect CIN2+ for reaching acceptable accuracy levels for future screening of cervical cancer.

METHODS Samples for HPV-testing were collected from 343 women (age: 25-60, median 37) of the Democratic Republic of Congo. The PreTect HPV-Proofer assay (NorChip) based on Real-Time NASBA was used to identify HPV16,18,31,33,45 from mRNA. Additionally, 4 types (HPV35,51,52,58) were also detected using the NASBA assay. HPV DNA was detected by consensus PCR (GP5+/6+) followed by enzyme immuno assay and reverse line blot typing performed by Chris Meijer's group. One colposcopically directed biopsy was taken from all women. If the transformation zone was not completely visible with colpsocopy, ECC was performed. Histology was regarded as gold standard.

RESULTS AND CONCLUSIONS Sixteen histologically confirmed CIN2+ (4.7%) were identified. DNA HPV16 was most frequently detected in CIN2+ and presence of HPV16 DNA solely revealed a sensitivity of 31% and specificity of 99%. An octet of HPV DNAs (i.e. HPV 16,33,18,51,66,56,45,35) resulted in a sensitivity of 100% and a specificity of 90% for CIN2+. 100% sensitivity for CIN3+ (with a specificity of 92.5%) was reached with HPV DNA testing using a pool of 5 types (HPV16,18,33,66,45). mRNA of HPV 16,18 where most frequently detected in CIN2+. The sensitivity for CIN2+ calculated for HPV16 mRNA was 31% with 100% specificity. A pool of HPV16,18,33 mRNA yielded a sensitivity of 75% and a specificity of 99%. However, for CIN3+ cases a pool of only 3 mRNA HPV types, (i.e. HPV 16,45,52) seems enough to reach maximum sensitivity (87.5%) with a specificity of 96%. Including HPV18 instead of HPV52 (i.e. pool of mRNA 16,45,18) yielded the same sensitivity (87.5%) but a specificity of 95%. These new data may create possibilities for world wide coverage both related to future vaccines and to screening methods.

P15

Formalin-Fixed Paraffin-Embedded Material is a Reliable Source of RNA for Molecular Genetic Analysis Provided Small Fragments are Targeted

{P} L Lam[1], F Berisha[1], TC Diss[2], A M Flanagan[3]
[1]. Royal National Orthopaedic Hospital, London, United Kingdom, [2]. University College London Hospitals, London, United Kingdom, [3]. Institute of Orthopaedics and Musculoskeletal Sciences, UCL, London, United Kingdom

Specific therapy for tumours increasingly requires diagnosis at a genetic level as more drugs against specific mutations become available. Analysis of RNA is required to detect fusion genes involved in translocations, a common event in sarcomas. RT-PCR for such analysis was originally performed on fresh/frozen material due to difficulties in acquiring suitable RNA from formalin-fixed, paraffin-embedded (FFPE) samples. However this is impractical when providing diagnoses for rare tumours at remote sites. Improvements to quality of reagents, plastic-ware, and the development of protocols for extraction of RNA from FFPE now permit study of routinely processed material. To assess the quality of RNA that can be obtained from routine histological samples we extracted RNA, using a commercial kit, from 281 consecutive samples, some decalcified, submitted for molecular genetic analysis. Each sample was reverse transcribed using a G6PD anti-sense primer and amplification of 86bp, 141bp and 200bp fragments of the G6PD gene attempted. Amplification of the 86bp fragment was achieved in 268 (95%), of the 141bp fragment in 253 (90%) and of the 200bp fragment in 220 (66%). These data show that FFPE tissue samples are a suitable source of RNA for RT-PCR analysis in most cases provided that small fragments are targeted.

P16

RNA quality from fresh human surgical specimens; a comparative assessment of tissue specific and general factors.

{P} L Willerton[1], A Lowe[1], J Hair[2], B Gusterson[1], A.M. McNicol[1]
[1]. University of Glasgow, Glasgow, United Kingdom, [2]. Glasgow Royal Infirmary, Glasgow, United Kingdom

Background. Analysis of gene expression requires high quality RNA but this is thought to be difficult to achieve from human tissue due to factors delaying freezing, particularly with surgical specimens. The detailed effects of such factors have been poorly documented. **Objective.** To determine the quality of RNA extracted from surgical specimens, assessing to what extent tissue type and individual case are associated with variation and to monitor the effects of delayed freezing. **Methods.** Human duodenum, breast and pancreas were obtained from surgical specimens transported on ice to the Pathology Department. Mouse duodenum was used as control. Samples were snap frozen at time points up to 4 hours after collection. RNA was extracted using a Trizol technique. Integrity of the RNA (RIN) was assessed using the Agilent Bioanalyser. Data were analysed using one way Anova with LSD post hoc analysis. **Results.** For breast and duodenum there was no significant difference between RIN values up to 3hrs, but a significant decrease after 4 hrs. The quality of RNA from pancreas was poorer at all time points. Differences were observed between individual cases of the same tissue. RNA integrity was also reduced when tissue defrosted after initial freezing. **Conclusion.** These results suggest that good quality RNA can be obtained from some tissues even when freezing is delayed, but that subsequent thawing may significantly reduce the quality.

P17

COLONIC STEM CELLS ARE UNABLE TO SELECTIVELY SEGREGATE THEIR GENOME

{P} N Mandir, SL Preston, T Hunt, NA Wright, MR Alison
[1]. Cancer Research, UK, London, United Kingdom, [2]. Cancer Research, UK, London, United Kingdom, [3]. Cancer Research, UK, London, United Kingdom, [4]. Barts and The London, Queen Mary's School of Medicine and Dentistry, London, United Kingdom, [5]. Centre for Diabetes and Metabolic Medicine, London, UK

Small intestinal stem cells may protect themselves from DNA replication errors by segregating genome at cell division - retaining 'template' strand and passing on only newly-synthesised DNA strand to their progeny. We examined if this took place in the colon when new stem cells and 'template' strands are being produced following irradiation.

Twenty C57Bl/6J mice received irradiation followed by 6-hourly injections of tritiated thymidine (^{3}HTdR) over 48 hours. After 8 days, the mice were injected with bromodeoxyuridine (BrdU), 6-hourly for 2 days and sacrificed in 25 days. Colons were stained with anti-BrdU and autoradiography. Crypts were scored and labelled cells mapped.

Nineteen percent of colonic cells were labelled with ^{3}HTtdR at day-8, none in day-20. Fewer labelled cells were distributed throughout the crypt. BrdU-labelling was almost complete in day-1, but declined sharply in 3-5 days. No labelled cells seen in day-25. Cells containing both labels were rare and only in day-1.

Presence of ^{3}HtdR-labelling at day-8 indicated retention of part of the genome. The disappearance of label over 12 days and dilution of BrdU at the same rate, indicates no selective retention of the 'template' strand in the colon. May contribute to the higher incidence of colonic tumours.

P18

Relationship of CXCR4 Expression to Stage in Colorectal Cancer

{P} C Lauder, P Daforno, JH Pringle
Leicester University, Leicester, United Kingdom

Purpose
The CXCR4 chemokine receptor mRNA has been shown to be a significant factor for local recurrence and survival in colorectal cancer (CRC). In this study we aimed to correlate the expression of CXCR4 protein to the stage of disease.
Methods
50 cases of CRC were identified to represent a spectrum of disease stage within the Dukes B and C subgroups. Paraffin embedded archival tissue was then used to provide sections from the primary tumour as well as lymph node and distant metastatic spread where found. Subsequent analysis by immunohistochemical methods was used to determine CXCR4 expression, as determined by scoring the intensity of staining. This was performed by two independent observers with results compared using a kappa score to measure the degree of agreement (K=0.84). These scores were then used to analyse the relationship of CXCR4 expression with TNM stage and metastatic sites.
Results
The expression of CXCR4 in the primary tumour was not found to be associated with regional lymph node (P=0.78) or distant metastatsis (P=0.7). No significant differences in positive staining were observed between primary and metastatic tumors (P=0.72). Positive CXCR4 staining was found more frequently in liver metastasis than lymph nodes (P=0.025).
Conclusions
Chemokine receptor CXCR4 expression in CRC specimens may not provide additional information in assessing the tumours likelihood to spread and recur when combined with conventional staging investigations.

P19

PML nuclear domains, intranuclear injury-responsive structures that may audit DNA damage and influence cell fate.

{P} T Rich, A Varadaraj, L Laredj, C Dovey, N Lubben, A. H. Wyllie
Department of Pathology, University of Cambridge, Cambridge, United Kingdom

PML nuclear domains (PML-NDs) are intranuclear bodies of around 0.2μm diameter that comprise a shell of PML protein and many cargo proteins. These include proteins involved in the DNA damage response, transcriptional regulation and apoptosis. Within 8h of exposure to ionising radiation, PML-NDs juxtapose with the protein complexes that accumulate at DNA damage sites. Cells lacking PML are deficient in the capacity to activate apoptosis. Here, we study the increase in number of PML-NDs following ionizing radiation. We show that this directly reflects the extent of DNA damage. It is initiated swiftly (within 30 minutes), is sensitive to doses as low as 1Gy, and reverts to pre-irradiation levels within 12 hours in cells destined to survive. It is delayed in cells that lack the damage-sensitive kinase Chk2. Interestingly, onset of long-term cycle arrest in untransformed IMR90 or WI38 fibroblasts, or apoptosis in HCT116 colorectal cancer cells is accurately predicted by failure of restoration of PML-ND number within 12 hours. The results support the view that PML-NDs may provide a nuclear microenvironment in which evaluation of the adequacy of DNA repair takes place, a process necessary to inform the critical decision between reversion to normal, or commitment to cytostasis or apoptosis.

P21

A Prospective Study Confirms Mismatch Repair Status Predicts Outcome in Colorectal Cancer

{P} I P Chandler[1], S Popat[2], H Pan[3], D Zhao[4], Y Shao[4], Z Chen[3], R S Houlston[2]
[1]. Dept of Cellular Pathology, St George's Hospital, London, United Kingdom, [2]. Section of Cancer Genetics, Institute of Cancer Research, Sutton, Surrey, United Kingdom, [3]. CTSU, Richard Doll Building, Roosevelt Drive, Oxford, United Kingdom, [4]. Department of Abdominal Surgery, Tumour Hospital, Beijing, China

Mismatch repair (MMR) deficiency is one of the most promising markers for colorectal cancer prognosis. Results from some published studies of MMR status as a prognostic marker may have been influenced by retrospective, non-blinded design, together with heterogeneous and small datasets. To address these deficiencies, we analysed 697 patients in a large prospective multi-centre randomised trial of adjuvant intra-portal 5FU. Tissue microarrays were generated from the tumours and MMR status assessed by immunohistochemistry for MLH1, MSH2, MSH6 and PMS2. Analysis was blinded to clinical data. An overall survival advantage was observed (HR=0.39, 95%CI: 0.18-0.85. p=0.02). The mortality in the MMR deficient group was 21.2% versus 43.3% in the MMR intact group (p=0.03). The classification of colorectal cancer based on MMR status provides a prognostic index that reflects tumour pathobiology. Cancers characterised by MMR deficiency have a markedly better prognosis and this suggests that MMR status should be used in therapeutic decision-making

Abstract P20 has been withdrawn

P22

DALM Associated With Longstanding Crohn's Disease – A Case Report

{P} S Nayak[1], H Denley[2]
[1]. Blackburn Royal Infirmary, Blackburn, Lancs, United Kingdom, [2]. Manchester Royal Infirmary, Manchester, United Kingdom

Dysplasia in inflammatory bowel disease (IBD) is a well recognised complication of long-standing disease, particularly in Ulcerative Colitis (UC). This has been categorised as either flat or associated with a raised lesion or mass (dysplasia associated lesion or mass [DALM]). DALM occurs in less than 5% of patients with dysplasia, almost all cases (except one) having been reported in cases of UC. We present a case in a 55 year old woman with a history of Crohn's disease who presented clinically with a rectovaginal fistula. On CT scan there was significant thickening of the rectum with an inflammatory mass involving the vagina and rectum. The patient underwent an abdominoperineal resection. Gross examination of the resection specimen showed a plaque like thickening with irregular nodularity in the rectum, particularly in the region of the fistula. Microscopically there was definite evidence of Crohn's disease in the rectum and presence of moderate dysplasia. We propose that this fulfils the currently accepted description of DALM. This case highlights the need for clinical vigilance to identify neoplastic complications in long-standing Crohn's disease, not just UC.

P23

The Role of Fat Clearance Technique in Colorectal Carcinoma

A Khattab, {P} H Awad, H Pitchamuthu, S Walsh, E Salmo, N Y Haboubi
Trafford General Hospital, Trafford, United Kingdom

Aim:
To establish the value of fat clearance technique in lymph node yield in cases of Dukes "A" and "B" colorectal cancer.
Methods:
We analysed 26 random cases of colorectal cancer, 25 of which were Dukes "B" and one was Dukes "A", which underwent xylene clearance after standard gross dissection.
Results:
The age ranged between 48 and 90 years (mean 76 years). Thirty-four percent were in the recto-sigmoid and 23% in the caecum. The length of the resection ranged from 145m to 810mm. The number of lymph nodes prior to fat clearance ranged from 0 to 37 with a mean of 11.27 nodes. The number of lymph nodes after fat clearance ranged from 3 to 68 with a mean of 19.88 lymph nodes. Four cases of Dukes "B" and one case of Dukes "A" were upgraded to Dukes "C1".
Conclusion:
Fat clearance technique is useful in improving the accuracy of the Dukes classification and has prognostic significance. It can be used in specimens, which on initial examination appear to be Dukes "A" or "B". We believe that the benefit of accurately staging the disease outweighs the slight delay in providing the final report as a result of this method.

P24

The Role of Immunohistochemistry in Differentiating Between Pseudo-invasion and True Invasion in Colonic Polyps

{P} A Khatab, P Hyder, E Salmo, N Haboubi
Trafford Healthcare NHS Trust, Manchester, United Kingdom

Introduction:
The aim of this study was to employ immunohistochemical markers in order to differentiate colonic adenomas with pseudoinvasion from those with true invasion.
Methods:
Fifteen cases of colonic neoplasia were analysed and categorised into three groups (adenomas, adenomas with pseudo-invasion or misplaced epithelium and adenocarcinomas). Ki67, p53, E-cadherin, alpha smooth muscle actin (SMA) and h-caldesmon immunostains were applied to all categories.
Results:
Ki67, p53, E-cadherin and SMA showed positivity (moderate to strong) in all three categories with positivity around dysplastic, misplaced epithelium and malignant glands. p53 showed positivity in misplaced epithelium in a range between 0 to 90% (average 22%). However, 60 to 80% of dysplastic glands and misplaced epithelium positively stained (moderate to strong) with h-caldesmon and only up to 5% of malignant glands showed moderate peripheral staining.
Conclusion:
We concluded that Ki-67, p53, E-cadherin and SMA are of no value to differentiate between true and pseudo-invasion in colonic polyps. *h*-caldesmon, however, may be of some assistance in separating pseudo-invasion versus true invasion with benign adenomas and adenomas with misplaced epithelium showing high percentage of positive staining than malignant glands.

P25

Radiation Colitis: Assessment By Grading

{P} Khalifa Sawalem, Paula Hyder, Mohammed Edilbe, Emil Salmo, Najib Haboubi
Trafford Healthcare NHS Trust, Manchester, United Kingdom

Introduction: Pre-operative radiotherapy may induce radiation colitis. Histological evaluation of radiation colitis needs to be reproducible to assess disease progression, post-operative complications and radiation dosage, but, to our knowledge, histological grading of radiation colitis has not been reported.
Aim: To evaluate radiation effects on rectal mucosa and assess their severity using a simplified grading system akin to that of inflammatory bowel disease.
Methods: Histological sections were examined from the rectal mucosa of 30 cases of rectal carcinoma where the patients had received preoperative short course radiotherapy. The irradiation changes were graded as mild, moderate or severe.
Results: Mild changes were characterised by an eosinophilic cell infiltrate and occasional crypt abscesses. Moderate changes were typified by numerous crypt abscesses, a mixed inflammatory infiltrate including eosinophils and mild epithelial atypia. Severe changes were distinguished by prominent epithelial cellular atypia, mucosal attenuation and fibrosis, and comparatively little eosinophilic infiltrate. Seven patients developed post-operative complications, of whom 2 showed severe, 4 showed moderate and 1 showed mild changes.
Conclusion: The severity of radiation colitis can be assessed and graded according to its histological features. Increased severity of disease appears to be associated with a higher degree of cellular atypia and a lesser eosinophilic infiltrate.

P26

Radiation Colitis:Grading and Complications; Are They Related?

{P} Khalifa Sawalem, Heyam Awad, Sheila Walsh, Emil Salmo, Najib Haboubi
Trafford Healthcare NHS Trust, Manchester, United Kingdom

Aim: To evaluate the effect of preoperative radiotherapy (RT) on the rectal mucosa and to investigate the relationship between preoperative radiation and postoperative complications.
Methods: We examined the histological sections of 34 cases with rectal carcinoma that received short or long course of preoperative RT. The rectal mucosa within the irradiation field was examined histologically for crypt abscesses, eosinophilic infiltrate, cellular atypia, surface erosion and inflammation. These changes were graded as mild, moderate or severe and correlated with postoperative complications.
Results: Of the 34 cases, 30 (88.24%) received short course RT and 4 (11.76%) received long course.
Histologically, the changes were mild in 9 cases (26.47%), moderate in 10 (29.41%) and severe in 15 (44.12%).
Clinically, only seven cases developed complications, all of these received short course RT. Of the cases that had complications, 1 case had mild, 4 had moderate and 2 had severe histological changes. The complications included bleeding, obstruction, and leakage.
Conclusion: There was no trend in the development of complications with the severity of histological changes. A larger study is needed to verify these results.

P27

Idiopathic gall bladder siderosis! An unusual case.

{P} J. K. S. S. Philip, S. Bhatnagar
Fairfield General Hospital, The Pennine Acute NHS Trust, Bury Lancashire, United Kingdom

We present a case of extensive siderosis of the gallbladder. To the best of our knowledge this condition has not been described in the english literature. The cause of iron deposition in soft tissues can be primary due to hereditary haemochromatosis or secondary to iron overload states like thalassaemias or rarely excessive iron ingestion. There is a single report of iron deposition in experimental piezoelectric shockwave application for the fragmentation of biliary calculi involving six animals. However, the deposits in these cases were small and microscopic. In our case extensive dark brown to black pigmentation was noticed on macroscopic assessment and on microscopy this was a golden brown pigment strongly positive with Perl's stain. The patient was a 73 years old woman who underwent simultaneous right hemicolectomy for an adenocarcinoma caecum and cholecystectomy for cholelithiasis. There is no known history of haemochromatosis or any other haematological disorder.

P28

Multiple Chylous Cysts - Our Experience Of Rare Mesenteric Masses

{P} S Patil, S Malhotra, J Harvey
Dept. of Histopathology, PathLinks, County Hospital, Lincoln, United Kingdom

Mesenteric chylous cyst is a rare lesion which is difficult to differentiate clinically from other mesenteric cysts such as enteric or dermoid cysts. The English literature contains a few cases of single chylous cysts, but only one case of multiple chylous cysts has been described. We report a second case.
A 52 year old female presented with an abdominal mass and underwent exploratory laparotomy following an inconclusive MRI. A multilocular cyst 20x20cm was seen arising from the mesentery of the sigmoid colon but not attached to uterus, ovaries or intestines. Hundreds of separate white mesenteric cysts were also seen. A sigmoid colectomy was done with en-bloc resection of mesentery.
The cysts contained milky fluid, and the internal and external surfaces were smooth.
On microscopic examination, the cysts were lined by endothelium with lymphoid aggregates in the wall and occasional calcified areas.
The patient remains alive and disease-free fifteen months after surgery.
Although chyle-filled cysts may occur secondary to lymphatic obstruction, we believe the clinical course indicates that this case is a primary chylangioma, a benign tumour occurring in misplaced foci of lymphoid tissue, which proliferate and accumulate fluid.
This spectacular lesion represents an extremely rare cause of abdominal mass.

P29

Primary Small Cell Undifferentiated Carcinoma Of The Large Intestine Associated With Ulcerative Colitis

{P} S Patil, N Manglik, A McGregor, J Xuereb
[1]. Dept.Of Histopathology,PathLinks,County Hospital, Lincoln, United Kingdom, [2]. Dept. Of Histopathology,Leicester Royal Infirmary, Leicester, United Kingdom

Longstanding Inflammatory Bowel Disease, particularly Ulcerative Colitis (UC), predisposes to carcinoma of the colon, usually adenocarcinoma. The risk is higher when the entire colon is involved, when the disease is continuous, unremitting and longstanding (over 10 years). Primary small cell undifferentiated carcinoma (SCUC) of the large bowel in the background of UC is an unusual occurrence. Only two such cases have been reported in English literature. We discuss two further cases, as correct diagnosis will predict the clinical course, management and prognosis.
Both our patients were middle aged males with a greater than 15yr history of severe UC with panproctocolitis and on surveillance. Panproctocolectomy contained a small cell undifferentiated carcinoma. Extensive liver and peritoneal metastases were noted. Chemotherapy with carboplatin and etoposide was given, with good tumour response.
SCUCs are highly aggressive tumours when compared with adenocarcinoma of the same stage, and are associated with mean survival of 11 months. They occur frequently with distant metastases and carry a poor prognosis, even if diagnosed in the early stage. Hence, it is very important to diagnose and distinguish them histologically from other tumour types as it plays an important role in the patient management and prognosis.

P30

Detection Of Pathogenic *Yersinia* DNA In Crohn's Disease By Real Time Polymerase Chain Reaction

{P} S Barnaras, O Rotimi
Leeds General Infirmary, Leeds, United Kingdom

Introduction
Crohn's disease (CD) is a chronic inflammatory bowel disease of unknown aetiology. Many infective agents have been implicated in the pathogenesis of CD including Yersinia and a recent study reported the presence of Yersinia DNA in 30% of a series of CD cases (Lamps LW *et al. Am J Surg Pathol,* 2003). Given the significance of such a finding, we undertook a similar study in a cohort of CD cases using the same primer pairs.
Methods
We carried out a real-time PCR study using specific primer pairs for pathogenic *Y.* enterocolitica (YE) *ail* and *Y.* pseudotuberculosis *(YP) inv* DNA in 75 test samples of 59 archival resections of CD patients 30 years old or less. 15 cases of active ulcerative colitis (UC) and five granulomatous appendicitis (GA) were used as disease controls. *YE* pure culture and *YP* genomic DNA were used for positive Yersinia controls. Albumin was used as housekeeping gene.
Results
None of the CD, GA or UC cases showed amplification of either *YE* or *YP* DNA. However, YE and YP DNA was amplified from the Yersinia control samples in every PCR run and albumin was amplified in all the samples.
Conclusions
This negative finding is in contrast to the recent publication and therefore, the role of Yersinia in CD remains controversial and demands further investigation.

P31

Differences In DNA Copy Number Profiles Between Gastric Cancers In Young And Elderly Patients

{P} TE Buffart[1], B Carvalho[1], E Hopmans[1], V Brehm[2], E Klein Kranenbarg[2], TBM Schaaij-Visser[1], PP Eijk[3], NCT van Grieken[1], B Ylstra[3], CJH van de Velde[2], GA Meijer[1]
[1]. Dept. pathology VU University Medical Center, Amsterdam, Netherlands, [2]. Dept. Surgery Leiden University Medical Center, Leiden, Netherlands, [3]. Micro-array Core Facility VU University Medical Center, Amsterdam, Netherlands

While most gastric cancers occur in elderly patients, a substantial number of cases occur in young patients. Gastric cancer is a heterogeneous disease at the genomic level and different patterns of DNA copy number alterations are associated with a different clinical behavior. Aim of the present study was to explore differences in DNA copy number alterations in relation to age of onset of gastric cancer.
DNA isolated from 46 gastric cancers of 17 patients <50 years of age (median 43 (21-49)) and 29 patients ≥70 years of age (median 75 (70-83)) was analyzed by genome wide microarray comparative genomic hybridization. Patterns of DNA copy number aberrations were analyzed by hierarchical cluster analysis, whereafter cluster membership was correlated to age group. In addition, supervised analysis was performed.
Hierarchical cluster analysis of the array-CGH date revealed three clusters with different genomic profiles that correlated significantly with age (p=0.006). Cluster 1 mainly contained young patients while elderly patients were divided over clusters 2 and 3. Chromosomal regions 11q23.3 and 19p13.3 contributed most to age related differences in tumor profiles.
Gastric cancers of young and old patients belong to groups with different genomic profiles, which likely reflects different pathogenic mechanisms of the disease.

P32

GLUT1 STAINING PATTERN IN CONTIGUOUS AND NON-CONTIGUOUS COLORECTAL ADENOMAS AND THEIR CORRESPONDING CANCERS

{P} S Bakhiet, J O'Sullivan, M Izzuan, G Keating, J Gorman, H Mulcahy, J Hyland, D O'Donhue, K Sheahan
[1] St. Vincent University Hospital, Elm Park, Dublin 4, Ireland,
[2] University College Dublin, Dublin 4, Ireland

Background: Glucose Transporter 1(GLUT-1) is a high affinity glucose transporter. Increased glucose uptake at cell membranes is a metabolic characteristic of tumour cells. GLUT-1 at the membrane represents the active form of the protein in cancers.
Elevated levels of Glut1 have been reported in many human malignancies. We have previously shown a correlation between GLUT-1 staining and a more advanced stage of colorectal cancer. In addition we have described distinct supranuclear and membranous staining patterns. However the correlation between different staining patterns in contiguous and non-contiguous adenomas in colorectal cancer has not yet been studied.
Design: Using tissue microarray and immunohistochemistry, the aim of this study was to evaluate the localisation of Glut1 in 31 adenomas and corresponding tumours. 14/31 adenomas were contiguous to the cancer. Four 6mm cores from each sample were scored and assessed for Glut1 staining pattern (either supranuclear, mixed or membranous).
Results: In contiguous adenomas (4/14), a mixed or membranous GLUT-1 pattern was detected. All these adenomas showed high-grade dysplasia. All corresponding cancers for these cases showed the same Glut1 staining pattern. However, no non-contiguous adenomas showed membranous Glut1 staining.
Conclusion: Membranous or mixed pattern of GLUT-1 correlates with high-grade dysplasia in adenomas and their corresponding cancers.

P33

Adenocarcinoma in Barrett's oesophagus, without previously detected dysplasia

{P} Y Alizadeh, D Treanor, N Scott, J Wyatt
Department of Histopathology, St. James's University Hospital, Leeds, West Yorkshire, United Kingdom

Introduction: Oesophageal adenocarcinoma (AC) is believed to develop from Barrett's oesophagus (BO) via the metaplasia-dysplasia-adenocarcinoma sequence, therefore surveillance can be useful in detecting early malignant changes. However, some tumours appear to develop without going through these stages. Our aim was to determine whether failure to detect this sequence was due to failed surveillance or rapidly developing malignant change.

Methods: Pathological reports of 8/265 (7 males) patients with the diagnosis of AC and previous BO were reviewed (1996-2003) noting date of the initial BO diagnosis and follow up examinations, number of biopsies per endoscopy, and their histology.

Results: Median number of endoscopies from presentation to diagnosis of AC was 3 (range 2-7), median interval 25 months (range1-52 months) and median number of biopsies per endoscopy 6 (range 4-25). Two patients developed AC despite good surveillance protocol but in 3 surveillance requirements were not met (delays 1.8 and 2.5 years between endoscopies and one with insufficient biopsies). Three patients were not in the surveillance program with biopsies interval >3years. Only 1/8 patient had dysplasia detected prior to AC, later diagnosed after 2.5 years between endoscopies.

Conclusion: Rapid disease progression (2/8), inadequate number of biopsies (1/8) and delay between endoscopies (5/8) may contribute to the discovery of AC without prior dysplasia. Our results indicate that it is important not to prolong the time interval between surveillance endoscopies in BO patients.

P34

Stomal Haemorrhage Secondary To Portal Hypertension Treated With TIPSS

{P} A Kabeer[2], A Widdison[2], G Maskell[3], J Mathew[1]
[1]. Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [2]. Department of Surgery, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [3]. Department of Clinical Imaging, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom

Aim: to describe a case of recurrent stomal variceal haemorrhage secondary to portal hypertension.
Case Report: A 60 year-old woman presented with recurrent ileostomy haemorrhage following panproctocolectomy and ileo-stomy for severe ulcerative colitis in 1997. Between 1998 and 2002 she had six admissions for stomal haemorrhage, was managed by local treatment and had ileostomy revision twice.
The histology of her stomal stump showed dilated vessels. Subsequent investigations including ultrasound, mesenteric angio-gram, CT liver and distal celiac angiogram, confirmed stomal varices, shunting of blood, cirrhotic liver changes and possible portal vein stenosis with portal hypertension. She subsequently underwent transjugular intrahepatic portosystemic shunt (TIPSS) placement twice, the last one in 2004 and she has had only one minor bleed since then which was managed by local treatment.
Conclusion: Haemorrhage from gastrointestinal varices is a major complication of portal hypertension but varices at ectopic sites other than the gastro-oesophageal junction or rectal sites is quite rare. Diagnosis of these presents a challenge and the treatment options can be quite cumbersome, ranging from local measures as suturing and silver nitrate application, revision of stoma and porto-systemic shunt placements to liver trans-plantation in cases of cirrhosis. Surgical porto-systemic shunts present a definitive treatment option for recurrent bleeding but can be associated with significant morbidity and mortality. TIPSS is a much less invasive technique which, by reducing the morbidity and mortality associated with surgical shunts, is a more attractive option. We describe an unusual case where recurrent stomal haemorrhage exposed underlying portal hypertension.

P35

Recurrent Ovarian Carcinoma Presenting As A Primary Rectal Tumour

{P} M Kabeer[1], P Arumugam[1], G Maskell[2], A St John[3], J Mathew[3]
[1]. Department of Surgery, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [2]. Department of Clinical Imaging, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [3]. Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom

Aim: To describe a case of metastatic ovarian carcinoma masquerading radiologically and pathologically as a primary rectal tumour.
Case report: A 58 year-old lady presented with rectal bleeding and constipation. On investigation by CT and MRI, a rectosigmoid junction lesion was found which showed an intraluminal component. This was thought to be a primary rectosigmoid lesion although she had been treated for a stage 2B endometrioid ovarian carcinoma five years earlier and had been on HRT; CA125 was marginally raised at 55ku/L (n=1-30ku/L).
Sigmoid colon biopsies showed an adenocarcinoma, with focal squamous differentiation in which the luminal aspect showed features of a dysplastic "tubular adenoma"; these features were considered by an expert to be more likely to be a primary colonic adenocarcinoma than metastasis. She subsequently underwent anterior resection: histology showed an adenocarcinoma that was strongly with CK7 and CA125 positive, CK20 negative and weakly ER positive: CEA was equivocal. On this basis she was treated as having colonic metastasis of an ovarian carcinoma and put on platinum chemotherapy. Two tears later, the patient is free from measurable recurrence.
Conclusion: Metastatic ovarian carcinoma can occur as intraluminal lesions with serosal sparing even in the absence of peritoneal disease and can present with localised obstruction. CK7, CA125, CEA and CK20 are useful in differentiating these from primary bowel tumours.
This case is a very good example of metastatic ovarian cancer managed by surgery and chemotherapy and this approach should be adapted in selected patients with similar disease.

P36

MCMs And Geminin: Similar Expression But Different Relationship To Survival In Small Bowel Compared To Gastric Cancer

{P} R Sekhon[1], J Knock[1], M Shah[1], K Maude[2], K Stoeber[3], G Williams[3], P Howdle[2], P Quirke[1], H Grabsch[1], R Watkins[1]
[1]. Leeds Institute of Molecular Medicine, University of Leeds, Leeds, United Kingdom, [2]. Academic Unit of Medicine, University of Leeds, Leeds, United Kingdom, [3]. Dept of Histopathology, University College London, London, United Kingdom

Small bowel adenocarcinoma (SBC) is a rare neoplasm and little is known about the molecular alterations involved in the development of SBC. The initiation of DNA synthesis is a final and critical step in growth control and is therefore of importance in carcinogenesis. Minichromosome maintenance proteins (MCM) and geminin are vital for the regulation of DNA replication. We showed previously that altered expression of these proteins is associated with patient survival in gastric adenocarcinomas (GC). MCMs and geminin have not been studied in SBC.

To further understand small bowel tumorigenesis we studied the expression of MCM2, MCM5, MCM7 and geminin in 160 SBC by immunohistochemistry. Their relationship with clinico-pathological data, proliferation marker Ki67 and patient survival was analysed. Cluster analysis was used to compare the expression of MCMs, geminin and Ki67 between GC and SBC.

High expression of all proteins was associated with high grade in SBC but not in GC. Expression of MCM2 and MCM5 was higher in patients with coeliac disease. Cluster analysis showed that the expression profile of GC is similar to that of SBC. In particular intestinal type GC is closely related to SBC. In contrast to GC, neither a relationship between MCMs, geminin, Ki67 and survival nor other clinicopathological data was seen in SBC.

This is the first study to demonstrate that MCMs and geminin play a role in tumour differentiation but not in tumour progression or survival in SBC. The similar expression pattern of the proteins but different relationship with clinicopathological data in SBC compared to GC indicates that their function may be impaired in either GC or SBC. This warrants further investigation as does the higher expression of MCM2 and MCM5 in coeliac related SBC.

P37

The Effect Of Sub-specialisation In Histopathology On Lymph Node Retrieval In Colorectal Cancer Resection Specimens

{P} M Gallacher, F Carey, S Walsh
Dundee University, Dundee, United Kingdom

Introduction: Lymph node staging is critical in colorectal cancer management. This study investigated if subspecialisation (SS) in histopathology influenced the number of retrieved lymph nodes (RLNs) and staging in colorectal carcinomas.
Methods: Colorectal carcinoma reports were analysed preSS (1995-96, n=344) and postSS (2002-05,n=468). Analysis included specimen type, length, RLNs and Dukes stage.
Results: Mean RLNs preSS was 7 (0-34) and postSS was 18 (0-74) ($p<0.001$). Pre SS specimens with <10 RLNs was 270 and ≥10 RLNs was 74. Post SS specimens with <10 RLNs was 70 and ≥10 RLNs was 398 ($p<0.001$). PreSS the number of Dukes C patients was 134 (39%) and post SS was 231 (49%) ($p<0.001$) Average specimen type and length were unchanged. In the preSS era specimens were dissected in general cut-ups. In the postSS era dissections were performed in dedicated colorectal cut-ups. In the preSS era consultants alone and trainees alone reported the majority of cases whereas in the postSS era almost all cases were dissected by trainees and reported with specialist gastrointestinal consultants.
Conclusion: Subspecialisation has increased the number of RLNs and increased the number of Dukes C patients. This occurred despite our hospital being a pilot site for colorectal cancer screening which would have been expected to decrease Dukes C patients numbers.

P38

FOREIGN BODY OBSTRUCTION IN A DISEASED BOWEL

{P} MONIKA TRIPATHI, ROGER LINDLEY
MEDWAY MARITIME HOSPITAL, KENT, United Kingdom

Gastrointestinal perforation results in an emergency situation that requires prompt treatment. There is a broad spectrum of etiologic factors that cause gastrointestinal perforation, neoplasm and foreign bodies among them. Ingested foreign bodies are likely to stop at any narrowing or angulations of the intestinal lumen, and perforations usually occur above the colon.
We report 3 case of a perforation caused by an ingested foreign body in a diseased bowel.
A 55 year old male presented with right iliac fossa pain, Appendicectomy done, showed, normal appendix histology, Meckel's diverticulectomy also done, showing ectopic gastric mucosa and perforation tract by a fishbone.
A 69 year old patient of colonic adenocarcinoma presented with acute obstruction, sigmoid colectomy done, showed a moderately differentiated adenocarcinoma(Duke's B) tumour in sigmoid colon and luminal blockage by a fragment of a chicken bone, apparently the cause of the obstruction.
A 65 year old known patient of Crohn's disease presented with small bowel obstruction, right hemicolectomy done for suspected ileocaecal mass, showed perforation by a plum seed.
Intestinal perforation occurs in less than 1% of cases of ingestion of foreign bodies.
Most perforations occur at narrowing and angulations. Eighty-three percent of foreign-body perforations occur in the ileum. Foreign-body perforations are more common in elderly patients who wear dentures, patients who have a mental impairment, and patients who chronically abuse alcohol. Although the imaging findings can be non-specific, the identification of a foreign body with an associated mass or extra luminal collection of gas in patients with clinical signs of peritonitis, mechanical bowel obstruction, or pneumoperitoneum can strongly suggests the diagnosis

P39

Nodular Hyperplasia Of The Terminal Ileum Mimicking Crohn's Disease

{P} A Naveed, H Denley
Manchester Royal Infirmary, Manchester, United Kingdom

Crohn's disease is a chronic inflammatory disease that may involve the entire length of the gastrointestinal tract. It may require treatment with systemic immunosuppressive therapy, necessitate long term patient follow up and is associated with a number of other clinical conditions.

In this report, we present the case of a 21 year old lady who initially presented with a five month history of diarrhoea and abdominal pain. Clinical investigation at that time included a barium follow through. This showed irregular nodularity of the terminal ileum, which was presumed to be due to Crohn's ileitis. She initially responded well to immunosuppressive therapy, but developed steroid dependence. Alternative treatments were either less effective or resulted in unacceptable side effects. Finally, surgery was considered necessary.

The right hemicolectomy revealed a localised area of nodularity within the terminal ileum, which on histology proved to be due to lymphoid hyperplasia. We will discuss the histopathological findings, necessary special studies, relevant differential diagnoses and putative mechanisms for this condition, with a review of relevant literature.

P40

Histopathological assessment of rectal carcinoma following pre-operative chemo-radiotherapy: development of a pilot dataset for reporting tumour response

{P} S M McGrath, R F T McMahon, K Skordilis
University of Manchester, Manchester, United Kingdom

Colorectal cancer continues to be a challenging clinical problem. Each year in the UK there are approximately 29,000 new cases and 18,000 deaths. The standard surgical treatment for distal rectal carcinoma is either a low anterior resection or an abdomino-perineal resecton. Pre-operative chemo-radiotherapy is offered to patients with advanced tumour when there is concern about the ability to perform a curative resection. Several studies have suggested that this approach can both reduce the incidence of local recurrence and improve survival rates. The effects of chemo-radiotherapy on the histology of rectal cancer have been addressed by several studies, each using differing methods of evaluation. Specific histological parameters may be related to the character and magnitude of the tumour response. It has therefore become necessary to consider differentiating between those rectal carcinomas that have received pre-operative therapy and those that have not using a more uniform approach. In this study we attempted to address this issue by performing a critical review of the available relevant literature. Our goal has been to suggest a supplementary pilot dataset for reporting colorectal specimens subjected to pre-operative chemo-radiotherapy. This would facilitate audit and hopefully help to identify important prognostic factors in these patients.

P41

The Role Of The Periglandular Myofbroblast Sheath In Gastric Carcinogenesis

{P} M G Deheragoda[1], S J Leedham[1], M Brittan[1], N C Direkze[1], S L Preston[1], D L S Stoker[2], M R Novelli[2], S A C McDonald[1], N A Wright[1]
[1]. Cancer Research UK, London, United Kingdom, [2]. University College Hospital, London, United Kingdom

Background: The stem cell is thought to be the site of genetic changes giving rise to gastric carcinogenesis. The gastric stem cell resides in a niche comprising myofibroblasts which make up the gastric periglandular myofibroblast sheath (PGMFS). These secrete growth factors and Wnt signalling factors which regulate stem cell activity.
Aims: To investigate the size of the gastric PGMFS in pre-neoplastic conditions.
Methods: Human gastric resection and biopsy specimens from antral and body type mucosa were evaluated for the presence of chronic gastritis, atrophic gastritis and intestinal metaplasia. Immunohistochemical staining with SMA and vimentin was used to detect the Gastric PGMFS and the size of the PGMFS was evaluated under different pathological conditions.
Results: The PGMFS was present in normal gastric mucosa. In both antral and body type mucosa the PGMFS showed a statistically significant increase in size (p value <0.05) compared to normal in atrophic gastritis and intestinal metaplasia.
Conclusion: The expanded PGMFS in pre-neoplastic conditions may promote gastric carcinogenesis.

P42

Epigenetic Changes In Familial And Sporadic Hyperplastic Polyps

{P} D Speake[1], DG Evans[2], S Ramsden[2], J O'Sullivan[2], F Lalloo[2], J Hill[1], RFT McMahon[3]
[1]. Department of Surgery, Manchester Royal Infirmary, Manchester, United Kingdom, [2]. Regional Genetics Service, St Mary's Hospital, Manchester, United Kingdom, [3]. Division of Laboratory and Regenerative Medicine, University of Manchester, Manchester, United Kingdom

Epigenetics describes the silencing of key tumour suppressor genes through methylation in the promoter regions of DNA. Its importance in carcinogenesis is receiving increasing attention, as methylation occurs in up to 30% of colorectal cancers (CRC) and has been linked to hyperplastic polyps (HPs) through a pathway independent of the adenoma-carcinoma sequence. The study objective was to determine the methylation status of 'sporadic' and 'familial' HPs. Patients were recruited via the routine endoscopy unit (sporadic) and the regional genetics service HNPCC database (familial). Methylation Specific PCR (MSP) was performed on DNA extracted from polyps to determine methylation status of the tumour suppressor genes MGMT, P16 and hMLH-1. 95 HPs were identified (55 sporadic; age range 28-78 years (median 51.1); surface area 0.03-0.79 (median 0.707) cm^2; males 47). 21 patients had confirmed methylation of MGMT: 17 'sporadic' patients (31%) and 4 'familial' patients (10%) (p=0.023). 21 patients had confirmed P16 methylation: 19 (35%) sporadic p and 2 (5%) familial (p=0.002). 12 patients had methylation of MLH-1: 11 (20%) sporadic and 1 (2.5%) familial (p=0.013). Significant differences are shown with methylation status in sporadic and familial HPs. Intuitively this may be expected since in familial polyp and cancer formation, defective mismatch repair due to mutation is more likely to be driving disease. However within the 'familial' polyp patients, there are individuals without mismatch repair mutations at increased risk of CRC. While it is appealing to consider methylation as a factor increasing this cohort's risk of CRC, our provisional results appear to refute this possibility. Our next strategy is to identify methylation in other genes and to perform immunohistochemistry on this cohort of polyps.

P43

The NOD2/CARD15 3020insC polymorphism is not associated with gastrointestinal disease in autistic patients.

{P} A Lynch[2], M Ring[2], O Sheils[1], CM Martin[1], JJ O'Leary[1]
[1]. Department of Pathology, Coombe Women's Hospital, & Trinity College, Dublin, Dublin 8, Ireland, [2]. Department of Pathology, Coombe Women's Hospital, Dublin 8, Ireland

A gastrointestinal lesion has been described in a cohort of children with developmental disorders. The intestinal pathology includes ileo-colonic lymphonodular hyperplasia and non-specific colitis, which lacks the specific diagnostic features of Crohn's disease or ulcerative colitis. The association between three polymorphisms within the NOD2/CARD15 gene on the IBD1 region of chromosome 16, and Crohn's disease is well established. The aim of this study was to investigate the presence of the most common of these risk alleles (3020insC) in autistic patients with gut pathology to predict the potential development of inflammatory bowel disease.
Genomic DNA extracted from bowel biopsies from a total of 13 autistic cases, 26 IBD and 20 non IBD controls was genotyped using a TaqMan SNP assay specific for the NOD2/CARD15 3020insC polymorphism. All autistic cases were homozygous for the wild type allele of the CARD15 3020insC gene, while two of the IBD controls were heterozygous for the CARD15 3020insC wild and mutant alleles.

This pilot study does not show or support an association between mutations in the CARD15 gene and the gastrointestinal lesion in autism.

P44

Cyclin D2 gene abnormalities in colorectal cancers identified by array comparative genomic hybridisation

{P} G Poulogiannis, K Ichimura, NGA Miller, IM Frayling, RG Morris, DJ Harrison, VP Collins, A Ibrahim, AH Wyllie, MJ Arends
University of Cambridge, Department of Pathology, Cambridge, United Kingdom

The retinoblastoma (RB) pathway regulates G1/S-phase transition and is known to be deregulated in colorectal cancer. Cyclin D is a key component of the RB pathway and one of the strongest predictors of shorter overall survival of diffuse large B-cell lymphomas is Cyclin D2 over-expression. The influence of large-scale chromosomal instability (CIN) and microsatellite instability (MSI) on the survival of sporadic colorectal cancer (CRC) patients is controversial. We analysed the chromosomal changes of >100 sporadic CRCs using high resolution 1Mb array-comparative genomic hybridisation (CGH). The tumours were evaluated for micro-deletions or micro-gains and these were correlated with Dukes' stage, survival and other clinicopathological features. Over 5% of the cases studied exhibited amplification of the chromosome 12p locus containing the cyclin D2 gene (CCND2). These tumours had a strong correlation with poorer survival and higher recurrence rate. Furthermore, flow cytometric analysis of these tumours showed that they all had a statistically significantly higher number of cells in both G2 and S-phases, indicating that the amplification of cyclin D2 is associated with evidence of a selective growth advantage. Analysis of all the small genomic abnormalities identified by 1Mb-array CGH identified novel genomic changes that could stratify microsatellite stable from unstable tumours, as well as tumours of higher and lower stage. We conclude that, as well as chromosome and microsatellite instabilities, other small localised genomic abnormalities, presumably affecting particular genes such as cyclin D2, may afford selective growth advantage in colorectal cancers.

P45

Role of Endoscopic Mucosal Resection in Staging and Therapy of Early Upper Gastrointestinal Neoplasia

{P} DN Poller, M Gwiggner, D Cowlishaw, S Toh, S Somers, P Bhandari
Queen Alexandra Hospital, Cosham, Portsmouth, PO6 3LY, United Kingdom

Introduction: Endoscopic mucosal resection (EMR) has been advocated for diagnosis and therapy of upper gastrointestinal neoplasia.
Methods: 11 patients underwent diagnostic /therapeutic EMR, 10 for Barrett's associated lesions, 1 for squamous cell dysplasia. Pre-EMR 9 patients were known to have high grade glandular dysplasia, 1 high grade squamous cell dysplasia and 1 intramucosal adenocarcinoma. Oesophagectomy was initially considered for all 11 patients. All patients had EMR with a targeted procedure using acetic acid (glandular dysplasia) or Lugol's iodine (squamous cell dysplasia).
Results: After EMR 6 patients were confirmed to show high grade dysplasia (5 glandular, 1 squamous), 1 intramucosal adenocarcinoma and 4 adenocarcinoma with submucosal invasion. Only 2 patients required oesophagectomy, neither showed evidence of residual tumour, 1 received chemoradiation and the remaining 8 patients remain under intensive endoscopic surveillance.
Conclusion: EMR is a new procedure with huge diagnostic and therapeutic potential in upper GI tract. EMR is an excellent staging tool in high grade dysplasia/early oesophageal cancer as it is more accurate than other staging modalities e.g. CT and EUS in assessing depth of tumour invasion and in confirming pathologically the presence or absence of invasive carcinoma. EMR is also useful therapeutically as an alternative to oesophagectomy or PDT.

P46

Angiogenesis in colorectal cancer - a comparison between three commonly used pan endothelial antibodies and two counting methods in assessing tumour angiogenesis

{P} C Johnson[1], S.J. Coates[2], D.M. Hemingway[3], J.H. Pringle[1], K.P. West[4]

[1]. Department of Cancer Studies and Molecular Medicine, University of Leicester, Leicester, United Kingdom, [2]. Leicester Medical School, Leicester, United Kingdom, [3]. Department of Surgery, Leicester Royal Infirmary, Leicester, United Kingdom, [4]. Department of Histopathology, Leicester Royal Infirmary, Leicester, United Kingdom

Angiogenesis is important in tumour growth and metastasis has been associated with a poor prognosis in a variety of tumours. However, there is much conflict as to the prognostic importance of microvessel density, which may be due to the wide variations in technique..
Tissue blocks from fifty patients with colorectal cancer were retrieved from the archives . Immunohistochemistry was performed using antibodies von Willebrand factor, CD31 and CD34. In addition a double staining method to identify proliferating endothelial cells using CD31 and Ki 67 was developed. Counting of microvessels was performed using two standard techniques (Weidner's and Chalkey's) and validated by an independent observer.
Preliminary results show that counts of CD34 are significantly higher than those obtained using CD31 and VIII (P =.003 and P= .000 respectively) However, CD34 tended to stain non – vascular structures, whilst CD31 was more specific for microvessels. Analysis of double staining shows that microvessel density counts currently in use overestimates the amount of endothelial cells undergoing proliferation.
This work is the first comparison between pan endothelial antibodies in colorectal cancer angiogenesis and emphasises the need for a standard approach to such work.

P47

Prognostic significance of bone marrow micrometastases in oesophagogastric cancer.

{P} Paul Ryan[1], Jaquie Kelly[2], Gearoid O'Sullivan[2], Garry Lee[4], Gerald C. O'Sullivan[4]

[1]. Dept of Histopathology, Cork University Hospital, Cork, Ireland, [2]. Cork Cancer Research Centre, National university of Ireland, Cork, Cork, Ireland, [3]. Dept of Surgery, Mercy University Hospital, and Cork Cancer Research Centre, National University of Ireland, Cork, Cork, Ireland, [4]. Department of Histopathology, Mercy University Hospital, Cork, Ireland

Aims: A majority of patients (70-80%) with oesophago-gastric cancer selected for curative surgery have cytokeratin positive micrometastatic tumour cells detectable in rib bone marrow. This study examined the prognostic significance of bone marrow micrometastases in patients with oesophagogastric cancer who were treated with en-bloc oesophagectomy with or without neoadjuvant chemoradiotherapy.
Methods: Rib segments were obtained from 54 consecutive patients. Marrow flushed from rib specimens was stained immunohistochemically using an anti-cytokeratin 18 antibody and examined by light microscopy. Patient survival was analysed with respect to tumour stage, histology, treatment and presence of CK18 positive micrometastases in marrow.
Results: Patients were followed up for on average 5.1 years (range 3 - 8.5 yrs). Univariate analysis showed cytokeratin 18 to be a significant prognostic indicator of overall survival ($p = 0.0198$). Tumour stage was also significant ($p = 0.00328$) whereas histology and neo-adjuvant chemoradiotherapy were not significant. Mutivariate analysis showed micromets were prognostically significant in node negative tumours ($p < 0.05$).
Conclusions: In patients undergoing potentially curative resection of oesophagogastric cancer the presence of bone marrow micrometastases is prognostically significant. The detection of micrometastases may be useful to refine the staging of these tumours, particularly in node negative cases.

P48

An Audit Of Duodenal Giardiasis Biopsies To Identify Possible Histological Features Of Immunodeficiency

{P} SJ Hayes

Hope Hospital, Salford, United Kingdom

Background and aims: Giardiasis caused by *Giardia lamblia* is a common intestinal protozoal disease. In industrialized countries it is often associated with water-bourne epidemics, but it is also known to be associated with immunodeficiency. The proportion of diagnosed giardiasis cases associated with immunodeficiency is uncertain, as specific histological features may not be routinely assessed.

Methods: Cases of giardiasis involving the duodenum were identified using the SNOMED system for the previous 10 years. Each of these biopsies were systematically examined for lamina propria plasma cells and nodular lymphoid hyperplasia, associated with common variable immunodeficiency and crypt cell apoptosis, a non-specific feature of *HIV* infection. A systematic search for evidence of the other infections which may be associated with immunosuppression, namely *Cryptosporidiosis, Cytomegalovirus, Microsporidiosis, Isospora Belli, Mycobacterium Avium Intra-cellulare and Strongyloidiasis* was made. In addition, other clinical data for these patients was reviewed, together with histology received from other sites.
Results: Of 15 cases reviewed, no specific features of immunodeficiency were found. Review of the clinical data did however reveal that one patient had a history of treated Follicular Lymphoma.
Conclusion: Our study supports a low associated incidence of immunodeficiency with giardiasis. A systematic approach for the identification of possible features of immunodeficiency is however advocated.

P49

Inhibition Of VEGFR Reduces Polyp Burden In The $Apc^{Min/+}$ Mouse Model Of Intestinal Cancer

{P} D Alferez[1], R Goodlad[1], J Watkins[1], R Poulsom[1], A Ryan[2], S Wedge[2], I Pyrah[2], N Mandir[1], R Wilkinson[2], N Wright[1]

[1]. Cancer Research UK, London, United Kingdom, [2]. AstraZeneca Pharmaceuticals, Macclesfield, United Kingdom

AZD2171, inhibitor of vascular endothelial growth factor receptor-2 (VEGFR-2) tyrosine kinase activity was used to study the effect of blocking angiogenesis in multiple intestinal neoplasia ($Apc^{Min/+}$) mice. $Apc^{Min/+}$ mice develop polyps due to a mutation in the adenomatous polyposis coli (APC) gene, as occurs in familial adenomatous polyposis in humans.
Studies were performed in which 5-mg/kg/day of AZD2171 was administered daily by oral gavage to 6-week old or to 10 week old $Apc^{Min/+}$ mice for 28-days after which the number and size of polyps in the small and large intestines were scored.
Firstly, AZD2171 reduced polyp number in the small bowel and colon by 46% and 62%, respectively $P<0.05$. Polyp diameter was also reduced by 39% in the small bowel ($P<0.001$) but was unchanged in the colon. Mean tumour burden (product of number and volume) in the small bowel was thus reduced 85% ($P<0.001$). Secondly AZD2171 reduced polyp diameter by 24%, decreasing tumour burden by 46% ($P<0.059$). AZD2171 reduced the number of cells expressing VEGFR-2 from 7.4 ±0.6 to 4.0 ±0.9 per villus ($P<0.022$).
AZD2171 reduced the number and size of polyps, in the earlier stages of polyp formation. VEGFR-2 signalling plays a key role in the development of intestinal adenomas.

P50

Bone Marrow Transplantation Induces Remission In Crohn's Disease: Where Do The Cells Go?

{P} S Schier[1], M Brittan[2], N Direkze[1], R Jeffery[1], MR Alison[2], NA Wright[1]

[1]. Cancer Research UK, London, United Kingdom, [2]. Queen Mary's School of Medicine & Dentistry, London, United Kingdom

Recent studies show that bone marrow (BM) transplantation induces remission in Crohn's disease. We used a mouse model to elucidate the underlying mechanism.

Female mice were lethally irradiated and rescued by a BM transplant from male donors. After 6 weeks experimental colitis was induced and colons analysed 1-14 days later. *In situ* hybridisation for Y chromosome was combined with immunohistochemistry for α-SMA, ICAM1, EphB4, ephrinB2 and other specific antigens to determine cell phenotype. A novel triple staining method combined *in situ* hybridisation, immunohistochemistry and autoradiography to show cell activity.

Cells derived from BM contributed significantly to activated myofibroblasts and to endothelial cells, pericytes and vascular smooth muscle lining cells (VSMLCs) in blood vessels. BM contributed to both angiogenesis and neovasculogenesis, confirmed by vessels composed entirely of BM-derived cells.

This is the first observation of BM-mediated neovasculogenesis in colitis. We provide an insight into the regenerative function of BM by highlighting the capacity of BM to engraft within inflamed colons and form multiple, functional lineages.

P51

The interleukin-6 (IL-6) 174 G/C promoter polymorphism and the inflammatory response in colorectal cancer (CRC).

{P} D O'Donnell[1], CW Steele[2], F Reid[1], D McMillan[2], AM McNicol[2]

[1]. Royal Infirmary, Glasgow, United Kingdom, [2]. University of Glasgow, Glasgow, United Kingdom

Background. IL-6 regulates transcription of C-reactive protein (CRP), a sensitive marker of inflammation. Raised CRP predicts poor survival in patients with apparently curative resection for CRC. We have demonstrated that high CRP levels are associated with low levels of CD4+ lymphocytes in the tumour and this is also associated with outcome. The 174 G/C polymorphism in the IL-6 promoter has been shown to influence the inflammatory response, the G allele being associated with higher circulating levels of IL-6, while the C allele, even in heterozygotes, appears to be inhibitory. Thus, the possession of a C allele might predispose to lower IL-6 levels, a less marked inflammatory response and a better outcome in patients with CRC.

Aims. We aimed therefore to correlate the distribution of the C and G alleles with circulating CRP levels, tumour specific survival and T lymphocyte infiltration in a series of patients who had undergone potentially curative resection for CRC.

Methods. To examine genomic sequences, DNA was extracted from normal colon sections from 66 cases of Dukes B and C CRC, amplified and digested using Nla III restriction endonuclease that cleaves the C allele. Products were analysed by acrylamide gel electrophoresis or using the CEQ^{TM}8000 Genetic Analyser. Patients were defined as C+ or C-.

Results. There was no significant difference in CRP levels, CD4+ lymphocyte infiltration or survival between the two groups. However, C+ individuals had higher levels of CD3+ ($p=0.052$) and CD8+ ($p=0.045$) intra-tumoural lymphocytes.

Conclusions. The 174 G/C IL-6 promoter polymorphism does not appear to play a major role in the regulation of circulating CRP levels in patients with CRC.

P52

Modelling of the expansion of normal human colonic crypts

{P} SAC McDonald[1], SJ Leedham[1], LC Greaves[2], D Oukrif[3], MR Novelli[3], M Deheragoda[1], DM Turnbull[2], JAZ Jankowski[4], NA Wright[5]

[1]. Cancer Research UK, London, United Kingdom, [2]. University of Newcaste Upon Tyne, Newcastle, United Kingdom, [3]. University College London, London, United Kingdom, [4]. University of Oxford, Oxford, United Kingdom, [5]. Barts and the London School of Medicine, London, United Kingdom

Recent data from our laboratory has shown that mitochondrial DNA (mtDNA) mutations can spread through the human colon by a process of crypt fission, where one crypt is able to divide into two daughters(Proc. Natl. Acad. Sci. 103:714-19).

The rate at which normal human crypts undergo fission is unknown. Bjerknes (J. Theor. Biol. 179:381-5) developed a mathematical model by which he calculated the rate at which crypts within aberrant crypt foci were able to expand. This predicts that if an aberrant crypt population was expanding at the same rate as normal crypts then the number of single aberrant crypts should be half of the number of those within foci.

Here we use this model to show that crypts deficient in mitochondrial cytochrome c oxidase (CoxSU1) expand at the same rate as those with normal CoxSU1 expression. We have counted the total number of deficient crypts within tissue sections stained with CoXSU1 from normal colons of 35 patients. 1298 deficient crypts were observed of which 597 were single. This gives a ratio of singletons to patches of 0.46 which are expanding only at 1.15 times faster than positive crypts.

These data suggest that crypts with or without mtDNA mutations expand at the same rate and that this is an appropriate model to calculate the crypt fission rate of the human colon.

P53

Phyllodes Tumour of the Vulva: Report of a Case and Review of the Literature

{P} Emil Salmo[2], Geoffrey Shaw[1], Veena Tagore[1]

[1]. University Hospital Aintree, Liverpool, United Kingdom, [2]. Trafford Healthcare NHS Trust, Manchester, United Kingdom

Phyllodes tumours are relatively uncommon in the breast with an overall incidence of approximately 1.5% of that of breast carcinoma. However, phyllodes tumour arising in mammary like tissue of the vulva is rare. We report a case of a 33-year-old lady who presented with a 2cm mobile labial swelling, which was subsequently excised. Macroscopic examination of the resected lump revealed a light brown cyst measuring 10x8x7mm.

Histologically, it showed the typical appearance of a benign phyllodes tumour with no atypia or mitoses. On immunohistochemistry, the stromal cells were positive for CD34, vimentin and bcl-2. They were negative for p53 and Ki67. The epithelial cells were positive for bcl-2 and MNF-116 and were weakly positive for p53 and Ki67 (<1% of cells). Oestrogen and progesterone receptors were strongly positive in 40% and 85% of epithelial cells respectively. The epithelial cells also showed moderate positivity with c-Kit (CD117).

Previous case reports revealed that the average age at presentation was between 20 and 39 years and most of the patients had a painless cystic swelling. In one case only, the lesion recurred after 8 months due to its large size (4 cm).

P54

Audit of correlation of cervical cytology and histology performed at different sites

{P} M LEONG, C SEN, J PAWADE
[1]. DEPARTMENT OF CELLULAR PATHOLOGY,SOUTHMEAD HOSPITAL, BRISTOL, United Kingdom, [2]. DEPARTMENT OF HISTOPATHOLOGY, BRISTOL ROYAL INFIRMARY, BRISTOL, United Kingdom

AIM
To assess discrepancies between cervical cytology and histology performed at different sites.
METHOD
Data of cases with more than one grade discrepancy between cervical cytology and histology over a 3-month period was collected. The clinical history, cytology and histology were reviewed. Reasons for any discrepancy were sought.
CONCLUSION
Twenty-six cases showed discrepancy; this figure dropped to 12 following review.
The reasons for cytology overcall included inflammation, HPV changes and squamous metaplasia. Misinterpretations of high-grade dyskaryosis or screening misses were reasons for cytology undercall. The reasons for histology undercall were observer errors related to small foci of dysplasia. HPV changes and immature squamous metaplasia were misinterpreted as CIN resulting in histology overcall.
This audit highlighted the following points that could reduce the number of discrepant cases:
1. Ideally, cervical cytology and histology should be performed at the same site.
2. In smears showing widespread mild dyskaryosis with occasional cells of moderate dyskaryosis, a comment to clarify this in the report text.
3. Every attempt should be made to recognise low-grade dysplasia in the presence of squamous metaplasia, inflammation or koilocytosis.
A re-audit to be performed in one year.

P55

Significance Of Psammoma Bodies In Cervical Cytology

{P} P Joseph, D Rana, J Marshall, M Desai
Manchester cytology centre, Manchester, United Kingdom

The presence of psammoma bodies in cervical smears is a rare finding. These structures have traditionally been associated with malignant ovarian tumours. However, a review of literature reveals that they are frequently associated with benign conditions. The more recent papers suggest that the presence of psammoma bodies should be considered an incidental finding, especially when found in normal cervical smears of asymptomatic women. The feature most predictive of the presence of an associated malignancy is the presence of atypical cells in close association with the psammoma bodies. The association between malignancy and psammoma bodies in cervical smears is also much stronger for postmenopausal women. This article illustrates a case that was flagged due to the presence of psammoma bodies in the cervical smear and subsequent identification of a boderline serous ovarian tumor. The aim of this poster is to alert cytopathologists, to the possibilities, on finding psammoma bodies in cervical smears and the management of these cases.

P56

Unusual Spindle Cell Lesions of the Female Genital Tract (I) Solitary Fibrous Tumour of the Vulva

{P} OJ Biedrzycki[1], H Habeeb[2], N Singh[1], A Faruqi[1]
[1]. The Royal London Hospital, Department of Histopathology, London, United Kingdom, [2]. Homerton Hospital, Department of Obstetrics and Gynaecology, London, United Kingdom

Solitary fibrous tumour (SFT) is a rare tumour first described in the pleura. Originally thought to be derived from mesothelium, new advances have identified a subset of fibroblast like cells as the cell of origin. Recently numerous extrathoracic have been reported; amongst the rarest of such sites is the female genital tract with only eight such cases.

We present a case of an incidental painless 60mm vulval lump, in a 45 year old female. Macroscopically the lesion was an encapsulated white mass with a whorled cut face. Microscopy showed bland spindle cells in a collagenised stroma. Areas of hyper and hypocellularity were noted with abundant hyalinised collagen in the hypocellular foci. Cytological atypia, mitoses and necrosis were absent. On immunohistochemistry the spindle cells were positive for CD34, Bcl2, vimentin and CD99. Stains for cytokeratin, desmin, smooth muscle actin, ER, PR, S100, p53 and HMF2 were negative. Ki-67 staining was <1%.

The features were those of a SFT. This is only the fourth primary case arising in the vulva. We review these together with the other 5 reported cases of primary SFT of the female genital tract, outlining the importance of its recognition, regarding prognosis and the important differential diagnoses.

P57

Unusual Spindle Cell Lesions of the Female Genital Tract (II) Nodular Fasciitis of the Vulva with Unusually Long Clinical History

{P} OJ Biedrzycki, N Singh, A Faruqi
Department Of Histopathology, The Royal London Hospital, London, United Kingdom

Nodular fasciitis (NF) is a benign connective tissue proliferation of uncertain aetiology which usually involves the upper limbs of young adults. It is characterised by a very short clinical history usually measured in weeks. Primary NF of the vulva is very rare and there is a considerable potential for misdiagnosis as a sarcoma. We present a case of NF presenting as a vulval mass in a 35 year old which had been present for just over one year.

Macroscopically, fragmented greyish-white rubbery tissue measuring 3cm was received. Microscopy revealed a typical example of NF with numerous CD68 positive osteoclast-like giant cells. Mitotic figures were present but no atypical mitoses were seen. Due to the uncharacteristically long clinical history a confirmatory expert opinion was sought. There has not been a recurrence in 3 months of follow up.

This is only the thirteenth case of primary NF of the vulva in the literature. In ten of the other cases the diagnosis was not considered by the referring pathologist, despite typical morphological appearances. In this case the unusually long clinical history caused concern and referral. We review all the cases and highlight the need to consider this diagnosis in such an unusual area.

P58

Psammomatous Calcification in a Cervical Leiomyoma – A Case Report

{P} S Katti[1], N Wilkinson[2]
[1]. Leeds General Infirmary, Leeds, West Yorkshire, United Kingdom, [2]. St James University Hospital, Leeds, West Yorkshire, United Kingdom

Calcification is a well-known secondary change in uterine leiomyomata. It is seen in about 4% of cases and is particularly common in postmenopausal women. The calcification tends to be irregular and is presumed to be dystrophic in nature. It frequently manifests macroscopically as a fibroid, which is hard to cut.
Psammomatous pattern of calcification (comprising psammoma bodies like calcific spherules set within interlacing bundles of smooth muscle) in uterine leiomyomata is extremely rare. Only a single case has been previously reported in the literature and this was in association with a uterine serous carcinoma with bilateral ovarian metastases.
We report a case of a cervical leiomyoma with extensive psammomatous calcification in a 77-year-old woman who had an associated ovarian torted endometriotic cyst. There was no evidence of any papillary neoplastic lesion in the uterus, tubes or ovaries.
We conclude that psammoma bodies in leiomyomata are a distinctive and rare pattern of calcification that can occur in the absence of any predisposing neoplasm.

P59

The Value Of Peritoneal Washing Cytology In The Staging Of Gynaecological Malignancy

{P} A K Evans[1], P Joshi[2], D N Rana[2], R J Hale[3], G E Wilson[2], M Desai[2], S M Ismail[1]
[1]. Department of Histopathology, Wythenshawe Hospital, Manchester, United Kingdom, [2]. Departments of Cytopathology and Histopathology, Manchester Royal Infirmary, Manchester, United Kingdom, [3]. Department of Histopathology, Stepping Hill Hospital, Stockport, United Kingdom

Current protocols for staging gynaecological cancers include cytopathological examination of peritoneal washings taken at the time of definitive surgery. We investigated the clinical usefulness of this procedure.
During 2004 and 2005, 140 peritoneal washings were submitted for cytopathological examination in our institutions for staging of 36 ovarian, 101 endometrial and 3 synchronous ovarian/endometrial cancers. The washings contained malignant cells in 39 cases (28%). 35 of these cases had high stage disease – not confined to the organ of origin (i.e. stage 2 or more for ovary and stage 3 or more for endometrial). The other 4 were stage 1C ovarian cancers where there was either rupture or tumour involvement of the capsule. In only 2 of the 39 positive cases the cancer was marginally upstaged by the positive washings – these were ovarian cancers upstaged from 2A/B to 2C.
These findings suggest that peritoneal washing cytology as a routine procedure for staging ovarian and endometrial cancer is of limited clinical value. A larger study is needed to determine whether this procedure should continue to be included in staging protocols for gynaecological cancer.

P60

Atypical Solitary Fibrous Tumour Of The Vulva

{P} M Powari[1], A Oladipo[2], A Malcolm[3], J Tisdale[4], J Mathew[1]
[1]. Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [2]. Department of Gynaecology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [3]. Department of Histopathology, Shrewsbury and Telford Hospital, Shropshire, United Kingdom, [4]. Probus Minor Surgery Unit, Probus, Cornwall, United Kingdom

Aim: To describe a rare case of atypical solitary fibrous tumour (ASFT) arising in vulva of a post-menopausal female.
Case report: A 60 year-old, mother of two, presented with a 15-year history of right sided vulval swelling. She had no other complaints. She had undergone a hysterectomy with right oopherectomy for fibroid and left oopherectomy for a benign cystadenoma in the past. Clinical examination was normal except for a 70mm circumscribed firm mass in the right vulva. Wide local excision was done.
Grossly the specimen was 70mm in diameter, firm to hard in consistency with a bosselated external surface. Microscopically, apart from a focal storiform and focal pericytomatous pattern, the tumour consisted of a cellular spindle to epithelioid cells with moderate cytoplasm and rich vascularity. In pericytomatous areas, there was prominent hyalinisation of vessel walls. Atypical features in form of mitosis up to 5/10HPF, focal tumour necrosis and increased cellularity were seen. The tumour expressed CD34 focally but was negative for desmin, smooth muscle actin, CD31, HMB45 AND MNF116, features in keeping with a cellular form of SFT with atypical features.
Conclusion: Solitary Fibrous tumours (SFT) are uncommon tumours originally described in pleura but can be seen elsewhere. Its occurrence in vulva is quite rare. Atypical and malignant forms are recognised although distant metastasis is uncommon.
We have described a rare case of atypical vulval SFT in a post- menopausal female. This tumour needs to be distinguished from more common tumours of this site

P61

Retroperitoneal Mucinous Cystadenoma In A Teenage Girl

{P} M Powari[1], A Oladipo[2], S Thorogood[3], J Mathew[1]
[1]. Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [2]. Department of Gynaecology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [3]. Department of Clinical Imaging, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom

Aim: To describe a case of mucinous cystadenoma arising in the retroperitoneum of a 19-year-old girl.
Case report: A 19 year-old girl presented with abdominal discomfort of a few weeks duration. A mass was palpable in the lower abdomen. Clinical examination was otherwise unremarkable. A USG showed a well-circumscribed cystic tumour which was confirmed to be in the retroperitoneum by a CT scan. CA125 levels were normal.
The tumour was excised and at surgery, both ovaries and uterus were found to be normal. The tumour was a multi loculated cystic mass measuring 135x85x75mm with a smooth outer and inner surface. It was lined by a tall columnar type mucin producing epithelium. In some area the lining was flat cuboidal. No tufting, nuclear atypia or mitosis was seen. The columnar cells were CK7 and CK20 positive while the cuboidal cells were negative. These cuboidal cells were calretinin positive suggesting mesothelial origin. Stromal cells expressed oestrogen and progesterone receptors akin to mucinous tumours of the ovary. The histologic features were of a mucinous cystadenoma and immunohisto-chemistry supported a metaplastic process with differentiation of mesothelium to mucinous type.
Conclusion: Retroperitoneal cysts are uncommon, usually asymptomatic lesions with most cases occurring in the fourth decade. We have described an uncommon retroperitoneal mucinous cystadenoma in a teenage girl. The morphology is quite similar to those arising in ovary. Histiogenetically, it appears to arise by mucinous metaplasia of a mesothelial cyst.

P62

A Rare, Uterine Case Of Extra-Pulmonary Lymphangioleiomyomatosis

{P} M Powari[1], L Hirchowitz[2], P Callen[3], J Mathew[1]
[1]. Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [2]. Department of Histopathology, Southmead Hospital, Bristol, Avon, United Kingdom, [3]. Department of Gynaecology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom

Aim: To describe a rare case of lymphangioleiomyomatosis (LAM) affecting uterus in a patient with tuberous sclerosis.
Case report: An 80-year-old female with tuberous sclerosis, presented with post-menopausal bleeding. She was being treated for chronic renal failure. She had had bilateral renal angiomyo-lipomata for which she had undergone right nephrectomy in the past.
On examination she had a bulky uterus. An endometrial pipelle showed atypical hyperplasia for which a total abdominal hysterectomy and bilateral salpingo-oophorectomy were done. Histology confirmed atypical endometrial hyperplasia along with adenomyosis. The myometrium appeared unusual with presence of nodules of spindle shaped cells arranged in irregular fascicles, some of which were related to complex lymphovascular spaces. Nodular areas of epithelioid cells and areas with clear cell morphology were also seen, separated by thin walled variably sized blood vessels. In addition, similar lesions were present within the right ovary and parametrium. The nodules were diffusely positive for desmin, smooth muscle actin and H-caldesmon. HMB45 was very focally positive. CD10 was negative in these areas. The overall histological appearances were those of uterine LAM involving also the right ovary and parametrium.
Conclusion- LAM is a rare disease and it is still rarer to affect female genital tract. It affects females in reproductive age group and is usually associated with tuberous sclerosis.
We have described a rare case of extra-pulmonary LAM affecting uterus and ovary in a patient with tuberous sclerosis. The histologic appearances are characteristic; HMB 45 stain can be focally positive or completely negative in these lesions.

P63

Vaginal angiomyofibroblastoma; a case report and a review of the literature.

{P} M Taylor, H Shaikh
Kings College Hospital, London, United Kingdom

Angiomyofibroblastoma (AGMF) is a rare lesion in the vulvo-vagina which poses problems for the surgical pathologist due to the wide variety of morphologicaly bland lesions that may involve this area and their overlapping morphological features. We report a case of vaginal AGMF and discuss the clinico-pathalogical features and recent developments in this area.

P64

Leptomycin B Induces Apoptosis in Keratinocytes Containing the Whole HPV 16 Genome

C Jolly, S Talukdar, L Gray, {P} C S Herrington
University of St Andrews, St Andrews, Fife, United Kingdom

We have shown previously that leptomycin B (LMB) induces apoptosis efficiently in primary human keratinocytes expressing the HPV 16 E6 and E7 genes. However, to be a useful treatment for HPV-associated neoplasia, this agent needs to be effective in cells expressing the whole viral genome. We therefore assessed the ability of LMB to induce apoptosis of two derivatives of the W12 cell line, one of which contains the whole HPV 16 genome in episomal form (W12E), the other containing integrated HPV 16 (W12I). Given our previous demonstration that LMB induces apoptosis most efficiently in PHKs expressing only the E7 gene, we hypothesised that W12I cells, which express a higher level of E7 protein than W12E cells, would be most sensitive to this treatment. W12 cells were analysed in both monolayer and raft culture and apoptosis was identified by immunocytochemistry for M30 and activated caspase 3. The effect of LMB on monolayer cells was also assessed using the colony survival assay. Immunocytochemistry demonstrated more widespread induction of apoptosis by LMB in W12E than in W12I, using both monolayer and raft culture. The colony survival assay also demonstrated better survival of W12I than W12E after LMB treatment. These data indicate that LMB can induce apoptosis in keratinocytes containing the whole viral genome and that factors other than E7 expression govern the response to LMB treatment.

P65

Endometrial Morphology With The Use Of The Mirena Coil

S CHAUDHRI[2], R HEJMADI[1], {P} R GANESAN[1], TP ROLLASON[1]
[1]. BIRMINGHAM WOMEN'S HOSPITAL, BIRMINGHAM, United Kingdom, [2]. QE HOSPITAL, BIRMINGHAM, United Kingdom

Introduction:The Mirena coil is an intrauterine hormone release device used in contraception, menorrhagia and to counter HRT. Objective: The object of this study was to illustrate the effects of this device on endometrial morphology associated with Mirena coil. Materials and methods: Endometrial morphology was assessed, particularly, for the appearance of glands, endometrial stroma, presence or absence of inflammation and ulceration and additional features of note. Results: Atrophic glands or secretory glands with interspersed atrophic glands were the dominant feature. A variety of metaplasias were noted. Nuclear atypia when present was predominantly random with occasional enlarged, hyperchromatic nuclei. Most cases demonstrated sheets of plump, polygonal, pseudodecidualised stromal cells. In some, the stroma exhibited extracellular mucinous material with a basophilic, myxoid appearance. Other changes included ulceration, infarction, superficial micropapillary change and presence of dense hyaline nodules. Discussion: The presence of hyaline nodules is an addition to the spectrum of changes recorded. In one case, there was extensive mucoid stroma mimicking a a myxoid sarcoma or mucinous carcinoma. Conclusion: We believe that with ever increasing Mirena usage it is important that the general pathologist is aware of the morphological features associated with its use.

P66

Antigen Processing And Correlation With Immunological Response In Vulval Intraepithelial Neoplasia

{P} K SINGH[2], Y YEO[3], H HONEST[1], R GANESAN[1], DM LUESLEY[2]
[1]. BIRMINGHAM WOMEN'S HOSPITAL, BIRMINGHAM, United Kingdom, [2]. CITY HOSPITAL, BIRMINGHAM, United Kingdom, [3]. WALSGRAVE HOSPITAL, COVENTRY, United Kingdom

Objective.: To study the antigen-presenting cells and co-stimulatory factors in different grades of vulval intraepithelial neoplasia. Material and methods.: Histology specimens were obtained from women who had undergone vulval biopsies for VIN. The CD1a (Langerhans cell/antigen-presenting cell marker) and co-stimulatory factors-HLA class 2 antigens (LN3) and the adhesion molecule (CD54)-were semi-quantitatively analyzed in all the specimens. Pearson chi-squared test was used for statistical analysis. Results.: There was an inverse correlation between CD1a and severity of VIN (pearson chi-squared = 26.876, p = 0.001). Qualitatively, there was a basal location of CD1a-positive cells in normal epithelium. There was no statistical significance in the distribution of LN3 and CD54 in different grades of VIN. Conclusion: this study shows an alteration in the numbers and spatial arrangement of CD1a-positive langerhans/antigen-presenting cells in different grades of VIN. Reduction in cd1a expression in high grade VIN may reflect the inability of the host to mount an adequate immune response due to reduced antigen presentation.

P67

PATHOLOGICAL CHARACTERISATION OF CERVICAL SMALL CELL CARCINOMA WITH CLINICAL FOLLOW UP

{P} A El-Ghobashy[1], M Hewitt[1], G Hall[2], N Wilkinson[3]
[1]. Department of Gynaecological Oncology, St James's Unoversity Hospital, Leeds, United Kingdom, [2]. Department of Medical Oncology, St James's Unoversity Hospital, Leeds, United Kingdom, [3]. Department of Pathology, St James's Unoversity Hospital, Leeds, United Kingdom

Introduction: Cervical small cell carcinoma (CSCC) is a rare and aggressive tumour. There is a lack of consistency in the management strategies for this tumour. It is often diagnosed at an advanced stage; its prognosis is generally poor and not infrequently seen in association with other tumours.
Patients and Methods: A retrospective review of cases of carcinoma of the cervix was conducted to identify CSCC over a 10-year period. The cervical tumours stained routinely by H & E were examined in order to establish the diagnosis and sections were stained immunohistochemically with a panel of neuroendocrine markers to confirm the diagnosis. Clinical follow-up data was retrieved for all patients
Results: 19 patients were identified at our centre with the diagnosis of CSCC. The mean patients' age was 49.2 years (range: 24-85 years). Eight tumours (42.1%) were stage I, 5 (26.3%) were stage 2 and 6 were of advanced stage. The median overall survival was 28.1 months. A significantly better survival probability was found in patients with stage I disease compared with other stages.
Conclusion: CSCC is an uncommon tumour with poor outcome, which requires a multi-disciplinary approach in treatment. Our series is the largest cohort of CSCC in the UK.

P68

Optimisation of sampling omentum in ovarian neoplasia

{P} T Duncan, R hammond, S Deen
Queen's Medical Centre, Nottingham, United Kingdom

Omental sampling for suspected ovarian cancer is recommended as part of FIGO staging but there are no published guidelines as to the number of blocks required for accurate assessment.
In an attempt to assess the most accurate and also cost effective strategy we audited 100 cases of ovarian neoplasia, with omental biopsy, in our department. This included 59 benign, 13 borderline and 28 malignant cases.

The median ratio was one block per 4.5cm with little variation when dividing the tumours according to malignant potential. Microscopy confirmed the macroscopic benign or malignant impression. All the blocks from the latter were positive for tumour.
Sampling was more generous when the macroscopic appearances of the omentum were equivocal (4%), in all these cases microscopic findings were negative.

It appears that macroscopically involved omentum in malignant cases only require a single block for confirmation. In benign cases a single block should be taken for completeness.
Multiple omental sampling should be reserved for those malignant/borderline tumours in which the omentum is macroscopically normal to rule out peritoneal implants and avoid under staging. Adopting this rationale would lead to a 65% reduction in the number of blocks taken, with a saving of £13 pounds per case.

P69

What factors play a role in achieving complete excision of CIN? An audit project.

{P} S Deen, T Duncan, M Singh, R Hammond
Queens Medical Centre, Nottingham, United Kingdom

Aim; To identify the factors that might have a bearing on completeness of excision of LLETZ.
Method; 250 LLETZs submitted as one piece of cervix in 2005 were reviewed. All cases were taken from the departmental archive, a secondary & tertiary referral practice. 37(15.8%) CIN1, 83 (35.5%) CIN2 & 114 (48.7%) CIN3. Specimens were embedded as one slice per cassette with special attention to the lateral ectocervical margin. The latter was embedded with the outermost margin face down.
Results; Excision was complete in 48.8%, incomplete in 45.6% and inconclusive in 5.6%. Incomplete excision was mostly reported in high grade CIN with a breakdown of 61% CIN3, 29.5% CIN2 and 9.5% CIN1. Incomplete excision was mainly at the ectocervical margin (31%) and in particular the lateral side of that margin (30% of the involved ectocervical margins). 11% of LLETZs had incomplete excision at the endocervical margin. There is a tendency for incomplete ectocervical excision in cases with a transverse os rather than a circular os. Size of the loop was not related to the various degrees of CIN.
Conclusion:1) By using the method described for assessment of the lateral margin, rather than the more common practice where more than one slice of tissue may be presented in one cassette, we have identified more loops with incomplete excision.

2) Probably a slightly wider excision is required to encompass about 1mm of apparently normal cervix in cases of CIN3 and perhaps those with a large transverse os.

P70

A Validated Method for Quantifying Macrovesicular Hepatic Steatosis in Chronic Hepatitis C.

{P} TH Boyles, S Johnson, N Garrahan, AR Freedman, GT Williams
University Hospital of Wales, Cardiff, United Kingdom

Hepatic steatosis is increasingly seen as an important prognostic factor in chronic hepatitis C infection (HCV). The commonly used semi-quantitative method of measuring steatosis is based on a study that excluded patients with HCV. Potentially useful methods of quantifying steatosis using computer assisted morphometric analysis have not been validated against a proposed gold-standard other than the method they were intended to replace. We present a novel method of computer assisted morphometric analysis of steatosis and propose a gold standard based on manual measurements. The manual method is time consuming but shows little inter-observer error and the mean value of three observations by separate investigators is proposed as the gold-standard. The computer assisted method has a single interactive step which shows minimal inter-observer variation. When compared to the gold-standard it accurately identifies biopsies with less than 1% steatosis (7 out of 7) and predicts the value for biopsies with greater than 1% steatosis with narrow confidence intervals (geometric mean ratio 0.85 with 95% confidence intervals 0.77-0.95). This novel method of computer assisted morphometric analysis is suitable for future research into steatosis in HCV and may be used to re-analyse previous studies. The semi-quantitative method remains appropriate for clinical purposes.

P71

Heterotopic pancreatic tissue at the ampulla of Vater causing bile duct obstruction

{P} E A Husain, A Biswas, A T Abraham, R Feakins
Barts and the London NHS Trust, London, United Kingdom

Heterotopic pancreatic tissue at the ampulla of Vater is an uncommon finding, and is a very rare cause of biliary obstruction and jaundice. A 47 year old man was admitted to this institution with obstructive jaundice. His radiological profile was highly suggestive of a malignant obstructing lesion at the distal end of the common bile duct. Accordingly he underwent pancreatoduodenectomy. Subsequent macroscopic examination revealed a well circumscribed nodule measuring 15×14×7mm immediately adjacent to the ampullary orifice. On histological examination, the nodule was composed of heterotopic pancreatic tissue. The distal common bile duct showed marked inflammation and fibrosis. There was no evidence of dysplasia or malignancy either in the bile ducts or in the pancreas. No other causes of obstruction were identified. This case serves as a reminder that benign lesions, including heterotopic pancreatic tissue, should be included in the preoperative differential diagnosis of causes of obstructive jaundice.

P72

Survivin Expression in Pancreatic Intraepithelial Neoplasia (PanIN)

{P} U.K. Bhanot, P. Möller, C. Hasel
Department of Pathology, University of Ulm, Ulm, Germany

Pancreatic ductal adenocarcinoma(PDA) is one of the most aggressive gastrointestinal cancers, thought to arise from noninvasive precursors - pancreatic intraepithelial neoplasi(PanIN). Aberrantly prolonged cell survival due to apoptosis suppression is likely to contribute to carcinogenesis and carcinoma progression where the inhibitor of apoptosis proteins(IAPs) may play an important role. IAPs specifically inhibit caspases 3,7, and 9 and prevent apoptosis. Survivin is a unique member of the IAPs family that is expressed in most human cancers including PDA but is not expressed in most normal adult tissues.
To measure survivin transcript levels in normal pancreatic ducts, PanINs, and PDA, we used laser capture microdissection and real-time PCR. Survivin protein expression in normal pancreatic ducts, PanINs, PDA, and lymph node metastases was evaluated by immunohistochemistry. In microdissected tissues, we found a steady and close to exponential increase in survivin transcript levels from low grade lesions (PanINs-1) to high grade lesions (PanINs-2 and 3) and further to PDA. This observation was strictly mirrored by survivin protein expression. In addition, survivin was localized to the nucleus in high grade lesions (starting at PanIN-2 stage), PDA and nodal metastases, suggesting that nuclear translocation of survivin may be an early event in transformation to malignancy.

P73

Islet Cell Survival in Chronic Pancreatitis :Role of NFκB and Inhibitor of Apoptosis Proteins (IAPs)

{P} U.K. Bhanot, C. Hasel, P. Möller
Department of Pathology, University of Ulm, Ulm, Germany

In advanced chronic pancreatitis (CP), islets are preserved even in the midst of scarring. We recently showed in CP local production of interferon(IFN)γ, transforming growth factor(TGF)β, and death receptor ligand TRAIL, along with functional death receptor neo-expression and apoptosis in exocrine but not in endocrine cells. However, islets are strongly induced for TRAIL-receptor(R)-4 lacking the functional death domain. TRAIL-R4 signaling in T-cells induces NFκB which activates anti-apoptotic programs. Here, we demonstrate that in insulinoma cells CM, TGFβ/ IFNγ/TRAIL in combination induced TRAIL-R4 surface expression. TRAIL/IFNγ, upregulated NFκB subunits and its target gene survivin while down-modulating IκBα mRNA. RelA transcriptional activity increased upon stimulation with IFNγ, and IFNγ/TRAIL. In situ, normal pancreatic epithelia had low mRNA levels of NFκB subunits. These were higher in parenchymal areas of CP with severe fibrosis and highest in islets. NFκB regulated proteins IκBα, survivin, and another apoptosis inhibitor - cIAP1, were found in corresponding sites, again, at highest levels in islets surrounded by fibrosis. In conclusion islets in CP not only evade immune attack by non-exposure of functional death receptors in presence of TRAIL-R4 but additionally neo-express NFκB and its target genes survivin and cIAP1 to protect themselves from apoptosis.

P74

A Ciliated Hepatic Foregut Cyst with Thyroid Tissue

{P} A.R. Darne, B. Haugk
Department of Histopathology, The Royal Victoria Infirmary, Newcastle-upon-Tyne, United Kingdom

We describe a rare case of a ciliated hepatic foregut cyst with thyroid tissue in a 45-year-old woman who presented with persistent right upper quadrant pain, requiring emergency admission.
Liver function tests and a full blood count were within normal limits.
Ultrasound imaging revealed a septate intrahepatic cyst. Subsequent computed tomography showed a 3 x 3 x 4cm well defined, non-enhancing cyst containing high density material in hepatic segment IV, with no definite malignant features. A left hemi-hepatectomy was performed following multi-disciplinary team discussion and patient request.
Macroscopic examination revealed a multilocular cyst adjacent to the gallbladder. Microscopically the cyst was lined by simple columnar ciliated epithelium with focal goblet cells. No multi-layering or solid areas were present. The sub-epithelial tissue contained sero-mucinous glands and smooth muscle. Of particular interest was a single focus of thyroid tissue within the cyst wall.
Ciliated hepatic foregut cysts are well documented in the literature, being derived from embryologic foregut remnants. However, the finding of thyroid tissue within the cyst wall has not been previously described. Thyroid tissue further confirms the histogenesis of the cyst as being foregut derived. Diagnostically, this also necessitated distinction from a hepatic teratoma.

P75

Correlation of Histological Features on Liver Biopsy in Patients with Alcohol-Induced Cirrhosis

{P} CL Verrill, H Markham, NJ Carr, N Sheron
Southampton University Hospitals NHS Trust, Southampton, United Kingdom

Introduction:
There is very little understanding of the correlation between the liver biopsy appearances in alcohol-induced liver cirrhosis and the wider clinical prognosis. Liver biopsies are usually given a grade of severity of cirrhosis such as 'early', 'established' or 'end-stage'. There is very little research into the clinical significance of these labels. For example does 'end-stage' mean a death sentence? The Laennec grading system is the only published grading system for alcohol-induced liver cirrhosis. Biopsies are divided into 4A (mild), 4B (moderate) and 4C (severe) cirrhosis.
Methods:
Retrospective study of 107 biopsies of alcohol-induced liver cirrhosis at Southampton General Hospital taken between 1st January 1995 and 31st December 2000. Cases were scored for Laennec grade, percentage of biopsy occupied by fibrosis, grade of steatohepatitis, grade of steatosis and pericellular fibrosis.
Results:
Significant Pearson correlations were identified between percentage of fibrosis and Laennec grade (0.776), pericellular fibrosis and steatohepatitis (0.416), grade of steatosis and grade of steatohepatitis (0.282)
Conclusions:
Pericellular fibrosis was significantly correlated with grade of steatohepatitis which provides definitive evidence for this observed link. Percentage of fibrosis correlates significantly with the Laennec Grade and is a novel and clinically relevant way to assess liver biopsies of alcohol induced cirrhosis.

P76

Hepatitis C - What Constitutes An Adequate Liver Biopsy?

{P} S Mussunoor, M Sheehan, A Fraser
Aberdeen Royal Infirmary, Aberdeen, United Kingdom

Introduction: Adequacy of liver biopsy material received for histological examination has been the subject of a great deal of debate. The minimum number of portal tracts required in a biopsy to ensure that the biopsy is representative of the whole liver and allows confident staging has been established for primary biliary cirrhosis and graft rejection. However, for diseases like hepatitis C minimum biopsy length criteria are less clear. Bedossa et al (2003) proposed an ideal minimum length of 25mm to allow accurate assessment of fibrosis in 75% of biopsies.
Methods: 179 consecutive liver biopsies performed to assess the histological stage of patients with chronic hepatitis C infection between the periods 1999 to 2005 were audited. The length of the biopsy and number of fragments received were documented. Necroinflammatory disease and architecture were assessed using the modified histological activity index (Ishak scoring system).
Results: The mean total biopsy length was 15.8mm. Median biopsy length was 14mm. Size of single largest core of tissue was 33mm. Largest total biopsy length was 42mm. The biopsy length most frequently fell in the 10-15mm range. The diagnosis of cirrhosis was made in 22 biopsies (13 %). The majority of the diagnoses were made on biopsies less than or equal to 15 mm. Twenty three (13%) cases showed stage 3 / 4 fibrosis.
Conclusions: Majority of biopsies fell in the 10-15mm range. Confident diagnosis and staging of fibrosis was made in 63% and 69% of cases respectively, on biopsies which were less than or equal to 15 mm in total length. Only 2 cases of cirrhosis were diagnosed on biopsies measuring more than 25mm. In our experience, only in a small minority of patients with hepatitis C infection was a liver biopsy of greater than 25 mm obtained. Despite this we could make adequate assessment of necroinflammatory disease as well as fibrosis in the majority of the cases.

P77

Promotion Of Tumourigenesis In Mucinous Cystic Tumours Of The Pancreas By Elevated Hormone Levels In Pregnancy: Two Case Reports

{P} M G Deheragoda, M Rodriguez-Justo, Z Amin, S Khorsandi, A Shankar, C Russell, M R Novelli
University College Hospital, London, United Kingdom

Background : Mucinous cystic neoplasms of the pancreas show an ovarian-like stroma with epithelial lined cystic spaces. Adenocarcinoma is typically found in patients aged 50 years and over.
We report two cases of mucinous cystadenocarcinoma of pancreas arising in patients a decade younger than the typical age expected for the development of carcinoma.
Both cases occurred in 40 year old women who underwent distal pancreatectomy in the postpartum period. Histology of both tumours demonstrated mucinous cystadenocarcinoma of the pancreas. There was positive immunohistochemical staining of the tumour stroma with ER, PR, HCG, inhibin and calretinin, consistent with steroid hormone responsiveness and biosynthetic function of the ovarian-like stroma. The epithelial component showed a high proliferation rate with MIB1.
Discussion: The finding of mucinous cystadenocarcinoma of the pancreas in relatively young patients in the post partum period raises the possibility that high hormone levels acting on the ovarian type stroma in pregnancy may have an effect by promoting epithelial tumourigenesis.

P78

A Cytogenetically Proven Desmoplastic Small Round Cell Tumour In Pancreas

B. Haugk, N. Brown, A. Ryan, {P} M. Jain
Department of Cellular Pathology, Royal Victoria Infirmary, Newcastle upon Tyne, United Kingdom

We describe an unusual case of a cytogenetically proven desmoplastic small round cell tumour (DSRCT) presenting as a mass in the tail of pancreas in a 35 year old lady.

A left upper quadrant mass was detected in a lady on fertility treatment. Computed tomography revealed a large mass in intimate relation with the tail of the pancreas. Liver masses and polycystic kidneys were also detected.

An initial core biopsy was not conclusive. A larger biopsy was sent fresh to allow cytogenetic analysis. Microscopic examination revealed a tumour infiltrating pancreas composed of desmoplastic stroma and irregular islands of small blue cells, some with intracytoplasmic inclusions.

Immunohistochemically the tumour was positive for low molecular weight cytokeratins and desmin with characteristic dot positivity. Cytogenetic analysis, using interphase FISH, showed SPLIT signal for EWS and WT1 "break-apart" probes consistent with translocation t(11;22)(p13;q12) typical of DSRCT. RT-PCR, using EWS and WT1 primers, generated a fusion transcript which was confirmed by direct sequencing of the PCR product.

DSRCTs are rare tumours occurring in children and young adults with a male predominance and a predilection for serosal surfaces, rarely involving viscera. The pancreatic involvement is unusual. The cytogenetic analysis has provided unequivocal proof.

P79

Immunohistochemistry Requests And Their Utility In The Diagnosis Of Metastatic Tumours Of The Central Nervous System

{P} M N D'Cruz, A Chakrabarty
Leeds General Infirmary, Leeds, West Yorkshire, United Kingdom

Introduction:
Metastatic tumours account for 24% of intracranial and 5% of intraspinal neoplasms, the common primary sites being respiratory tract, breast and melanoma. Oncologists can now tailor patient management depending on the diagnosis of a definitive primary tumour site.
Aim:
To evaluate the request patterns and utility of immunohistochemistry in the diagnosis of metastatic tumours of the central nervous system.
Results:
In this 8-year retrospective study, we audited a total of 130 cases with a histological diagnosis of metastatic tumour. The cases were retrieved using the appropriate SNOMED codes. Immunohistochemistry was done in 50 cases. The most useful markers were neural markers, melanoma markers, prostate markers, germ cell markers, CA-125 and epithelial markers. In 10 cases the primary site was unascertained despite immunohistochemistry. In 2 cases, the diagnosis of the primary tumour was altered in the light of immunohistochemistry results.
Conclusion:
There was no consistent panel of immunostains used for investigation of metastatic tumours. It is recommended that a wider panel of immunostains as well as a mandatory workup of all cases with a suspected or unknown primary tumour be performed.

P80

Extraventricular Neurocytoma In A 21 Year Old Male: A Case Report

{P} M J Alemkunnapuzha, A Shivane, A Ismail
Leeds General Infirmary, Leeds Teaching Hospitals NHS Trust, Leeds, West Yorkshire, United Kingdom

The neurocytic tumours are uncommon neuronal tumours principally affecting young adults. The central neurocytoma, most common of the neurocytic tumours, are typically located in the lateral and/or third ventricle and usually have a benign outcome. We report a rare and unusual tumour resembling central neurocytoma but lacking the characteristic intraventricular location. The differential diagnosis includes oligodendroglioma, clear-cell ependymoma and ganglioneurocytoma. The immunohistochemistry and electron microscopy confirmed the neurocytic nature of this tumour. Such neurocytic neoplasms located within the brain parenchyma are referred to as extraventricular neurocytomas. We emphasize the role of morphological, immunohistochemical and electron microscopic studies in recognizing this rare tumour.

P81

Astrocytoma With Unusual Histological And Cytogenetic Profile In A Ten Year Old Girl- A Case Report.

{P} AQ Ganjifrockwala, A Shivane, A Ismail
Leeds General Infirmary,Leeds Teaching Hospitals NHS Trust, Leeds, West Yorkshire, United Kingdom

We report a case of low-grade glioma with histological differential diagnosis of pilocytic astrocytoma, pilomyxoid astrocytoma and diffuse astrocytoma in a child. The tumour was in the left posterior medial temporal lobe lesion. Histological evaluation revealed a fibrillary tumour composed of a biphasic pattern with hyper and hypocellular areas, single mitosis and a myxoid background. Immunohistochemistry showed a strong positivity for GFAP and occasional positivity for neuronal markers suggesting infiltration. MIB-1 proliferation index (PI) was 5-7%. The above findings suggested a low-grade glioma but in view of the MIB-1 PI we proposed, it might show an aggressive behaviour. Cytogenetic analysis revealed loss of 1p and 19q, which are consistent with a diagnosis of a low-grade astrocytoma and loss of 9p (presumed loss of p16) that is generally associated with a higher grade of the disease.

The above findings suggested a low-grade astrocytoma histologically with IHC and cytogenetics suggesting an aggressive behaviour.

The purpose of this case report is to emphasise the need for more research to understand the histological criteria for grading and biological behaviour of gliomas in children.

P82

Audit of Muscle Biopsy Practice: An Evolving Service

{P} S M L Paine, M S Damian, M Daruwalla, N W Yii, A Fletcher
Leicester Royal Infirmary, Leicester, United Kingdom

Introduction: Skeletal muscle biopsy is a valuable tool in the assessment of neuromuscular disorders. Previously in this centre, general neurologists referred patients for a needle biopsy of the quadriceps or biceps muscles that was performed and reported by a general histopathologist. Following service development, a specialist neurologist now refers patients, often after neuromuscular magnetic resonance imaging, for an open biopsy that is performed by plastic surgeons and reported with a visiting specialist pathologist. Has the service improved and maintained pace with diagnostic advances?
Methods: A retrospective review of reports from 1987 to 1992 and 2000 to 2005 recording diagnostic yield and factors on which it depends (patient selection, use of ancillary tests, biopsy technique, muscle biopsied and amount of tissue taken). Biopsy practice in specialist centres was surveyed by postal questionnaire.
Results: 168 reports were reviewed. Since the service changes, the proportion of normal or diagnostic biopsies has increased from 66% to 87%. Non-specific biopsies have fallen from 34% to 8%. Biopsy practice varies considerably between specialist centres. The range of diagnoses will be presented along with radiological correlation.
Conclusion: It is possible to produce a diagnostically robust muscle biopsy service even in a centre without on-site specialist histopathologists.

P83

Molecular Characterization of c-KIT in Gliomas

AL Gomes[1], {P} JS Reis Filho[2], MB Lambros[2], A Martins[1], F Pardal[3], JM Lopes[4], RM Reis[1]
[1]. Life and Health Sciences Research Institute (ICVS), School of Health Sciences, University of Minho, Braga, Portugal, [2]. The Breakthrough Breast Cancer Research Centre, Institute of Cancer Research, London, United Kingdom, [3]. Department of Pathology, Hospital S. Marcos, Braga, Portugal, [4]. IPATIMUP, Porto, Portugal

Gliomas are the most common primary brain tumours. Despite the therapeutic advances, the majority of gliomas do not respond either to chemo or radiotherapy. C-kit is a class III receptor tyrosine kinase (RTK) frequently involved in gastrointestinal stromal tumour (GIST) tumorigenesis. Activating mutations of KIT are predictive of therapy response to RTK inhibitors (e.g. Imatinib). C-kit is expressed not only during brain development but also in a subset of glioma cell lines. To determine whether patients with glial tumours might benefit from therapy with RTK inhibitors, we analysed c-kit expression and its correlations with KIT gene amplification and activating gene mutations in a large series of gliomas. Immunohistochemistry for c-kit was performed in a series of 75 gliomas of various grades. C-kit positive tumours were further subjected to c-kit mutation analysis (exons 9, 11, 13 and 17), and gene amplification analysis by chromogenic in situ hybridisation (CISH) with in-house generated BAC probes. Immunopositivity was detected in 13/75 (17.3%) of cases. No c-kit mutations were identified. CISH analysis revealed KIT amplification in 4/13 (30.8%) of c-kit positive cases. Our results suggest that a subgroup of patients with gliomas may benefit from therapy with anti-c-kit RTK inhibitors.

P84

Aberrant Expression of the Rb Pathway Proteins in Soft Tissue Sarcomas

{P} M Sabah, R Cummins, M Leader, E Kay
Royal College of Surgeons in Ireland, Dublin, Ireland

Cell cycle regulation depends on a fine balance between cyclins, cyclin-dependent kinases and cyclin-dependent kinase inhibitors that block the cycle progression. Alterations of the cell cycle regulators are a common feature of many malignant tumours and some have been shown to have prognostic significance.
152 cases of different types of soft tissue sarcomas were evaluated for alterations of cell cycle regulator proteins that control the cell cycle progression from G1 to S phase and govern the Rb pathway. Immunohistochemical stains for proteins Rb, E2F1, cyclin D1, CDK4, CDK6, p16 and p27KIP1 were carried out on tissue microarrays. The relationship between the expression of these proteins and the histologic grade of the sarcomas was assessed. Altered expression for Rb and p16 proteins was identified in 67.8% and 65.1% of the cases, respectively. Overexpression of E2F1, cyclin D1, CDK4 and CDK6 was detected in 50.7%, 24.3%, 92.1% and 10.5%, respectively. Overexpression of E2F1 was associated with altered expression of Rb protein. Overexpression of cyclin D1, CDK4 and CDK6 showed an association with normal Rb expression. CDK6 expression revealed a positive correlation with the histological grade of the sarcoma and p27KIP1 expression was inversely correlated with sarcoma grade. These results suggest that alterations of the Rb pathway proteins are common in soft tissue sarcomas and may participate in their tumourigenesis. CDK6 and p27KIP1 showed correlation with the histological grade of the sarcomas, suggesting that these proteins could be used as prognostic markers.

P85

Immunoreactivity of p53, Mdm2, $p21^{WAF1/CIP1}$, Bcl-2 and Bax in soft tissue sarcomas: Correlation with histologic grade

{P} M Sabah, R Cummins, M Leader, E Kay
Royal College of Surgeons in Ireland, Dublin, Ireland

Tumour growth depends on two distinctive pathways: cell proliferation and apoptosis. The p53 pathway is an important regulator of the cell cycle as it triggers growth arrest or leads to apoptosis in response to cellular stress and therefore is commonly targeted during tumourigenesis. Apoptosis is also controlled by the Bcl-2 family, which includes proapoptotic and antiapoptotic proteins.
152 cases of different types of soft tissue sarcomas were analysed for the expression of proteins that are involved in the p53 pathway and apoptosis. The cases consisted of 54 low-grade, 40 intermediate-grade and 58 high-grade sarcomas. Immunohistochemical staining for $p21^{WAF1/CIP1}$, p53, Mdm2, Bcl-2 and Bax proteins was carried out on tissue microarrays. Nuclear reactivity for p53 was detected in 49 cases (32.2%). Overexpression of Mdm2 was found in 18 cases (11.8%) and $p21^{WAF1/CIP1}$ immunostaining was seen in 28 tumours (18.4%). p53 and $p21^{WAF1/CIP1}$ expression correlated with the tumour grade (low grade, 5.6% and 3.7%; intermediate grade, 22.5% and 20%; high grade, 63.8% and 31%, respectively). Expression of Bax protein was a common finding in soft tissue sarcoma cases. It was detected in 141 cases (92.8%). Bcl-2 was identified in 59 tumours (38.8%) and was more prevalent in high-grade sarcomas (low grade, 25.9%; intermediate grade, 32.5%; high grade, 55.2%). It was concluded that alterations in the p53 pathway and genes that regulate apoptosis are common events in soft tissue sarcomas. The expression of p53, $p21^{WAF1/CIP1}$ and Bcl-2 is closely associated with the histologic grade of the tumour and therefore these proteins may be used as prognostic markers.

P86

Immunolocalisation Of Matrix Metalloproteinase 7 In Human Intervertebral Discs (IVD).

{P} CL Le Maitre, AJ Freemont, JA Hoyland
University of Manchester, Manchester, United Kingdom

Matrix metalloproteinase 7 (MMP 7) is known to cleave the major matrix molecules found within the IVD. However MMP 7 has only been investigated in herniated discs. In this study we investigated the localisation of MMP 7 in human non-degenerate, degenerate and prolapsed intervertebral discs Immunohistochemistry was performed to localize MMP 7 in 42 paraffin embedded human IVDs consisting of 11 non-degenerate, 14 low grade degenerate, 8 severe degenerate and 9 prolapsed discs.
MMP 7 immunopositive cells were seen in the nucleus pulposus (NP) and inner annulus fibrosus (IAF), with little immunopositivity in the outer AF. Non-degenerate discs showed very low numbers of immunopositive cells (~6%), with a significant increase in the proportion of immunopositive cells in discs with severe grades of degeneration (~50%, $P<0.05$). Prolapsed discs showed higher numbers of cells with immunopositivity (~22%) compared to non-degenerate discs but lower levels than that seen in severe degenerate grades. This study has shown that MMP 7 is produced by the cells of the NP and IAF of human IVDs, and that MMP 7 immunopositivity is increased with degeneration. This suggests that MMP 7 plays an important role in the degradation of the extracellular matrix during disc degeneration, and LBP.

P87

The Identification of Large Conductance Potassium (MaxiK) Channels in Human Intervertebral Disc (IVD) Cells

{P} SM Parker, D Purmessur, SJ Millward-Sadler, CL LeMaitre, AJ Freemont, JA Hoyland
The University of Manchester, Manchester, United Kingdom

Classically associated with membrane potential control in excitable cells, the calcium-activated MaxiK channel has been identified in cells of connective tissues, namely chondrocytes and osteoblasts. However, no studies have identified the expression of this channel (or any other ion channel) in human IVD.

Fourteen human IVD samples; 4 non-degenerate (mean age 51 years) and 10 degenerate (mean age 43 years) discs were analysed for features of degeneration and separated into nucleus pulposus (NP) and annulus fibrosis (AF) tissue. Cells and RNA were extracted and RT-PCR performed for MaxiK channel (alpha subunit) and neural markers: GAP43, enolase2 and neuropeptideY. Articular chondrocytes and neuroblastoma cell line SH-SY5Y served as positive controls.

RT-PCR confirmed MaxiK channel expression in the degenerate AF and NP. However, no expression of the channel was observed in normal tissue, irrespective of disc region. There was no expression of neural cell markers in any IVD tissue.

We have shown for the first time that human IVDs express MaxiK channels. Expression is not dependent on disc region or patient age but is dependant upon degeneration. These findings have implications for understanding the phenotype of cells within the IVD and will help to elucidate the pathophysiology of IVD degeneration.

P88

Optimisation of Methods to Apply Mechanical Load to Mesenchymal Stem Cells

{P} SM Parker, AP Fotheringham, SJ Millward-Sadler, CE Evans, AJ Freemont, JA Hoyland
The University of Manchester, Manchester, United Kingdom

Mesenchymal stem cells (MSCs) are multipotent stem cells, derived from bone marrow that can differentiate into cells of connective tissue (e.g. chondrocytes, osteoblasts and adipocytes). Mechanical stimulation has been shown to affect differentiation of connective tissue cells, however to date the effect of mechanical load on MSC differentiation has not been elucidated.

MSC samples (♀, 57yrs, post-menopause; ♀, 38yrs, pre-menopause; ♀, 66yrs post-menopause and osteoporotic; ♂, 62yrs) were seeded in 24-well plates with standard, osteogenic or adipogenic media and subjected to a pressurised loading regime of 7Psi (48KPa) at a frequency of 1Hz for 1 hour ever other day for 14 days. Time points were taken at regular intervals between day 0 and day 28 and osteogenic and adipogenic differentiation was determined by alkaline phosphatase (ALP) assays, Oil red O staining and real-time PCR. Unloaded plates provided controls.

Osteogenesis and adipogenesis was slightly increased by load. However, following withdrawal of load a more prominent effect on MSC differentiation was observed.

Compressive load affects differentiation of MSCs down the osteogenic and adipogenic lineages. The nature of the response is dependant on patient age and menopausal status and the time duration post-load.

P89

Primary Aneurysmal Cyst Of Soft Tissue

{P} S Malhotra, S Patil, J Xuereb
Dept.of Histopathology,PathLinks, County Hospital, Lincoln, United Kingdom

Aneurysmal bone cyst (ABC) is a rare, locally aggressive bone lesion mainly affecting the metaphysis of long bones. ABC arising in soft tissues is even rarer. Only twelve cases have been reported in English literature. The diagnosis depends on radiological and histological features.
We report an unusual case of this entity in a 61-year-old woman who presented with a six months' history of a lump in left thigh. There was no history of trauma. On exploration, the lump was situated within the subcutaneous soft tissue with no deep extension beneath the deep fascia or to the underlying bone. Histology showed a well-circumscribed lesion with blood filled spaces lacking an endothelial lining surrounded by cellular stroma containing fibroblasts, mononuclear cells and osteoclast-like giant cells. There were occasional seams of lace like osteoid, woven bone and islands of calcification in the stroma. Extraskeletal telangiectatic osteosarcoma, myositis ossificans and ossifying fibromyxoid tumour are important differential diagnoses.
Histopathologists should be aware of the existence of soft tissue ABC that is histologically indistinguishable from intraosseous ABC and highlights the importance of clinical, radiological and pathological correlation. Soft tissue ABC infrequently recurs after marginal excision and hence, complete excision is the recommended mode of therapy.

P90

The Expression Of The Aggrecanolytic ADAMTSs And Their Endogenous Inhibitor TIMP-3 In Human Intervertebral Discs

{P} AJ Pockert[1], CL Le Maitre[1], DJ Buttle[2], AJ Freemont[1], JA Hoyland[1]
[1]. University of Manchester, Manchester, United Kingdom, [2]. University of Sheffield, Sheffield, United Kingdom

Loss of the proteoglycan aggrecan from the intervertebral disc (IVD) is an indicator of IVD degeneration (DIVD) which is implicated in low back pain (LBP). While members of the ADAMTS enzymes (-1, -4, -5, -9, -15) are aggrecanases with involvement in proteoglycan breakdown in cartilage, their expression in IVD is unknown. The aim of this study was to investigate expression of the aggrecanolytic ADAMTSs and their inhibitor TIMP-3 in normal and degenerate human IVDs.

Gene expression was assessed using relative and absolute quantification of real-time PCR for the ADAMTSs and TIMP-3 in 24 normal and 30 degenerate human IVDs. Immunohistochemistry localised ADAMTS-4 in 42 IVD samples of varying grades of DIVD.

Normal and degenerate IVDs expressed all target genes. The ADAMTSs were seen in more samples, and at higher levels, in degenerate discs than normal, with significant increases for ADAMTS-1 and -5. No difference was observed in TIMP-3 expression. ADAMTS-4 protein expression was significantly increased in degenerate discs than normal.

Aggrecanolytic ADAMTSs and TIMP-3 are expressed in human IVDs with increased gene and protein expression in degeneration, suggesting a dysregulation of the homeostatic mechanisms. This could be contributing to the pathogenesis of DIVD, therefore, a potential target for LBP management.

P91

Disc cell/Neural Cell Interactions in the Intervertebral Disc (IVD) and their role in Degeneration of the IVD

{P} D Purmessur, JA Hoyland, AJ Freemont
University of Manchester, Manchester, United Kingdom

Degeneration of the lumbar IVDs has been implicated in the pathogenesis of chronic low back pain. Little is known of the cellular and molecular events that underlie disease initiation and progression. However studies have demonstrated an association between disc degeneration and innervation of the usually aneural IVD.

The aim was to investigate disc cell/neural cell interactions in co-culture with and without cell contact

SH-SY5Y cells were differentiated and co-cultured with degenerate human nucleus pulposus (NP) cells with and without contact for up to 48hours. Real time PCR was carried out for gene expression of aggrecan, type II collagen and ADAMTS 4.

NP cells co-cultured without contact demonstrated a decrease in the relative gene expression of aggrecan and type II collagen (cell ratio disc: neural 75:25) compared to NP cells alone. ADAMTS-4 expression was detected in SH-SY5Y cells with a 5 fold increase with co-culture.

Neural cells may influence the extracellular matrix (ECM) profile of disc cells through release of mediators, thus favouring a phenotype characteristic of degeneration. ADAMTS-4 is an ECM degrading enzyme and was expressed by SH-SY5Y cells. Further investigations are required to identify mediators which may alter disc cell biology.

P92

Improved Detection of FUS-CHOP Fusion Transcript in Myxoid Liposarcoma Using Real-Time RT-PCR

N Etessami[1], L Lam[1], F Berisha[1], A M Flanagan[2], {P} T C Diss[3]
[1]. Royal National Orthopaedic Hospital, London, United Kingdom, [2]. Institute of Orthopaedics and Musculoskeletal Sciences, University College London, London, United Kingdom, [3]. University College London Hospitals, London, United Kingdom

Myxoid liposarcoma (MLS) may be difficult to distinguish from a wide range of small round cell tumours, including lymphomas, other myxoid lesions and well-differentiated liposarcomas. This is particularly so at unusual sites such as the foot where one of our cases was located. Detection of the characteristic t(12;16) which results in FUS-CHOP fusion transcripts can help the diagnosis. Detection of fusion transcripts using conventional RT-PCR with multiple primer sets is complex and may suffer from low sensitivity. We have developed a multiplex real-time RT-PCR approach using a dual-labelled probe to detect all common transcript variants in a single reaction. Transcript sub-types were determined by gel electrophoresis. Comparison of real-time RT-PCR with conventional RT-PCR was performed by analysis of RNA extracted from 29 cases of MLS. Fusion transcripts were detected in 18 cases (62%) using conventional RT-PCR, in 25 (86%) using real-time RT-PCR and in 26 using both (90%). The frequencies of the variants were similar to published data (Type I 19%, Type II 65%, Type III 15%). The three negative cases may carry rare transcript variants. Real-time RT-PCR offers a significant improvement to sensitivity of detection of the FUS-CHOP fusion transcripts in MLS.

P93

Differentiation of Mesenchymal Stem Cells to Nucleus Pulposus-like Cells in Chitosan/Glycerophosphate: A Potential Tissue Engineering Therapy for Degenerate Intervertebral Discs

{P} N Hughes[1], S M Richardson[1], J Hunt[2], A J Freemont[1], J A Hoyland[1]
[1]. University of Manchester, Manchester, United Kingdom, [2]. University of Liverpool, Liverpool, United Kingdom

A correlation between low back pain and intervertebral disc (IVD) degeneration has been reported. Degeneration results in loss of disc matrix, especially proteoglycan (PG) from its central, gelatinous, hydrophilic nucleus pulposus (NP). Current treatments, such as surgery, have limited success. Thus tissue engineering therapies are being developed to restore the IVD.
Aim: To replace the degenerate NP using a 'smart' hydrogel, chitosan/glycerophosphate (C/Gp) and autologous mesenchymal stem cells (MSCs), to produce a biomatrix with the same or improved properties of the non-diseased IVD.
Methods: Cells (MSCs, NP cells and articular chondrocytes) were cultured in C/Gp layers in vitro. At time points analysis of cell viability, mRNA expression and collagen/PG production was performed to determine cell phenotype.
Results: MSCs remain viable in C/Gp for up to 3 weeks and express NP marker genes after 2 weeks of culture similar to NP cells cultured in C/Gp. Also, matrix production occurred in C/Gp, with the greatest amount of PG produced by NP cells, followed by MSCs, with chondrocytes producing the least.
Conclusions: MSCs in C/Gp differentiate to NP-like cells and produce a matrix rich in PG, although not equivalent to that found in vivo. The addition of matrix enhancing growth factors is currently underway.

P94

Evaluation of Chitosan/Glycerophosphate-Hydroxyethylcellulose (C/Gp-HEC) for use in Tissue Engineering of the Intervertebral Disc

{P} N Kalson, S M Richardson, N Hughes, A J Freemont, J A Hoyland
University of Manchester, Manchester, United Kingdom

Introduction: Tissue engineering of the degenerate intervertebral disc (IVD) offers an exciting alternative to current symptomatic and surgical techniques. However the main challenges are to find a suitable scaffold to mimic the hydrophilic, gelatinous nature of the nucleus pulposus (NP) and to find a suitable cell source as recent evidence suggests degenerate NP cells are not ideal. Therefore we are investigating whether C/Gp-HEC 'smart' hydrogels, which have increased mechanical strength over standard C/Gp gels, seeded with mesenchymal stem cells (MSCs) are suitable candidates.
Methods: Bovine NP cells and human MSCs were cultured in C/Gp-HEC gels for up to 8 weeks. Cell viability, cell phenotype and matrix production were all analysed over the timecourse and expression of NP matrix components by MSCs was compared to that expressed by NP cells.
Results: High cell viability was observed throughout the timecourse and both cell types were found to express high levels of both collagens and proteoglycans. MSCs also expressed genes for other NP matrix proteins.
Conclusions: MSCs in C/Gp-HEC gels differentiate into NP-like cells and produce a matrix similar to that of the NP. Further optimisation of culture conditions may result in a system that is usable for tissue engineering of the IVD.

P95

Frequent Detection of β-catenin Gene Mutations in Paraffin-Embedded Sporadic Desmoid-Type Fibromatosis by Mutation-Specific Restriction Enzyme Digestion

{P} L Islam[1], MF Amary[2], B Idowu[2], T Diss[3], AM Flanagan[2]
[1]. Royal National Orthopaedic Hospital, Middlesex, United Kingdom, [2]. Institute of Orthopaedics and Musculoskeletal Sciences, UCL, London, United Kingdom, [3]. University College Hospital, London, United Kingdom

Fibromatosis (desmoid-type) is a locally aggressive deep soft tissue tumour with a high recurrence rate. Some cases are associated with familial adenomatous polyposis (FAP) and inherited mutations of the APC gene. Point mutations of the β-catenin gene have been identified in sporadic fibromatosis. These produce amino acid substitutions, stabilising β-catenin which binds transcription factors and switches on the Wnt signalling pathway. To assess the frequency of the most common β-catenin gene mutations in a large series, 73 cases of fibromatosis were studied. Exon 3 of the gene was amplified and PCR products digested by restriction enzymes to identify the three most common point mutations involving codons 41 and 45. A restriction site was present for analyses of mutations in codon 41. Mismatch primers were designed for two mutations in codon 45. Mutations were present in 56 cases (77%). Of these, 29 (52%) were in codon 45 (TCT>TTT), 22 (39%) in codon 41 (ACC>GCC) and 5 (9%) in codon 45 (TCT>CCT). β-catenin gene mutations in fibromatosis play an important role in tumorigenesis and can be targeted for restriction enzyme digestion. Diagnosing fibromatosis can be difficult and exploiting these mutations in this manner can complement light microscopy in reaching a diagnosis.

P96

SYT-SSX Fusion Transcript can be Detected in a Very High Proportion of Formalin-Fixed Paraffin-Embedded Synovial Sarcomas using Conventional, Real-Time RT-PCR and FISH

{P} MFC Amary[4], L Lam[2], FDC Bernardi[4], M Michelagnoli[3], F Berisha[2], T Diss[3], AM Flanagan[1]
[1]. Institute of Orthopaedics & Musculoskeletal Science - UCL, Lonon, United Kingdom, [2]. Royal National Orthopaedic Hospital, London, United Kingdom, [3]. University College London Hospitals, London, United Kingdom, [4]. Santa Casa School of Medical Sciences, Sao Paulo, Brazil

Synovial Sarcoma (SS) is a high-grade soft tissue tumour with several histological subtypes and poses potentially problematic differential diagnoses. The disease is characterised by t(X;18) resulting in SYT-SSX fusion. 119 cases (104 paraffin) classified as 'almost certain', 'possible' or 'probable' SS, were investigated for SYT-SSX fusion transcripts by real-time and conventional RT-PCR. Age varied from 5-81. A fusion was detected in 114 (96%) cases (61 SYT-SSX1, 43 SYT-SSX2, both in 5 cases, 5 SYT-SSX undetermined). 107 (94%) were positive by both methods: five were positive only by real-time PCR, all with poor quality RNA. Two SYT-SSX2 cases were positive only by conventional RT-PCR. None was positive for SYT-SSX4 or SS18L1-SSX. No fusion was detected in 5 cases (4%) using both RT-PCR methods and FISH (SYT break-apart probe). Of the negative cases, 4 were at unusual sites and classified as 'possible'. Only one, a recurrence in the lower extremity, was classified as 'almost certain': these may represent different diseases or carry variant mutations. Conventional and real-time RT-PCR detected SYT-SSX in a high proportion of cases. Combining methodologies offers a powerful aid to diagnosing these tumours in paraffin-embedded samples. Real-time RT-PCR has the advantages of speed, reduced contamination risk and improved sensitivity.

P97

Human Meniscus Cells Contain A Progenitor Cell Population

AB Adesida, {P} SJ Millward-Sadler, TE Hardingham
University of Manchester, Manchester, United Kingdom

De-differentiated human articular chondrocytes have been shown to have the capacity differentiate towards the adipogenic and osteogenic lineages, and also the potential to re-differentiate in response to chondrogenic stimuli. Furthermore, a subpopulation of chondrocytes with progenitor-like characteristics have been isolated from the surface zone of bovine articular cartilage using differential adhesion to fibronectin. The aim of this study was to determine whether human meniscus cells could be differentiated down mesenchymal cell lineages in culture.
Passaged meniscus cells were cultured in chondrogenic, adipogenic, osteogenic or control medium. Expression of PPARγ, lipoprotein lipase (LPL), osteocalcin, alkaline phosphatase, collagens type I and II and aggrecan were determined by RealTime PCR and immunohistochemistry, and cultures were stained with oil red O or alizarin red.
Meniscal cells cultured in chondrogenic medium showed no alteration in expression of collagens type I or II or aggrecan when compared with controls. Meniscal cells cultured in adipogenic medium showed an increase in PPARγ and LPL expression and an increase in oil red O staining. Only one out of four samples cultured in osteogenic medium showed any increase in osteogenic markers.
These results indicate that meniscus contain a population of progenitor cells.

P98

Low Oxygen Concentrations Modulate Matrix Production In Human Meniscus Cells

AB Adesida[1], LM Grady[1], WS Khan[1], {P} SJ Millward-Sadler[1], DM Salter[2], TE Hardingham[1]
[1]. University of Manchester, Manchester, United Kingdom, [2]. Edinburgh University, Edinburgh, United Kingdom

The biomechanical properties of the meniscus are dependent on the composition and organisation of the tissue and its extracellular matrix. The outer zone of the meniscus is composed of fibroblast-like cells surrounded by a type I collagen-rich matrix. The inner zone is avascular, and composed of cells of a more chondrocytic phenotype surrounded by matrix composed of a mixture of type II and type I collagen, and aggrecan. The aim of this study was to investigate the effect of oxygen concentration on matrix production by meniscus cells.
Human meniscus cells were cultured in monolayer or pellet form under 20% or 5% oxygen and analysed by real-time PCR and immunohistochemistry.
Pellets showed an increase in SOX9 and COL2A1 mRNA relative to monolayers. SOX9 was increased further by 5% O_2, which also induced hypoxia inducible factor-1α (HIF-1α), prolyl-4 hydroxylase α(I) (P4Hα(I)) and HIF prolyl-hydroxylase 2 (PHD2) expression. Type II collagen was increased in 5% O_2 compared with 20% O_2, but type I collagen was unchanged. COL2A1, HIF-1α, P4Hα(I) and PHD2 mRNA expression were higher in the inner region compared with the outer zone of tissue.
These results suggest that oxygen concentration plays an important role in modulating meniscus matrix production.

P99

Cherubism is a Clinical Phenotype Defined by Genetic Mutations in The RAS/ERK/MAPKinase Pathway

{P} B.D. Idowu[1], J.M. Mangion[2], P.H.G. Hogeman[3], R.A. Gale[4], A.M. Flanagan[1]
[1]. Institute of Orthopaedics & Musculoskeletal Science, London, United Kingdom, [2]. MRC Clinical Sciences Centre, London, United Kingdom, [3]. Meander Medisch Centrum, Amersfoort, Netherlands, [4]. Department of Haematology, London, United Kingdom

Cherubism is a clinical phenotype characterised by bilateral enlargement of the jaw. The histopathology shows an osteoclast-rich lesion that is difficult to distinguish from giant cell neoplams. The disease is inherited as an autosomal dominant trait with variable expressivity and approximately 80% penetrance. It is associated with mutations in SH3BP2 and PTPN11. We report on 50 families with Cherubism: 24 with a family history; of the remaining 12, five had de novo mutations, 7 could not be determined. Nine nonsynonymous SH3BP2 point mutations were identified in 72% (36) of the families, 3 being novel mutations. These involved 5 amino acids spanning 7 codons in exon 9. Four (8%) families had mutations (2 novel) in PTPN11. For the first time, a NF1 mutation was found in a patient with Cherubism: this was a de novo mutation involving a splice donor site (IVS37+1) and associated with mild clinical features of NF.
NF1, PTPN11 and SH3BP2 genes are all involved in the RAS/ERK/MAPKinase pathway. The remaining 9 (18%) cases did not have a mutation in KRAS and these cases are currently been screened for mutations in HRAS and BRAF.
This study implicates activation of the RAS/ERK/MAPKinase pathway in the pathogenesis of osteoclast-rich neoplasms.

P100

The Significance of the p130/p107 Switch in Adipocytic Neoplasms

{P} E Sarkady[1], J Milward Sadler[2], A.J. Freemont[2]
[1]. Tameside General Hospital, Manchester, United Kingdom, [2]. University of Manchester, Manchester, United Kingdom

Aim: Is to identify the presence and role of some members the retinoblastoma tumour suppressor gene family and other key elements in adipocytic tumors.
Background: stages of adipocytic differentiation are coordinately linked to cell cycle events. One of the many proteins are the E2F family of transcription factors. Their action is mediated through members of the retinoblastoma suppressor gene family, such as Rb, pRb, p107, p130. The p130/p107 switch is necessary for the physiological clonal expansion phase of adipocytes. TNF-α has been recognized to impair this and it also downregulates certain genes involved in normal adipocyte differentiation, such as PPARγ.
Method: Immunohistochemistry for TNF-α, its receptor, E2F, pRb, p130, p107 and PPARγ2 on a selection of adipocytic tumors using tissue microarray.
Results: p130 was expressed in all lipocytes of normal adipose tissue, lipoma, hybernoma or atypical lipoma, in some tumour cells of myxoid liposarcomas, but in none of round cell and pleomorphic liposarcomas. P107 and E2F was infrequently seen in the nuclei of benign lesions, but was strongly expressed in all liposarcomas.
Conclusion: Our preliminary results suggest that p130, p107 switching and E2F may play a role in adipocytic tumourgenesis.

P101

Preservation of RNA Integrity and Morphology in Surgical Tissue

{P} S.E. Bray, F.E.M. Paulin, D.A. Levison, R.J.C. Steele
University of Dundee, Dundee, United Kingdom

A tissue bank acts as a repository for tumour tissue, gifted from patients, for use in approved research projects. From these small fragments of tissue, DNA, RNA and protein can be extracted for molecular studies. RNA is particularly labile and its extraction from these specimens has yielded inconsistent and disappointing results. We have therefore investigated a procurement procedure using RNAlater™ to determine if this preserves RNA integrity without compromising other molecular investigations or tissue morphology for diagnostic purposes.

Ten patients with large colorectal tumours had tumour biopsies removed intra-operatively via colonoscopy. One was placed immediately in RNAlater™ and others were snap frozen at fixed timepoints thereafter. RNA from tissue placed immediately in RNAlater™ was found to have a substantially higher RNA integrity number (RIN) following analysis by Agilent 2100 Bioanalyser than all other samples.

Histological examination revealed that samples incubated in RNAlater™ for 24 hours prior to formalin fixation displayed poor morphology and immunoreactivity in comparison to samples placed immediately in formalin.

Whilst RNAlater™ is a suitable collection and storage medium for tissue intended for subsequent functional genomic studies, it renders surgical tissue unsuitable for pathological diagnosis. Therefore to obtain most value from research specimens a multi-procurement protocol is necessary.

P102

Spectral imaging for automated detection of acute myeloid leukaemia in bone marrow trephine samples

{P} S MacDermott, RJ Byers
University of Manchester, Manchester, United Kingdom

Automated morphological classification would facilitate high throughput histological analysis and has in the past been attempted with little success. Recent advances in spectral imaging have enabled greater precision of spectral definition that may improve morphological classification. As a proof of principle we used spectral imaging to define the spectral characteristics of bone marrow trephines containing acute myeloid leukaemia (AML) and reactive bone marrow and applied these to cases to test ability to classify on spectral data alone.

Spectral cubes were collected using a CRI Nuance system, which collates images at multiple wavelength steps, at 10nm intervals, over the visible range (500-700nm), from which spectral profiles can be calculated for each pixel in the image field. The spectral profiles of AML and reactive bone marrow were calculated in 2 training cases and used to classify 40 test cases (13 reactive & 27 AML). Ten of the 13 reactive cases and 18 of the 27 AML cases were correctly classified, with a sensitivity and specificity of 76% and 73% respectively.

The results demonstrate the ability of the technique to provide limited classification but further work is required to improve accuracy. Addition of spatial data to achieve this is being investigated.

P103

A Novel Loading Rig To Mimic The *In Vivo* Environment Of The Human Intervertebral Disc

{P} CL Le Maitre, JF Frain, AP Fotheringham, AJ Freemont, JA Hoyland
University of Manchester, Manchester, United Kingdom

Mechanical loading is a contributory factor in degenerative disc disease. The "load environment" on intervertebral discs in humans is different from that of experimental animals, as such their use in studies on loading of IVD cells is inappropriate. Thus, we have designed a compressive loading apparatus to mimic the *in vivo* environment experienced in the human IVD.

The system consists a computer controlled hydraulic piston connected to a pressure vessel filled with water maintained at 37°C containing samples suspended in sterile bags allowing the application of hydraulic load. Loading regimes are constantly monitored with an internal pressure transducer. Several loading regimes have been established to represent a range of normal and abnormal activities encompassing static and dynamic loading regimes. Using human IVD cells and tissue samples we have shown this test system generates, in a fully controllable manner, a loading environment similar to that seen *in vivo* without any loss of cell viability.

In conclusion this test system allows the maintenance of human IVDs in an environment that reflects the load experienced *in vivo*. Such a system permits studies of the effect of load in human IVD samples and the pre-clinical testing of tissue engineered IVD constructs.

P104

Tissue Preservation. Is Galvanisation The Answer?

{P} Dimitrios Lykidis, Susan Van Noorden, Mahrokh Nohadani, Bradley Spencer-Dene, Gordon Stamp
Imperial College London, London, United Kingdom

The aim of this study was to identify a reliable, cost effective, non-toxic 'ideal fixative' to meet the needs of contemporary molecular research and diagnostics. The effects of 'routine' and 25 novel or modified fixatives were investigated on tissues from male/female C57Bl6 mice. Colon, liver and spleen were fixed and processed to paraffin.

A modified zinc-based fixative (zinc chloride, zinc trilfuoroacetate, calcium acetate in Tris buffer) significantly improved protein, DNA and RNA preservation compared to the standard zinc fixative and to neutral buffered formalin as shown by immunohistochemistry (cytokeratin, muscle-specific actin, CD3), PCR (0.6Kb, 1Kb), RT-PCR (0.3Kb), RNA Agilent Bioanalyser, Real-Time PCR (GAPDH, beta-actin) and Real-Time RT-PCR (S15). The advantage was sustained in stored blocks. Fresh frozen samples served as controls for DNA and RNA integrity.

In addition, studies on tissue morphology, using 14 different zinc components at various concentrations, showed that the above modified zinc based fixative was overall the best candidate. Since the function of zinc as a fixative is unknown, future work will focus on the molecular basis of zinc fixation via mass spectrometry.

The findings of this work provide an improved tissue fixation method that could be used routinely in diagnostic labs and for molecular studies.

P105

The Tissue-Tek® Xpress™ Processor in routine practice – Technical Validation and Impact on Patient Pathways

{P} N R Griffin, R I Banks
Leeds Teaching Hospitals NHS Trust, Leeds, United Kingdom

The Tissue-Tek® Xpress™ processor represents a paradigm shift in tissue processing technology with a 1 hour processing time for paraffin embedded tissues and potential automated embedding, enabling routine high volume, rapid throughput processing with genuine potential for routine same day reporting.

In order to validate this technology for use in routine diagnosis we have carried out an extensive assessment the quality of H&Es, common special stains, immunohistochemistry and molecular pathology in tissues processed through the Xpress™, in parallel with material processed by a conventional Tissue-Tek® VIP processor.

Sections were assessed using NEQAS methodology by a team of 2 biomedical scientists and 2 histopathologists. Results showed that H&E, special stains and immunocytochemistry from tissues processed by the Xpress™ provided sections of high quality which scored well using the above criteria. We have also had some success with In Situ hybridisation and nucleic acid extraction, and development work is ongoing in this area.

These results indicate that the Xpress™ produces acceptable results and can be used with confidence for routine diagnosis. Modelling of laboratory workflows and skill-mix exploiting this technology to the full shows significant benefits to the service. The potential for routine same-day reporting will enable fundamental re-engineering of patient pathways.

P106

A Dual Beam Fibre Trap for Raman Micro-spectroscopy of Single Cells

P Jess[1], V Garces-Chavez[1], D Smith[2], M Mazilu[1], A Riches[2], {P} CS Herrington[2], W Sibbett[1], K Dholakia[1]
[1]. School of Physics and Astronomy, University of St Andrews, St Andrews, Fife, United Kingdom, [2]. Bute Medical School, University of St Andrews, St Andrews, Fife, United Kingdom

Raman spectroscopy permits acquisition of molecular signatures from both cellular and sub-cellular samples. When combined with optical trapping, an isolated cell can be interrogated, reducing extraneous signals from the local environment. To date, schemes have employed combinations of a single beam optical tweezers trap and Raman spectroscopy using either the same beam or separate beams for Raman interrogation and trapping. A key problem in optical tweezers is the ability to hold and manoeuvre large cells. We demonstrate the use of a dual beam fibre trap to hold and manoeuvre cells whilst using an orthogonally placed objective to record Raman spectra. Due to its divergent light fields, the dual beam trap offers an as yet unexploited ability to hold and move large cellular objects. We additionally show how this system permits movement of large primary human keratinocytes (approximately 30 microns in diameter), such that Raman spectra can be recorded from local parts of a trapped cell with ease. Finally, we have developed a rudimentary microfluidic system using a microcapillary such that we can flow cells, hold them using our trap, and acquire Raman spectroscopy from chosen cell samples. This approach holds promise for the optical analysis of clinical samples.

P107

Challenges of Computer-Based Image Analysis in Histopathology

{P} C.C. Chibelushi[1], B. Sharp[1], C. Platt[2]
[1]. Staffordshire University, Stafford, United Kingdom, [2]. Guy's and St Thomas' NHS Foundation Trust, London, United Kingdom

Images are increasingly used for diagnosis, prognosis, therapy, healthcare planning and health monitoring. Histopathology primarily handles tissue images obtained from gross specimens and light microscopy. The acquisition, storage, transmission, and processing of digital images have seen large decreases in price-performance ratio. The power of computers could be harnessed in histopathology, to enhance the quality and speed of healthcare delivery.
Research into automated image analysis in histopathology has made significant advances; partial automation is becoming feasible. However, despite the potential benefits of digital image analysis, the use of assistive tools in histopathology is still hampered by costs, technological limitations and non-technical issues such as data protection.
Although digital image analysis techniques give promising performance in controlled settings, they generally have insufficient accuracy or reliability for the often stringent requirements of clinical practice. In addition many histopathologists have limited familiarity with digital image analysis and there is little training in this aspect of pathology.
This paper discusses developments in digital image analysis for histopathology. It focuses on outstanding problems which hinder automated image analysis.

P108

A new approach for validating antibodies in diagnostic histopathology; dispel the myth

{P} S Deen, R Allibone, P James, K Kulkarni, I Leach, D O'Neil, J Padfield, K Robson, J Lowe
Queen's Medical centre, Nottingham, United Kingdom

Aim: To use composite tissue microarrayed blocks to assess the performance of antibodies as a diagnostic tool.
Method: Microarrayed blocks of triplicate 0.6-mm cores from 84 cases representing benign and malignant cases of variable origin including gastrointestinal, neuropathology, skin and gynaepathology, identified from case archives were constructed and stained with commonly used antibodies according to our departmental protocols.
Results: Staining results as a range of samples were compatible with those described in the literature for most antigens. We identify two diagnostically important issues:
1) Mesothelin staining showed variable expression different to that of the published data by being negative for ovarian serous tumours while the antibody was validated by a positive mesothelioma tissue control. Pancreatic adenocarcinoma was positive. 2)P53 staining was seen in normal tissue as well as the expected and unexpected tumours.
Conclusion: Multiple tissue microarrays offer an efficient method to assess the clinical performance of antibodies and raise the issue of adjusting primary antibody concentration to detect an appropriate threshold of positivity. As a tool for quality assurance, this technique takes into account local differences in tissue fixation, processing and staining. Our preliminary results confirm the limitations of currently available antibodies in giving unequivocal tissue-specific staining patterns and reveal the need for a formal performance assessment for antibody staining in the local laboratory using Receiver Operating Characteristic (ROC) curve to provide a pure index of accuracy.

P109

Value of perinatal autopsy of 74 dead fetus and neonates (Isfahan , Iran 2001- 2005).

{P} Parvin Rajabi dehnavi, Mehrangiz Ghafari, Zeinab Aram
Isfahan university, Isfahan, Iran

Objective: Perinatal death complicates 1.5% of all births. A perinatal autopsy can provide an explanation for a loss, often relieving the parents and their physician, and may reveal a specific disorder for which precise recurrence risk or strategies for prevention are available. Our aim was determining causes of death based on pathologic findings.
Methods: This descriptive cross- sectional study was done on 74 perinatal autopsy during 2001-2005 in Alzahra hospital in Isfahan -Iran.
Results: Seventy four dead fetus and neonates which their parents allowed us for autopsy were studied during 2001-2005. Causes of death were determined in 82.4 percent (61cases). Anatomical abnormality were seen it 64.8 percent of cases. There were no pathologic finding in 8 cases (10.8%) and unexpected pathology (different from clinical diagnoses) were seen in 9 cases (12.1%). Maceration was seen in 5 cases(6.75%).
Conclusion: The cause of fetal or Perinatal death was determined by autopsy in 82.5 percent of cases in our study. Asphyxia, IUGR (Intra uterine growth retardation) and renal agenesis were the most common cause of death . These findings show the importance of screening for these disorders.
Key words: Autopsy, Dead fetus , Perinatal , Asphyxia , renal agenesis, Intra uterine growth retardation

P110

Audit Of Forensic Autopsies In Leicester: Investigations Conducted And Report Turnaround Time

{P} L R Dunk[1], G N Rutty[2]
[1]. Leicester Royal Infirmary, Leicester, Leicestershire, United Kingdom, [2]. University of Leicester, Leicester, Leicestershire, United Kingdom

This audit investigates the time it takes to produce forensic autopsy reports in Leicester, the investigations that are requested and the possible reasons for any delay in production of the autopsy report. In general, the process for the production of an autopsy report is as follows: autopsy is conducted and investigations are requested, investigation results are received, a draft report is reviewed by a forensic pathologist not involved with the case, a final report is produced.

Autopsies conducted by forensic pathologists in Leicester for the year 1st October 2004 until 30th September 2005 were included (149 cases). The median time for production of a report was 66.5 days (range 5-167 days). In only 12 (8%) of cases the report was produced within the contracted time of 21 days. Common investigations requested were: histology (87% of cases), toxicology (86%), neuropathology (15%) and microbiology (10%). Receiving the results of investigations constituted the longest step in report production, taking a median of 37 days (range 2-266 days).

Conclusion: The time taken to receive the results of investigations is the longest step involved in production of a forensic autopsy report, often alone taking longer than the 21 days allowed.

P111

Improving The Standard Of Autopsy Reporting In A Busy Hospital Autopsy Unit

{P} N.P. West, S.M. Claydon
Department of Histopathology, Leeds General Infirmary, Leeds, United Kingdom

The Royal College of Pathologists published their latest minimum dataset for autopsy reporting in 2002. An audit was performed to determine the impact of the Royal College guidelines on the standard of autopsy reporting in a busy hospital autopsy unit. The overall aim was to identify how to further improve the current standard of reporting.

100 autopsy reports from both 1995 and 2005 were compared to the minimum dataset. The mean percentage of essential details omitted from autopsy reports had decreased from 23.5% to 19.1% following the publication of the Royal College guidelines. This represented a significant improvement ($p<0.001$).

It was noted that many of the essential details omitted from autopsy reports were historically badly recorded. We hypothesised that the introduction of a standardised electronic proforma containing the full minimum dataset would further improve the standard of autopsy reporting.

Following the introduction of the proforma, the mean percentage of essential details omitted from autopsy reports fell to 11.2%, representing a further significant improvement ($p<0.001$). We conclude that the use of electronic proformas can significantly improve the standard of autopsy reporting in a busy hospital autopsy unit.

P112

Can Cause Of Death Be Predicted From The History Alone?

{P} MJP Biggs, LJR Brown, GN Rutty
Leicester Royal Infirmary, Leicester, United Kingdom

Aim: To determine whether the cause of death can be accurately predicted from the available history, before carrying out a necropsy.
Methods: In this prospective study pathologists gave a predicted cause of death after reading the history. They then noted any relevant information from the external examination prior to carrying out the necropsy.
Results: A total of 101 necropsies were included in the study. The cause of death was deemed to have been correctly predicted from the history in 66 cases (65.3%). In 35 cases (34.7%), an unexpected cause of death was found. Findings from the external examination were noteworthy in only 8 cases (7.9%) and did not alter the predicted cause of death in any case.
Conclusions: In certain circumstances an accurate cause of death could be given with confidence without a necropsy. However, many common causes of death can present with similar or misleading scenarios. This study supports the current system of performing necropsies, despite seemingly predictable circumstances, if an accurate cause of death is to be recorded.

P113

Fatal Overdose Due to Prescription Fentanyl Patches in a Patient With Sickle Cell/β-thalassaemia and Acute Chest Syndrome. A Case-Report and Review of the Literature

{P} OJ Biedrzycki[1], DH Bevan[2], SB Lucas[3]
[1]. Department of Histopathology, The Royal London Hospital, London E1, United Kingdom, [2]. Department of Haematology, St Georges' Hospital, London SW17, United Kingdom, [3]. Department of Histopathology, King's College London School of Medicine, St Thomas' Hospital, London SE1, United Kingdom

The analgesic fentanyl is a 100 times more potent than morphine. Methods of administration include the Duragesic® patch, used widely in management of chronic pain. The first fentanyl-related deaths were reported in 1979, and since then ingenious methods of abuse have evolved.

We describe an accidental overdose and death of a 32 year old male with sickle cell/β-thalassaemia who was a long term user of the Duragesic® patches.

At post mortem an enlarged heart (440g) and spleen (700g) were noted and the lungs were congested and oedematous. Histopathologically, there was marked contraction band necrosis of the right ventricle and acute sickling of red blood cells in the lungs. The cause of death was initially thought to be acute chest syndrome due to sickle cell crisis (SCC). Following re-evaluation of the clinical history, toxicology analysis revealed grossly elevated blood and urine fentanyl levels.

Fentanyl is not detected in all toxicology laboratory screening processes so the total number of deaths that involve accidental or deliberate overdose may be underestimated. We discuss the interplay between the fentanyl overdose and SCC. Given the widespread use/abuse of fentanyl, we remind pathologists to consider it when confronted with non-specific features of narcotic overdose at post-mortem.

P114

An Evaluation Of The Causes Of Cardiac Tamponade, At Postmortem, In Rural District General Hospital

A Swaminathan[1], {P} M Powari[1], K Kandasamy[2], J Mathew[1]
[1]. Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [2]. Department of Cardiology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom

Aim: To define the causes of cardiac tamponade (CT) in routine post-mortem practice in a rural population.
Materials and Methods: The postmortem register and database were used to identify all patients who died as a result of CT between 1995 and 2004 (inclusive).
Results: 14,368 postmortems were performed in this period: 461 patients (3.2%) were identified as having died from CT. Three of these patients died as a result of non-haemorrhagic pericardial effusion defined as passive (n=2) or infective (n=1).
Haemopericardium (HP) (n=458) was the commonest cause of CT. In the group of patients dying from HP, the commonest causes were rupture of an acute myocardial infarction (RAMI) (n=311: 67.9%) and intrapericardial rupture of dissecting aortic aneurysm (RDAA) (n=138: 30.1%).
Post-traumatic HP (n=5: 1.1%) and miscellaneous causes (n=4: 0.9%), including pericardial angiosarcoma, metastatic lung cancer, warfarin therapy and malignant haemorrhagic pericardial effusion - unknown primary, were less frequent causes.
In the non-traumatic category and, excluding miscellaneous causes, the male (n=182) to female (n=267) ratio was 1:1.46. Intrapericardial blood volume was recorded in 246 cases (54%) ranged between 150ml and 1650ml. The volume of intrapericardial haemorrhage was broadly similar in men (mean = 507ml) and women (mean = 438ml) and also similar whether as a consequence of either RAMI (mean = 440ml) or RDAA (mean = 498ml) in both sexes
Conclusion: At postmortem, CT is most often related to HP, attributable to either RAMI or intrapericardial RDAA. Post -traumatic and other causes of CT are infrequent.

P115

The Role of Autopsy Teaching in Medical Education

{P} L Rogers-Holmes, P Quirke, P.A. Burns
Pathoology, Leeds Institute of Molecular Medicine, Leeds University, Leeds, Yorkshire, United Kingdom

This study investigated the role of the autopsy within medical education at the University of Leeds by surveying the attitudes of students on the MBChB course`. All 774 students in the first three years of the course were invited to complete a questionnaire designed to survey their opinions on the importance of seeing an autopsy, which is voluntary at Leeds, as part of their medical education. They were asked what they felt they would learn, or had learnt, from seeing one. The 31 medical schools of the United Kingdom were also surveyed about their attitudes to teaching autopsies. Most UK universities encourage, but do not compel, their undergraduates to attend an autopsy
In Leeds, compliance ranged from 42% of third year students up to 90% of first years. The survey indicated an overwhelming (99%) desire among first-year students to observe an autopsy. Second- and third- year students that had seen autopsies felt that they had gained a better understanding of anatomy and the effect of disease on organs, although the number who actually attended was much lower than data from first-years interest would indicate (32% of second years and 26% of third years). When asked over two thirds of students claimed they would not feel competent to consent relatives for an autopsy if they had not seen one.

P116

An audit of B3 breast needle core biopsy reports and their correlation with excision / mammotome biopsy histology reports

{P} U CHANDRAN, S WELLS
Devi Ahilya University, Indore, India

Histopathological evaluation of breast needle core biopsies of palpable as well as screen detected nonpalpable breast lesions are classified according to the NHSBSP guidelines from category B1 to B5. B3 category lesions are known to be associated with in-situ or invasive carcinoma. Hence, all such lesions are followed up by either wide local excision or mammotome biopsy to rule out malignancy.
The aim of this audit was to evaluate the prevalence of malignancy in subsequent excision histology of B3 category lesions. A total of 87 cases of B3 category were analysed.
The overall prevalence of malignancy was 24% [20/87]. Lesions with a diagnosis of atypical ductal hyperplasia (ADH) alone or in association with other B3 lesions had a high incidence of in-situ or invasive malignancy i.e. 58% [17/29]. Whereas the prevalence of malignancy in other lesions like intraductal papilloma and radial scar without associated ADH was low i.e. 5% [3/58].
Hence, we propose that B3 category lesions could be further subdivided into two groups : (a) atypical ductal hyperplasia with an increased risk of malignancy and (b) remaining lesions with a low risk of malignancy.
It is important to develop a local policy to follow up these cases depending upon the clinical and radiological assessment. A further audit can be done to correlate the clinical and radiological picture with different groups of B3 category lesions.

P117

Expression of the Tenascin-binding α9β1 Integrin in Normal Breast, DCIS and Invasive Breast Cancer

{P} HL Nitch-Smith[1], R Walker[2], IR Hart[1], R Carpenter[3], JL Jones[1]
[1]. Institute of Cancer, Queen Mary's School of Medicine and Dentistry, London, United Kingdom, [2]. Breast Cancer Research Unit, University of Leicester, Leicester, United Kingdom, [3]. Breast Unit, St Bartholomew's Hospital, London, United Kingdom

Cellular interactions with the extracellular matrix are essential for the transduction and integration of signals from the microenvironment. The predominant receptors involved in mediating these signals are the integrins. The signal produced is precisely dependant on the specific integrin-ligand interaction, and with changes in both the matrix composition and tumour cell integrin expression, there exists the potential to generate novel integrin-ligand interactions in the tumour setting. Tenascin-C is frequently upregulated in breast cancer and the recently characterised α9β1 integrin uses this as a ligand, however it's expression in breast is unknown.
This study aimed to establish the expression and localisation of α9β1 integrin in normal, pre-invasive and invasive breast lesions using standard immunohistochemical techniques on frozen breast tissue.
Normal breast exhibits strong staining at the basal aspect of myoepithelial cells and this was maintained in all DCIS lesions. 16% of invasive ductal carcinomas were also positive for α9β1 (n=50), and these were predominantly grade III lesions. Interestingly, α9β1 was also detected on lymphatic endothelium.
In conclusion, we have identified α9β1 as a myoepithelial-associated integrin and shown that a subset of grade III infiltrating ductal carcinomas also express this integrin. The effect of α9β1on breast tumour cell behaviour is currently being investigated.

P118

Adequate processing of breast core biopsies for mammographically detected microcalcification

{P} E A Husain, S Agrawal, J Tan, D Al-Okati
BHR NHS Trust, Essex, United Kingdom

Introduction: Mammographically detected microcalcification is a frequent finding accounting for almost half of all biopsies taken from non-palpable breast lesions. This represents a major part of the workload of any histopathology laboratory involved in the breast screening programme.
Objectives: To assess the number of histological levels required to optimize the workload of the laboratory staff and consultants and ensure cost-effectiveness. Assessment of the optimum number of cores required for adequate reporting.
Materials and methods: Seventy biopsies of mammographically detected microcalcification taken from 64 patients were included in this study. The cores were counted and radiographed before processing. The H & E stained sections were examined for microcalcification and histological diagnosis was established. The levels at which calcification appeared on histological section were recorded. For cases with no histological evidence of microcalcification on initial 4 levels, further levels were requested to reveal calcification till the core was exhausted. All the negative cases were polarised to rule out calcium oxalate deposits.
Results: Only 41 (58%) cases showed histological microcalcification on examining 4 levels and this increased to 54 (77%) on level 6. Further levels did not result in significant increase in the rate of microcalcification detection. Increasing the number of cores received upto 5, correlated with higher rate of microcalcification detection.
Conclusions: To ensure a high rate of detection of microcalcification in breast core biopsies, a minimum of 5 cores and six histological levels are required.

P119

North West Regional Breast Screening Pathology B3/4 Audit

{P} S A Hales[1], F Knox[2]
[1]. Countess of Chester, Chester, Cheshire, United Kingdom, [2]. South Manchester Hospitals, Manchester, Greater Manchester, United Kingdom

This region had one of the highest benign biopsy rates in the country in recent years. This audit was undertaken to see if this was due to a high number of inappropriate B3 and B4 core biopsy diagnoses by the regional pathologists. The wide bore needle biopsy QA statistics standard report was obtained for each unit via the screening offices for the years 2002/3 and 2003/4. B3/4 cases with a benign outcome were identified and the subsequent excision specimen reports analysed. The type of lesions identified mirrored that of published data. Over the two years there was an increase in B3/4 diagnoses, but a decrease in the benign biopsy rate. In some units, the use of the B3 and B4 categories was very low, but it was unclear if this was due to misreporting or miscoding of diagnoses such as radial scar or to a genuinely low incidence of these. The numbers of B3/4 diagnoses with a benign outcome did not correlate with the benign biopsy rate in all units.
The high benign biopsy rate does not appear to be due to a high number of inappropriate B3/4 diagnoses by NW breast screening pathologists.

P120

A diffusion-limited-aggregation (DLA) model of human breast lobe growth

{P} J.J. Going[1], P. Bourke[2]
[1]. University of Glasgow, Glasgow, United Kingdom, [2]. Swinburne University, Melbourne, Australia

Background. The 'sick lobe' hypothesis emphasizes a lobar dimension in breast intraepithelial neoplasia. Separate parenchymal lobes differ greatly in size, but it is not known whether this variation is random, or deterministic. 'Virtual lobes' were grown '*in silico*' to explore size variation in a model of breast lobe growth. *Methods*. The virtual lobes were grown inside an 'envelope' bounded by a 3D breast surface scan and a plane representing the chest wall. 3D diffusion-limited aggregation (DLA) used seed points representing lobar anlagen of the breast bud with visualisation in POV-Ray. *Results*. Branching lobe-like structures were created, with striking similarities and differences from real breast structure. Highly variable lobe size mirrored the *in vivo* situation, but long unbranched central ducts, which occur in human breast, did not appear. *Conclusions*. Highly variable lobe size emerges readily in a random model of breast lobe growth. It is certainly possible, therefore, that variable lobe volume in human breasts could arise by stochastic processes. However, differences between virtual and real lobe branching emphasize that DLA modelling over-simplifies actual human breast growth, and lobe-size determining events such (hypothetical) growth promoting mutations prior to thelarche are not excluded.

P121

The Role of Peritumoural, Peripheral and Intratumoural Lymphatics (PTL, PPL and ITL) in Lymph Node Metastasis of Primary Breast Carcinoma

{P} R.A.A. Mohammed[1], EC Paish[2], AR Green[3], IO Ellis[2], SG Martin[1]
[1]. University of Nottingham,City Hospital, Clinical Oncology Department, Nottingham, United Kingdom, [2]. University of Nottingham,City Hospital, Histopathology Department, Nottingham, United Kingdom, [3]. University of Nottingham,Queens Medical Center, Histopathology Department, Nottingham, United Kingdom

Metastasis of breast carcinoma to lymph nodes (LN) is a key event in tumour progression. Many studies reported that metastasis to LN occurs through PTL and PPL while ITL do not play a major role. To investigate this issue, sections from 125 primary breast carcinomas were stained immunohistochemically with podoplanin. Lymph vessel density (LVD) and presence of lymphovascular invasion (LVI) were examined in three tumour zones; PT, PP and IT and then correlated with LN status. Although all specimens contain PTL and PPL only 55/125 (44%) contained ITL. The mean LVD (per mm^2) of PPL, ITL, PTL and total LVD were 0.45, 0.33, 1.02 and 1.73 respectively and all were significantly associated with presence of LN metastasis. LVI was detected in 38/125 specimens, 29 of which had LVI in PPL and PTL while in 9 specimens additional LVI was in ITL. 14/29(48.27%) of cases with LVI in PPL and PTL have LN metastasis while 7/9 (77.7%) with additional emboli in ITL were LN positive
Conclusions: Increased LVD and presence of LVI are significantly associated with LN metastasis of breast carcinomas. ITL are present in a subset of tumours and invasion of these vessels appears to increase the probability of metastasis.

P122

Lupus Mastitis

{P} S Patil, S Malhotra, N Arsenovic
Dept of Histopathology,PathLinks, County Hospital, Lincoln, United Kingdom

Aim: Lupus mastitis (LM) is a rare subset of lupus erythematosus panniculitis limited to breast. It occurs in 2-3% of patients with Systemic lupus erythematosus (SLE). English literature review revealed less than ten cases of lupus mastitis.
Case report: A 33-year-old white woman with history of SLE presented with pain and swelling in right breast. Her double stranded DNA antibody level was 98 IU/ml.
The microscopic examination of breast skin showed an atrophic epidermis with extensive hyaline necrosis of the underlying fat. There was an associated lymphocytic infiltrate surrounding necrosis and within the lobular septae. Characteristic microcalcification of vessel wall, fibrinoid necrosis & perivascular lymphocytic infiltrate were seen.
Lupus mastitis may be clinically mistaken for carcinoma of breast (particularly inflammatory and scirrhous carcinomas). The other differential diagnoses include lipoatrophy with heterogeneous calcification, morphea & granulomatous angiopanniculitis.
Conclusion: This case highlights the awareness of existence of LM for histopathologists and they should consider the possibility of this diagnosis when microscopy shows a lymphocytic mastopathy with microcalcifications in the vessel walls and vasculitis. Correlation with mammography and lupus serology in such cases would help establish the diagnosis, which would allow clinicians to initiate appropriate therapy for the patient.

P123

Characteristics Of Clinically And Ultrasonically Negative But Histologically Positive Axillary Nodes In Patients With Breast Cancer

{P} A Hanfi, MS Bashir, J Jeffree, Z Saad
SALFORD ROYAL HOSPITALS NHS TRUST, HOPE HOSPITAL, SALFORD, NORHTWEST REGION, United Kingdom

Biopsy of sentinel node is becoming a standard procedure in patients with Primary Breast cancer. Clinical examination is carried out to detect any axillary lymphadenopathy with ultrasound as an adjunct tool. Patients with negative lymph nodes on ultrasound and/or clinical examination are offered a sentinel lymph node biopsy.

We did a retrospective study of Primary Breast cancer patients, operated in Hope hospital from June 2003-January 2006, and found 21 such cases where axillary lymph nodes were negative on ultrasound and clinical examination but showed metastases when examined histologically. We studied the pathological characteristics of these nodes.

95% (n=20) of cases showed involvement of 1-3 nodes with 80 % (n=16) showing involvement of single lymph node. In 20 % (n=4) of these cases, the size of metastases was less than 2 mm. Another 70 %(n=14) were in the range of 2-10mm and in only 10 % (n=2) of cases, it was greater than 10mm. In 15 % (n=3) of cases, the size of the involved lymph nodes was less than 5 mm. The size of the nodes in this group ranged from 2.5-18.4mm. Tumours of grade I, II, & III were present in a proportion of 19%, 47% and 19%, respectively.

Physical examination when combined with axillary ultrasound yields a low percentage of false negatives. In 80% of these cases, a single positive node was identified and except one almost none of them had more than 3 positive nodes. The possibility that these nodes represent sentinel nodes cannot be excluded.

P124

Metaplastic breast carcinomas are basal-like tumours

{P} JS Reis-Filho[1], F Milanezi[2], D Steele[1], K Savage[1], PT Simpson[4], JM Nesland[5], EM Pereira[6], SR Lakhani[4], FC Schmitt[3]
[1]. The Breakthrough Breast Cancer Research Centre, Institute of Cancer Research, London, United Kingdom, [2]. Life and Health Sciences Research Institute (ICVS), School of Health Sciences, University of Minho, Braga, Portugal, [3]. IPATIMUP – Institute of Molecular Pathology and Immunology, University of Porto, Porto, Portugal, [4]. Molecular & Cellular Pathology, Mayne Medical School, University of Queensland, Brisbane, Australia, [5]. The Norwegian Radium Hospital, University of Oslo, Montebello, Norway, [6]. Laboratório Salomão & Zoppi, Sao Paulo, Brazil

An immunohistochemical panel comprising antibodies against HER2, oestrogen receptor (ER), epidermal growth factor receptor (EGFR) and cytokeratin (Ck) 5/6 has recently been reported to accurately identify basal-like breast carcinomas, as defined by cDNA microarrays. Our aim was to analyse a series of metaplastic breast carcinomas (MBCs) using this panel plus two other basal markers (Ck 14 and p63) and progesterone receptor (PgR), to further define how frequently MBCs show a basal-like immunophenotype. 65 MBCs were retrieved from the pathology archives of the authors' institutions and reviewed by 3 of the authors. Immunohistochemistry with antibodies for HER2, ER, EGFR, Ck 5/6, Ck14, and p63 was performed according to standard methods. All but six cases (91%) showed the typical immunoprofile of basal-like tumours (ER and HER2 negative, EGFR and/or Ck 5/6 positive). When Ck 14 and p63 were added to the panel, 2 additional cases could be classified as basal-like. The majority of MBCs lacked PgR, except 4/19 (21%) carcinomas with squamous metaplasia. Our results demonstrate that MBCs show a basal-like phenotype, regardless of the type of metaplastic elements. Moreover, as these neoplasms frequently overexpress EGFR (57%), patients with MBC may benefit from treatment with anti-EGFR drugs.

P125

Immunoprofile of Invasive Tubular Carcinoma, Tubulolobular and Invasive Lobular Carcinoma and their Co-existent Putative Precursor Lesions

{P} Tarek Abdel-Fatah[1], Desmond Powe[1], Zsolt Hodi[2], Andrew Lee[2], Ian Ellis[1]
[1]. Nottingham University, Nottingham, United Kingdom, [2]. Nottingham City Hospital, Nottingham, United Kingdom

Aim This study was undertaken to determine the morphological and immunophenotypic features of putative precursor lesions involved in the development of some special types of breast carcinoma: invasive tubular (TC), tubulolobular (TLC) and lobular carcinoma (ILC).
Method: Immunoprofiling of tissue microarrays containing 1000 cores of invasive, terminal duct lobular units (TDLUs) and co-existent precursor lesions was performed by examining the expression of luminal, basal and myoepithelial markers, ER-α and β, Her2-neu, MIB-1, Cyclin D1, p53, Bcl-2, E-cadherin and FHIT. Fluorescence in-situ hybridization (FISH) was performed to detect amplification of the Her-2 gene.
Results: All TC, TLC and ILC expressed luminal markers. E-cadherin was reduced in TC and absent in lobular neoplasia (LN) and ILC. Cyclin D1 and the ratio of ER-α to ER-β increased in a stepwise manner from TDLUs: CCLs: in-situ: invasive lesions. Bcl-2 and FHIT were reduced in invasive lesions. The aforementioned lesions show no or low expression of MIB-1, p^{53} and HER-2.
Conclusion: The similarity in immunoprofile and co-existence of low grade DCIS, CCLs and LN in TC, TLC and ILC suggests that these represent a family of low grade intraepithelial precursors and invasive lesions. This suggests low grade breast carcinoma develops via CCLs, DCIS and LN.

P126

A case of adenomyoepithloma of the breast (tubular variant) presenting on screening mammography

{P} Y. A. MASANNAT, S. MUSA, M. DILANI, G. COAST, A. HANBY, A. M. SHAABAN
Leeds General Infirmary, Leeds, United Kingdom

Adenomyoepithelioma is a rare tumour with luminal epithelial and myoepithelial differentiation. Here we present a mammary adenomyoepithelioma excised from a 70 year old female whose lesion was detected by breast screening. The 20mm lesion was seen on previous mammogram but felt to have increased in size. FNAC showed a cellular specimen with nuclear atypia and features suggestive of carcinoma. Core biopsy was suggestive of benign adenomyoepithelioma. Wide local excision was done with no postoperative complications. The lesion was grayish/white and lobulated. Microscopy show a co-proliferation of tubular and myoepithelial elements infiltrating breast tissue. No significant atypia or mitotic activity was seen. Benign adenomyoepithelioma is a rare tumour that has epithelial and myoepithelial components. Spindle cell, tubular and lobulated variants have been described. Treatment is surgical excision with clear margins as the lesion can recur. The epithelial, myoepithelial or both components may become malignant.

P127

Do dyes used in sentinel node biopsy affect immunocytochemistry of hormonal receptors in breast cancer cells?

{P} Y. A. MASANNAT[1], G. COAST[2], V. SPEIRS[3], A. M. SHAABAN[2], P. JACKSON[2], K. HORGAN[1], A. HANBY[2]
*[1]. Department of Breast Surgery, Leeds General Infirmary, Leeds, United Kingdom, [2]. Department of Pathology, Leeds General Infirmary, Leeds, United Kingdom, [3]. *** Leeds Institute for Molecular Medicine, University of Leeds, Leeds, United Kingdom*

Sentinel Lymph Node Biopsy is the procedure of choice for axillary staging in clinically node-negative breast cancer. Nodes localization is achieved with dyes, radioactive tracer or both. The biological effect of the dyes materials has not been fully investigated.
Aims: To determine the effect of localisation dyes; 2.5% Patent Blue V (PBV), 1% Methylene Blue (MB) and 0.4% Indigocarmine (IDC) on immunocytochemical expression (ICC) of oestrogen (ERα) and progesterone receptor (PR) on a known ERα and PR positive mammary cell line. Methods: The MCF-7 breast cancer cell line was treated with the dyes PBV, MB and IDC in three dilutions, 1:10, 1:100 and 1:1000 for 4hours and 24hours. Subsequently, embedded cell blocks were prepared from cell pellets, stained for both ERα and PR (ER-6F11 and PgR636) and scored by two breast histopathologists using the Allred score.
Results: A marked decrease in ERα and PR immunoreactivity with higher concentrations of MB dye (1:10 dilution of the 1% MB preparation), but not PDV and IDC was observed.
Conclusion: Our data show that *in vitro*, MB can interfere with the results of ERα and PR ICC. If a similar effect is seen *in vivo* it could lead to inappropriate choice of antihormonal treatment.

P128

Reliability of core biopsy in pre-operative diagnosis of papillary lesions of the breast

R Swamy, {P} J Loane, J Thomas
Western General Hospital, Edinburgh, United Kingdom

NHSBSP guidelines recommend that most core biopsy-confirmed papillary breast lesions are excised because of possible sampling adequacy and lesional heterogeneity. Recent literature proposes conservative management of benign papillary lesions, suggesting that core biopsy histology predicts accurately excision histology.
We examined all papillary lesions diagnosed on core biopsy with subsequent excision biopsy (n=66) from a large breast screening centre between 2001 & 2005. NHSBSP guidelines were used for categorisation of core biopsies, allowing for a B2 diagnosis on cases without proliferation. Core biopsy diagnoses were later correlated with their paired excisions.
Core biopsy predicted accurately benign or malignant excision histology in most cases but failed to identify one of 5 papillomas with atypia

Core Biopsy Designation

Excision Diagnoses	B2	B3	B4	B5
Papilloma	15	6	0	0
Papilloma with DEH	7	11	0	0
Papilloma with atypia	1	4	0	0
Papillary carcinoma	0	1	3	18

We discuss core biopsy sampling, lesional heterogeneity and the definition of atypia in papillary lesions.

We conclude that it is premature to depart from the NHSBSP recommendations for core biopsy reporting of breast papillary lesions. It is timely to pool similar case series from other institutions to improve our understanding of the clinical implications and management of breast papillary lesions.

P129

The value of intra-operative axillary sentinel lymph node (SLN) imprint cytology preparations in the diagnosis of metastatic breast carcinoma.

{P} S Angra, A C Girling, R Y Ball
Norfolk & Norwich University Hospital NHS Trust, Norwich, United Kingdom

Aims and Objectives: To assess the value of axillary SLN imprint cytology preparations in the intra-operative diagnosis of metastatic breast carcinoma and to identify limitations of the technique.
Study Design: Cytopathology reports for axillary SLN imprint preparations from August 2004 to March 2006 were reviewed and compared with the histopathology reports on the nodes.
We compared the intra-operative report, based on DiffQuik staining (and which determined whether the axillary nodes were cleared), with the final report, which included preparations stained with Giemsa and for cytokeratins. In addition, we correlated the final cytopathology and histopathology reports.
Results : 152 cases were reviewed; in 34 of them metastatic carcinoma was identified on histological examination of the SLNs. Fifteen (44.1%) of these cases were detected by intra-operative cytopathology (overall intra-operative sensitivity = 64.2%). In a further 6 cases, malignant cells were recognised on immunostaining of the imprint preparations (final cytopathology sensitivity = 72.3%). No false positive cytopathology reports were issued. Of the 19 intra-operative false negative cases, 14 were from invasive carcinomas of lobular type and 14 of the cases proved to be micrometastases histologically.
Conclusion: Intra-operative assessment of SLN imprint cytology detected nearly half of the cases of metastatic breast carcinoma in this series. Most false negative results were cases of metastatic lobular carcinoma or micrometastases. Cytokeratin immunostaining added information in only 6 of the 19 cases reported as negative in the intra-operative setting. However, this was not clinically relevant in the operative time scale.

P130

Three Dimensional Reconstruction Of Breast Carcinomas Using Routine Laboratory Equipment & Immunohistochemistry

{P} RO Jones[1], AR Green[1], T Kurien[1], EC Paish[1], J Maddison[2], IO Ellis[1]

[1]. University of Nottingham, Nottingham, Nottinghamshire, United Kingdom, [2]. Medical Solutions PLC, Nottingham, Nottinghamshire, United Kingdom

This study used a simple technique to produce 3D reconstructions of breast carcinomas from histological sections using basic laboratory equipment to evaluate and characterise the spatial arrangement of the parenchymal cells of the breast. Case #1 was reconstructed using 128 4μm sections (20μm apart), immunohistochemically stained for AE1/AE3, c-erb-B2 and P63. Case #2 was reconstructed by using 47 serial sections and immunohistochemically stained for AE1/AE3, P53, and a cocktail of P63/SMM. Sections were digitally imaged using a flat bed scanner, at 1200dpi resolution, linked to a conventional PC. Accurate alignment of the images was carried out and the volume was reconstructed using maximum, minimum point projection and ''back to front'' opacity blending. The quality of the reconstructed images was clear, providing a comprehensive and explicit view of the normal and malignant parenchymal tissues of the breast that is not possible by viewing 2D histological sections. A novel subtractive technique was employed to differentiate between the invasive carcinoma, DCIS and normal tissue. This simple and reproducible approach enables the spread and infiltration of invasive carcinoma to be understood and could also be used to analyse the spatial relation between atypical hyperplastic and malignant in situ lesions of the breast.

P131

The Prognostic & Predictive Significance of Oestrogen Receptor Beta in Ductal Carcinoma In Situ

{P} INC Okonkwo, AR Green, EC Paish, IO Ellis

University of Nottingham, Nottingham, Nottinghamshire, United Kingdom

In situ breast tumours are benign precursor lesions of invasive breast cancer. The mitogenic action of steroids and dysregulation of steroid receptor expression may significantly influence disease pathogenesis, prognosis and therapeutic response. This study characterised expression of Oestrogen Receptors (ER)-α, -β and Progesterone Receptor (PR) in ductal carcinoma in situ (DCIS) using immunohistochemistry (n=55). Furthermore, gene expression of ER-α, ER-αΔ3, ER-αΔ2-3, ER-β1 and ER-β2 was quantitated in laser microdissected DCIS using real-time PCR (n=11). Results were compared with steroid receptor expression in lobular carcinoma in situ (LCIS; n=52). In DCIS, ER-β protein was more commonly expressed than ER-α or PR. Gene expression of ER variants were also found in DCIS. There were no differences in steroid receptor expression between DCIS which lacked associated invasive tumour compared with those that did. Significant differences in nuclear protein expression between ER-α and ER-β1 gene was observed between DCIS and LCIS. These findings suggest that ER-β might have a major biological role in DCIS along with ER splice variants. However, steroid receptor expression may not be directly associated with disease progression. The lack of similarity in steroid receptor expression between DCIS and LCIS may indicate fundamental differences in their biology.

P132

Gene Expression Signatures Of Oestrogen Receptor Positive And Negative Breast Tumour Cells

{P} R Day, AR Green, EC Paish, IO Ellis

University of Nottingham, Nottingham, Nottinghamshire, United Kingdom

Oestrogen is a potent mitogen in breast cancer and its action is mediated via Oestrogen Receptor (ER). This study has compared global levels of gene expression between ER positive and ER negative cell populations isolated from the same breast tumour cases in order to establish ER regulated genes. ER+ and ER- breast tumour cells were identified in frozen tissue sections rapidly immunostained using an ERα antidody (ID5 Dako) and isolated using laser microdissection in duplicate. Total RNA was extracted and amplified using two rounds and labelled with Cy3 or Cy5 using the Amino Allyl MessageAmp II aRNA Amplification Kit. Differences between gene expression of ER+ and ER- cells were analysed using a Human 30K MWG pan oligonucleotide microarray. The expression of several genes were altered more than 2-fold between ER+ and ER- tumour cells commonly identified in all cases. These included up-regulation of Group X secretory phospholipase A2 precursor cells, thyroid hormone receptors alpha and beta and lecithin-cholesterol acetyltransferase in ER+ tumour cells. This study has successfully identified several genes that are known to be regulated via ERα, as well as some novel gene candidates. Their relevance and role in breast cancer remains to be determined.

P133

Expression of invasion promoting Tn-C isoforms in DCIS

{P} K Lloyd[1], JH Pringle[1], RA Alcock[1], JL Jones[2], RA Walker[1], JA Shaw[1]

[1]. University of Leicester, Leicester, United Kingdom, [2]. Barts and The London, London, United Kingdom

We have previously identified two tumour specific isoforms of Tenascin C (Tn-C) in breast cancer tissues, containing either exon 16 (Tn16) or exons 14 and 16 (Tn14/16), which promote tumour cell invasion. The aim of the study was to examine the precise cellular localisation of these isoforms in cases of ductal carcinoma in situ (DCIS). 13 cases, 2 with small areas of invasion, were assessed. mRNA *in situ* hybridisation (ISH) was carried out using isoform-specific digoxigenin labelled single stranded DNA probes. Probe specificity was confirmed by ISH to transfected MCF7 cells expressing each isoform. Protein expression of total Tn-C and Tn-C FnIIIB was examined by immunohistochemistry (IHC) using BC24 (Sigma) and αIIIB (Chemicon) antibodies respectively. 10 of 12 cases showed variable positive staining by ISH and IHC. In 8 cases Tn16 and Tn14/16 mRNA were expressed in occasional myoepithelial cells and stromal fibroblasts surrounding DCIS. 4 cases showed strong staining of malignant luminal epithelial cells as well as occasional myoepithelial cell staining. 2 of these 4 cases, which had small areas of invasion, showed clear labelling of malignant cells in DCIS and strong staining of invading tumour and surrounding stroma for both isoforms. These different patterns of Tn-C isoform expression may influence early tumour cell invasion and lead to disease progression.

P134

Mammotome Biopsy in the Management of Screen-Detected Breast Lesions: Experience of a Large Screening Centre

{P} V Kumaraswamy, J Liston, G Coast, S Lane, A Hanby, A Shaaban
Leeds General Infirmary, Leeds, United Kingdom

Aims: Mammotome biopsy is a minimally invasive, image guided procedure that yields larger quantities of tissue for diagnosis compared to a core biopsy. We aimed to audit the indications for mammotome biopsies and the clinical usefulness of the technique by analyzing the diagnoses that were modified with mammotome biopsies compared to the previous core biopsies.
Method: All patients who underwent mammotome biopsy were identified from the database of the Breast Screening Unit. The histopathology reports of all the mammotome biopsies done in Leeds from inception in September 2002 to May 2005 were reviewed. The diagnoses on a precedent core biopsy were compared to the diagnoses on the mammotome biopsy.
Results: 60 cases were performed with an average patient age of 56 years. All except 8 cases (the learning curve) were preceded by a core biopsy. The indications for mammotome biopsies were to identify the nature of calcification; a core biopsy diagnosis of B1, B3 (in particular columnar cell atypia and atypical intraductal proliferation) or B4; discordance between radiology and pathology on the original core and where a benign core was felt not to be representative. Of the 52 cases with previous core biopsies, 39 had a modification of diagnosis after mammotome biopsy. 15 new cases of B5a were diagnosed; 5 of which were B1 (inadequate) on the original core, 2 were B2, 3 were B3 and 5 were B4. One case of invasive carcinoma (B5b) was diagnosed on mammotome biopsy without a preceding core.
Conclusion: Mammotome biopsy is a useful technique that has the potential of saving patients unnecessary/multiple surgery. In a reasonable proportion of cases it helps to arrive at a definitive diagnosis with consequent better planning of patient management. It is particularly useful in cases with unrepresentative/inadequate initial sample.

P135

Epithelial apoptosis detected by cytodeath (M30) immunohistochemistry correlated with BCL2 expression in infiltrating ductal breast carcinoma.

{P} Paul Ryan[1], Calvin Coffey[3], Ashling O'Connor[3], Gordon Watson[3], Robert Landers[2]
[1]. Dept of Histopathology, Cork University Hospital, Cork, Ireland, [2]. Dept of Histopathology, Waterford regional Hospital, Waterford, Ireland, [3]. Dept of Surgery, Waterford Regional Hospital, Waterford, Ireland

Background: Regulation of apoptosis plays a role in the growth and progression of solid tumours. Cytodeath (M30) is a new antibody which selectively identifies epithelial apoptosis. Bcl2 is an antiapoptotic protein expressed in normal breast ductal epithelium. This study assessed apoptotic and antiapoptotic protein expression in infiltrating ductal carcinoma.
Methods: Archival cases (n = 48) of infiltrating ductal carcinoma were stained immunohistochemically for M30 and BCL2 and evaluated by light microscopy. Correlation with 5 year survival was also determined. Control normal breast and fibroadenoma samples (n = 18) were similarly stained.
Results: M30 immunoreactivity was observed only in grade 3 carcinomas (9/33=27%), and was not seen in grade 1 or 2 tumours, fibroadenomas, and normal breast duct epithelium. BCL2 was expressed in 100% control tissue and decreased with increasing grade. In grade 3 tumours M30/BCL2 co-expression was associated with 88% 5year survival, versus 50% 5year survival in M30/BCL2 negative cases.
Conclusion: High grade ductal carcinoma of breast shows increased apoptosis and decreased antiapoptosis marker expression. The findings indicate a possible phenotype associated with improved outcome within the subgroup of grade 3 tumours but correlation with other known prognostic factors in prospective trials appears warranted.

P136

An Audit of Breast Core Biopsies Performed for Radiological Architectural Distortion: Is Irregular Fibrosis an Acceptable Histopathologic Result?

{P} I Tosounidis, SA Pritchard
South Manchester University Hospitals NHS Trust, Manchester, United Kingdom

Introduction: Focal fibrosis of the breast has been described in the radiological literature but there is little information in pathology journals on correlation of core biopsy findings in cases of mammographic architectural distortion. Within the department we noticed many core biopsies carried out for such stromal abnormalities (usually category R3) showed a characteristic form of fibrosis that was dense and irregular.
Aim: To determine the frequency and characteristics of fibrosis diagnosed at core biopsy and to compare the histological and imaging features.
Methods: All core biopsies performed between January and May 2005 were retrospectively reviewed. Cores taken from patients with architectural distortion on imaging that showed fibrosis only microscopically were reviewed histologically. Follow up of pathological data was reviewed when available.
Results: 489 breast core biopsies were performed. 53 were taken for architectural distortion. In total 60 cases showed fibrosis as the only pathological finding, (category B1), of these 30 were from cases of architectural distortion. These were reviewed and 26 showed similar features with dense, hyalinised fibrosis that appeared irregular and disrupted resembling pieces of a jigsaw puzzle. We termed this irregular fibrosis. On follow up 2 cases had cytology that was benign and 4 cases had further histology, 3 of which revealed a malignant diagnosis.
Conclusions: The irregular fibrosis described correlates with focal breast fibrosis described in the radiological literature. It was found in 87% of cases taken for architectural distortion, pathology category B1. Irregular fibrosis is a histopathologic entity and is an acceptable histopathologic result of core biopsy in cases of architectural distortion following careful radiologic-histopathologic clinical correlation. It is debatable whether this should be classed as category B1 or B2.

P137

Unusual Malignant tumours of the breast

{P} Y Alizadeh, A Shaaban
Department of Histopathology, Leeds General Infirmary, Leeds, United Kingdom

INTRODUCTION: Breast cancer is the commonest non-skin cancer in females. The majority of breast malignancies are carcinomas and very few are non-epithelial tumours. The aim of the study was to evaluate the malignant non-epithelial neoplasms diagnosed in Leeds General Infirmary (LGI) in the past 10 years.

MATERIALS AND METHODS: Cases with a diagnosis of non-epithelial breast malignancy were identified within the databases of the Department of Pathology and HMDS in LGI. The reports of the patients with the diagnoses of non-epithelial breast tumours between 1995 and 2005 were reviewed. Age, gender, clinical details, type of specimen, diagnosis and where available the size, margins and grade of the tumours were recorded.

RESULT: A total of 25 patients were identified, all of them were females. The median age at time of presentation was 63 years (range 19-97). Out of 25 patients, 23 presented with breast lump, of which two had other lesions (ovarian lesion and splenic vessel abnormality). Two patients presented with mammary skin lesions of which one was a recurrent angiosarcoma in a mastectomy scar. The size of the lesions ranged from 5-150mm (median 67mm). The most common diagnosis was lymphoma (11/25) including follicular, marginal zone, diffuse large B-cell lymphoma and o mycosis fungoides. This is followed by angiosarcoma (9/25), and sarcomas (2/25). Sarcoma arising in phyllodes tumour (1 case), plasmacytoma (1/25) and malignant fibrous histiocytoma (1/25) were also identified.

CONCLUSION: Malignant non-epithelial tumours were rare and represented approximately 3.84% of all malignant breast tumours received in our institution.

P138

The Significance Of Lobular Neoplasia On Needle Core Biopsy Of The Breast

{P} S Menon, GJR Porter, AJ Evans, IO Ellis, Z Hodi, CW Elston, AHS Lee
Nottingham City Hospital, Nottingham, United Kingdom

The significance of lobular neoplasia of the breast is controversial. There is some evidence that it is a risk factor for invasive carcinoma in both breasts and some that it is a precursor for invasive carcinoma at the same site. This study was performed to examine the significance of lobular neoplasia in breast needle core biopsies.

39 breast cores from July 1998 to May 2005 showing lobular neoplasia with no DCIS, invasive carcinoma, atypical ductal hyperplasia or pleomorphic LCIS were included in the study. Detailed histopathological and radiological review was performed.

Surgical excision was performed in 25. Carcinoma was found in 10 (7 invasive and 3 DCIS): radiological/pathological review showed that the core missed a mass in 6 and calcification in 3. 6 patients had radiological follow up of at least 2 years. Carcinoma developed in 2 (1 invasive and 1 DCIS). In one of these the carcinoma was mammographically occult.

In conclusion most of the cases with carcinoma after a core biopsy showing lobular neoplasia were the result of the core missing the radiological lesion. This emphasises the importance of clinicopathological review and further investigation of any discordance.

P139

Backpain: An Unusual First Presenting Sign Of A Primary Breast Liposarcoma

{P} S Nagaraju, M Powari, R Marshall, J Mathew
Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom

Aim: To describe an unusual first presentation of a breast liposarcoma.
Case report: A 44 year-old female presented with an 8-month history of back pain. An MRI examination suggested infiltrative pathology of the vertebra bodies.
The peripheral blood showed leukoerythroblastic picture. Bone marrow examination was done. The bone marrow smear showed non-haematopoietic vacuolated cells. The trephine biopsy showed complete replacement of marrow spaces by tumour cells with focal alveolar pattern, increased vascularity and some cells showing cytoplasmic vacuoaltion.
Subsequently, on further work up, a non palpable breast mass was detected by mammogram and ultrasound. A lumpectomy was done: this showed high grade sarcoma with cytoplasmic vacuolation and occasional lipoblasts. The breast mass and the bone marrow metastasis were reported as Trojani grade 3 liposarcoma.
Conclusion: <1% of breast tumours are sarcomas but between 3 to 24% of these are liposarcomas. They often arise against a background of mixed tumours. Although extra-pulmonary metastasis is common with liposarcomas, metastasis to bone marrow is rare; even rarer still is the presentation of this disease as backpain with a non-palpable breast lesion.

P140

How Does Recurrent Breast Cancer Compare To Its Primary?

{P} M Manickavasagom[1], O Taiwo[1], D Wheatley[2], J Mathew[1]
[1]. Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [2]. Department of Oncology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom

Aim: To compare the histologic appearances of recurrent breast cancer to its primary.
Material and methods. Patients attending the oncology clinic for treatment of recurrent breast cancer over a period of one year were identified and their hospital numbers used to identify the histology of the "recurrent" cancer against the primary lesion.
Results: 76 patients were identified; data was available for 51 patients. Average age of first diagnosis was 59.1yrs (33.8-85.7yrs) and recurrence was 64.7yrs (36.6-87.8yrs); time to recurrence was 5.7yrs (0.07-14.5yrs). The primary breast lesion compared to the histology of the recurrent breast lesion is shown in table 1; in table 2, the grade of the primary lesion is compared to the recurrence (nm=not mentioned). The histology and grade of the primary and recurrent lesion were broadly similar. Histologic type variation between invasive ductal (IDC) and lobular (ILC) is attributed to mixed pattern tumours. Occasional tumours appear to have a poorer grade than its primary and vice-versa; this could reflect observer variation or reflect clonal selection within a tumour in some instances.

Table 1		Histology: first biopsy			
		ADH	DCIS	IDC	ILC
Histology second biopsy	ADH	0	0	0	0
	DCIS	0	1	1	0
	IDC	1	3	36	2
	ILC	0	0	1	6
Total		1	4	38	8

Table 2		Grade of first biopsy					
		GR1	GR2	GR3	DCIS	ADH	NM
Grade of second biopsy	GR1	2	1	0	0	0	2
	GR2	6	10	5	2	1	1
	GR3	0	5	9	0	0	1
	DCIS	1	0	0	0	0	0
	NM	0	2	2	0	0	1
Total		9	18	16	2	1	5

Conclusion: Recurrent tumours, for the most part tend to remain true to the type and grade of its primary.

P141

Needle Core Biopsy Diagnosis of Papillary Breast Lesions

{P} N Sahasrabudhe, JD Coyne, MC Howe
Wythenshawe Hospital, Manchester, United Kingdom

Papillary lesions (PLs) of the breast often show intralesional heterogeneity, potentially increasing the risk of misdiagnosis on needle core biopsy. This study aimed at assessing the reliability of core biopsies in the evaluation of PLs.
All B3/B4 category core biopsies in the period 1998-2004 (inclusive) were considered. In all those containing PLs the diagnosis was compared with the subsequent excision specimen where available. A total of 294 biopsies were classified as B3. Of these, 23 cases of PL with a subsequent excision specimen were identified. On core biopsy 19 showed papillomas without atypia. The subsequent excisions contained benign papillomas (13) and other benign breast lesions (6). In one excision specimen there was an incidental focus of low grade DCIS separate from the papilloma. 4 cases were diagnosed as papillomas with atypia on core biopsy. The excisions contained papillomas with focal atypia (2) and low grade DCIS (2).
There were 61 cases designated as B4 and a PL was present in 5. The excision specimens contained intracystic papillary carcinoma (3) and micropapillary DCIS (2).
A diagnosis of PL with atypical/suspicious features on core biopsy reliably predicts atypia/malignancy in the subsequent excision specimen. The absence of atypia/malignancy within the excised PLs that were diagnosed as papillomas without atypia on core biopsy suggests that there may be little justification for excising this subgroup.

P142

Can sudden cardiac deaths be predicted?

Debbie Chase, Hayley Burnley, {P} Patrick J Gallagher, Paul Roderick, Paul Roberts, John Morgan
Southampton University Hospitals, Southampton, United Kingdom

The objective of this study was to identify which, if any, Sudden Cardiac Death (SCD) victims could have been identified before their death and considered for an implantable cardioverter defibrillator (ICD) on the basis of their previous cardiac history.
Autopsy proven cases of SCD were identified over a 1 year period in a defined population. They were categorised according using a modification of Davies' method. Those with evidence of myocardial scarring, coronary narrowing or ventricular hypertrophy may have died because of ventricular arrhythmia. However cases with clear evidence of an acute coronary thrombosis or acute myocardial infarction were excluded. Hospital and GP records provided information on previous symptoms, investigations and past history of previous cardiac disease. Two cardiac electrophysiologists judged the appropriateness of each case against pre-defined criteria
366 of 975 post mortems were sudden cardiac deaths and in 232 of these there was no evidence of acute myocardial infarction or coronary thrombosis (63% of all SCDs).215 lived within the catchment area, 64% were male, the median age was 75 and 37% of events were witnessed. Agreement between experts on appropriateness for an ICD was good (kappa score 0.64). One case was considered appropriate for an ICD. 41% of cases had no evidence of confirmed or suspected heart disease or arrhythmia and were not therefore appropriate for investigation. 49% of the remainder had previous cardiac events or symptoms suggestive of ventricular arrhythmias but were not referred for further investigations. In particular some patients with a previous myocardial infarct were not referred for 24 hour ECG tracings and and some of those with suspected heart failure did not have echocardiography.
Our study has demonstrated that circa 40% of SCDs could not have been predicted. A significant proportion of the remaining patients were not referred for appropriate investigations. One patient should have received an ICD.

P143

Surgical Reporting Of Cardiac Valves – "It's All In The Macro"

{P} G L Rowlands, D F R Griffiths
University Hospital of Wales, Cardiff, United Kingdom

The gross descriptions of cardiac valve specimens can often indicate the aetiology of the valvular pathology, allowing a meaningful report from the pathologist, as the microscopy is often non-specific.

An audit of 195 cardiac valve reports over three years revealed 57% of cases had "non-specific" macroscopic description - as a consequence, 48% of cases had non-diagnostic microscopic conclusions.

A scoring system was designed to assess the quality of the gross descriptions. Points were given for the inclusion of various valvular components, such as chordae length and presence of vegetations. Up to nine points were available in total and a percentage score was calculated for each case, indicating the comprehensiveness of the macroscopy. Results: Mean Score 37.2%, Median Score 33.0%, and Range Of Scores 0-100%. Six components scored poorly (less than 50%) and the only component that had a relatively high score was "Number of Cusps" (86%).

The results showed most cases had sub-optimal scores for their gross descriptions, resulting in many non-specific reports, despite often having had special stains requested, to assist the microscopic reports. Thus there is a real need for pathologists to improve the gross descriptions of cardiac valve specimens in order to provide meaningful reports.

P144

Ruptured Acute Myocardial Infarction, Haemopericardium And Death: A Post Mortem Review.

{P} A Swaminathan[1], M Powari[1], K Kandasamy[2], J Mathew[1]
[1]. Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [2]. Department of Cardiology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom

Aim: to define some of the chararcteristics of ruptured acute myocardial infarction (RAMI), as a cause of haemopericardium (HP), in a post-mortem (PM) setting
Materials and Methods: The PM register and departmental database was used to identify all patients who died as a result of HP due to RAMI between 1995 and 2004 (inclusive).
Results: Of 14,368 PMs, 311 cases of HP-related RAMI were identified in a population of 458 HP-related deaths, with a male (n=120; mean age=74.2yrs) to female ratio (n=191; mean age=78.7yrs) of 1:1.6. Coronary artery thrombosis (CAT) was recognised in 209 PMs (82M, 127F); CAT was not identified in the remainder (n=102: 38M, 64F).
The ventricular rupture site was defined in all PM reports, with the anterior wall (33.8%) being the most common. Cumulative figures show anterior (42.8%) and posterior wall rupture (37.3%) being more common than apical (6%) or lateral (22.5%) rupture.
Males were more likely to have posterior (12.2%) than anterior (10.6%) or lateral (7.4%) ventricular wall rupture; females, by contrast, had anterior (23.2%) ventricular wall rupture more commonly than posterior (19.3%) or lateral (5.8%) wall rupture.
The volume of blood in the pericardial space was recorded in 166 (54%) instances (64M, 102F); it ranged between 150ml and 1000ml of blood. The volume of haemorrhage was, on average, greater in men (mean = 473ml) and women (mean = 418ml).
Conclusion: RAMI is the commonest cause of HP-related deaths and is often associated with CAT. It is commoner in males than females and occurs 4 years earlier in the former; the HP was 55ml higher in males. Although anterior wall rupture was commonest overall, and in females, posterior wall rupture was slightly commoner in males.

P145

The Diagnostic Yield Of Transbronchial Biopsy Compared With Endobronchial Biopsy In Pulmonary Sarcoidosis.

{P} AI Finall[1], R MacJanette[2], O Dempsey[2], KM Kerr[1]
[1]. Department of Pathology, Aberdeen Royal Infirmary, Aberdeen, Scotland, United Kingdom, [2]. Department of Respiratory Medicine, Aberdeen Royal Infirmary, Aberdeen, Scotland, United Kingdom

The diagnostic yield of endobronchial (EB) bronchoscopic biopsies has been reported as being lower than transbronchial (TBB) in Causasians. The British Thoracic Society suggest TBB is the gold standard for diagnosis. We analysed the increase in diagnostic yield from EB by performing a retrospective analysis of histopathological specimens. Patients had a radiological diagnosis of sarcoidosis, compatible clinical/biochemical features and follow-up. 35 patients had EB and TBB, 14 had TBB alone and 17 patients had EB alone. This resulted in 101 specimens (49 TBB and 52 EB). Overall, TBB had a diagnostic yield of 80% and EB of 54% and these results are statistically different (p=0.01). In four (11%) combined procedures EB was positive when the TB was negative, thereby adding to the diagnostic yield. We suggest that the diagnostic gain of 11% taking an EB in addition to TBB during a combined procedure outweighs possible side effects and reduces the possibility of a false negative result.

P146

The Effect Of Size And Number Of Transbronchial And Endobronchial Biopsies In Successful Tissue Diagnosis Of Patients With Pulmonary Sarcoidosis.

{P} AI Finall[1], R MacJanette[2], O Dempsey[2], KM Kerr[1]
[1]. Aberdeen Royal Infirmary, Department of Pathology, Aberdeen, Scotland, United Kingdom, [2]. Aberdeen Royal Infirmary, Department of Respiratory Medicine, Aberdeen, Scotland, United Kingdom

British Thoracic Society guidelines state that 4-6 fragments are required for optimum diagnostic yield of pulmonary sarcoidosis in bronchoscopic transbronchial (TBB) and endobronchial biopsies (EB). We carried out a retrospective, observational study analysing the number and size of fragments submitted to the pathology laboratory for diagnosis, and investigated their relationship with a positive diagnosis. Our hypothesis was that the greater the number and/or size of fragments received, the more likely a positive diagnosis. 101 biopsy specimens were retrieved from patients with a diagnosis of sarcoidosis either proven on lung biopsy, confirmed by radiology and/or clinical follow-up and/or by non-pulmonary tissue biopsy. The number of fragments in the positive group for TBB did not differ statistically from the negative group ($p>0.05$). This was the same for EB ($p>0.05$), suggesting that number of fragments may not be related to diagnostic yield. The difference in size of fragments differed significantly between the positive and negative groups for both TBB and EB ($p=0.001$ and <0.05 respectively), indicating that larger fragment size significantly improves diagnostic yield. Further research may lead to changes in current BTS guidelines, including procedures to maximise fragment size as well as number.

P148

Appropriate SNOMED Coding In Histological Reporting – A Departmental Audit

{P} Paula Hyder, Emil Salmo
Trafford Healthcare NHS Trust, Manchester, United Kingdom

Introduction: SNOMED is an established and reliable catalogue of clinical terminology, which allows enforcement of high standards in patient care, and enables quality control measures and research. In our department, coding is performed by the histopathology secretaries at the time of typing the histology reports.
Aim: To ascertain whether SNOMED coding is being accurately carried out in our department, and if not, to identify ways in which this can be rectified.
Methods: A representative sample of 100 histology and cytology reports from 2004-2005 (selected at regular intervals of histology specimen number) were analysed and the coding reviewed.
Results: 74% of reports were coded satisfactorily or showed very minor inaccuracies. The remaining 26% of cases showed major inaccuracies and non-coding, including a seborrhoeic keratosis coded as Bowen's disease, 'proliferating smooth muscle' coded as leiomyosarcoma and non-coding for CIN2 in a LLETZ specimen.
Conclusion: Although the majority of cases were adequately coded, a significant minority showed major discrepancies in coding especially where reports show more than one abnormality. Where coding is performed by a person other than the reporting pathologist, the code(s) must be checked at authorisation. More care must be taken in ensuring accurate and relevant coding for histological reports.

Abstract P147 has been withdrawn

P149

Reporting Colorectal Cancer In Accordance With The RCPath Minimum Dataset Guidelines - A Departmental Audit

{P} Paula Hyder, Ammar Khatab, Emil Salmo, Najib Haboubi
Trafford Healthcare NHS Trust, Manchester, United Kingdom

Introduction: The evidence-based Minimum Dataset Guidelines for colorectal cancer incorporate information required for appropriate patient management, prognosis and quality control.
Aim: To assess accuracy of colorectal cancer reporting in our histopathology department in relation to Minimum Dataset guidelines.
Methods: Histology reports for all colorectal cancer resections received to our department in 2004 were reviewed (52 cases).
Results: Dukes and TNM stage, number of lymph nodes, local invasion, histological subtype, differentiation and lymphovascular invasion were mentioned in >95% cases. Apical node was mentioned in 32.7%. Background abnormalities were mentioned in 23.1%. Where staging for Dukes (1 case) and TNM (4 cases) were not given, the depth of invasion and presence of nodal metastasis were described in the text. Differentiation and histological subtype were not explicitly mentioned in 2 cases, namely a 'tubulovillous adenoma showing focal malignant transformation' and a 'malignant tubulovillous polyp'.
Conclusion: Deficiencies in narrative reporting exist, although the key information can nearly always be extrapolated from the text. However, reporting of apical node and background abnormalities is inadequate. Dataset forms should be used in conjunction with narrative reports to avoid such omissions, and a case exists for the latter being phased out to prevent unnecessary duplication.

P150

AUDIT OF ENDOPSCOPIC BIOPSIES FOR CONFIRMING UPPER GI CANCER DIAGNOSIS

R Swamy, {P} R Perera, HM Kamel
NHS Lothian, Edinburgh, United Kingdom

Currently, local rather than national protocols dictate biopsy sampling in the evaluation of oesophageal and gastric malignancies.
The aims of the audit were to assess the number of biopsies required for diagnosing upper GI cancer and to assess discrepancies in reporting tumour type and grade between biopsies and resection specimens.
A total of 129 resections over a two-year period, comprising 47 gastric and 82 oesophageal carcinomas were audited.
A definitive cancer diagnosis was made following a single endoscopic procedure in 81.6% and 93.3% of oesophageal and gastric cases respectively. Multiple biopsy sampling at same endoscopic procedure for evidence of intestinal and Barrett's metaplasia were taken in 9/ 47 cases (19%) of gastric cancer and 9/56 cases (16%) of oesophageal adenocarcinoma respectively. Multiple biopsies involving more than one endoscopic procedure to diagnose malignancy was more frequent in oesophageal (18.4%) than gastric cancer (6.7%). Correct grading on endoscopic biopsy was 73% and 53% for oesophageal and gastric cancers respectively whilst no discrepancies were observed in histotype

P151

Pathologists' perception of the significance of immunohistochemistry - A questionnaire study

{P} R Perera, F Rae, L Brett, HM Kamel
NHS Lothian, Edinburgh, United Kingdom

Immunohistochemistry is an established technique in histopathology contributing mainly in diagnosis, classification and prognosis of tumours and also in non neoplastic diseases such as demonstration of microorganisms. The aim of the study was to explore pathologists' perception of the contribution of immunohistochemistry tests in diagnosis within a teaching hospital setting. A total of 333 immunohistochemistry requests were made for a total of 1340 tests during the period of study. A total of 113 (33.93%) requests had completed questionnaires. Almost half (49.55%) of the responders indicated that request were essential for diagnosis, 32.74% to confirm a diagnosis and 17.69% to exclude a diagnosis. However, when asked to indicate what is the expected contribution of the requested tests, 72.56% of the responders felt they help to make a definitive diagnosis while 10.61% indicated they help to narrow a differential diagnosis. These findings perhaps indicate that histopathologists believe in a significant number of cases (33.62%) immunohistochemistry is a confirmatory rather than being diagnostic or essential by itself. Other uses that were indicated by respondents included 8.84% to identify a specific structure, 2.65% to plan management and 1.76% for prognosis.

P152

An Audit of Renal Transplant Biopsy Reports

V Etezadi[2], A Asderakis[2], {P} D F R Griffiths[1]
[1]. University of Cardiff, Cardiff, United Kingdom, [2]. Cardiff and Vale NHS Trust, Cardiff, United Kingdom

Renal graft biopsies are costly and potentially hazardous; their use and accuracy should be audited. A detailed review of each biopsy is beyond the resources of most units. However if biopsy use is appropriate and biopsy diagnosis accurate then the distribution of diagnoses should be within expected ranges.

We have analysed the results of clinically indicated graft biopsies reported in our department from January 2001 to October 2005. All biopsies were coded according to the Banff (97) criteria.

Of the 674 biopsies the frequencies of individual diagnoses were: Normal (24%), Borderline changes (12%), Acute Rejection (22%) [cellular (62%) & vascular (38%)], ATN (7%) and Chronic Allograft Nephropathy (21%). The occurrence of acute rejection was 33% and 28% within the first and second 6 months post transplant respectively. ATN was an extremely rare (<1%) diagnosis after the first 3 months post transplant and infrequent (15%) before that. Chronic Allograft Nephropathy had a gradual increase from 0% within 3 months post transplant to 48% after the first year.

This distribution is similar to previously published work and offers a benchmark against which other units can audit. Any substantial deviation from this distribution should be investigated in more detail (e.g. absence of vascular rejection might suggest inadequate sampling).

P153

Gastrointestinal Histopathology Of Limited Or No Clinical Value: Following The Guidelines – What Are The Risks?

{P} E L Porter, A Jeffery, A McGregor
Leicester Royal Infirmary, University Hospitals Of Leicester NHS Trust, Leicester, United Kingdom

Background and Aims: The publication "Histopathology and Cytopathology of Limited or No Clinical Value" by the Royal College of Pathologists recommends that biopsies from the GI tract should only be taken in specific contexts, not randomly. A previous audit showed a large number of "inappropriate" biopsies. The audit was extended and all inappropriate biopsies were reviewed to evaluate how many showed significant pathology.
Method: Information including clinical history, endoscopic appearance, number and site of biopsies, was collected over a 2 month period. Biopsies were categorised as appropriate or inappropriate based on the Royal College document. A borderline category was used for cases which did not clearly fit either category. Slides from all borderline and inappropriate cases were reviewed.
Results: 2078 lower, and 1735 upper GI biopsies were included in the study. 23% of lower and 29% of upper GI biopsies were inappropriate or borderline. On review of these, one case (one biopsy) showed significant changes. This biopsy showed coeliac-like changes in a patient who was not anaemic and had negative serology.
Outcomes: The risks of missing pathology by following the RCPath guidelines are very small. We have presented this data to gastroenterologists locally and are currently reviewing biopsy protocols.

P154

Immunofluorescence in Lupus Erythematosus Skin. Are we doing the right thing?

{P} M N D'Cruz, S Edward, M J D Goodfield
The General Infirmary at Leeds, Leeds, West Yorkshire, United Kingdom

Immunofluorescence (IF) is a major diagnostic tool in skin biopsies of suspected Lupus erythematosus (LE) for diagnosis as well as for subtyping. Though of utmost importance, IF may be equivocal in some cases. We aim to determine whether IF is a useful diagnostic adjunct in LE skin biopsies. We also seek to determine whether a negative or equivocal IF test makes a difference in clinical management and to determine the number of cases with equivocal IF which responded to LE treatment. We also propose to evaluate the usefulness of the site of the biopsy.
All cases with a diagnosis of LE were retrieved using the relevant SNOMED codes. During a 5-year study period, there were a total of 239 cases of which a clinical diagnosis of LE was made in 103 cases. In another 60 cases, the clinical diagnosis was also confirmed with a positive histopathological and immunofluorescence result. In a further 29 cases, though the histopathological features were consistent with LE, the IF was equivocal. In another 14 cases, despite a negative history, the diagnosis of LE was made on characteristic histomorphological and immunofluorescence studies.

P155

Audit Of External Referral Pathway At A Pathology Network

{P} S Patil, S Malhotra, D Clark
Dept. of Histopathology,PathLinks,County Hospital, Lincoln, United Kingdom

Aims:
To determine turnaround times for external referral cases
To see if the external referrals correspond to the departmental agreed referral pathologists' list
Method:
All histology referral case reports, excluding lymphomas were retrieved from the Pathology database and analysed for the period between Feb-July 2005.
Results:
A total of 50 nonlymphoma cases were referred to external pathologists for further opinions & the average total turnaround time (specimen received to final report authorised) was 44 days. The time between specimen reception and referral and between specimen referral to the report being typed at the referral hospital were 20 days each. The time between report typing at referral hospital and final authorisation was 10 days.
The provisional & final referral reports were similar in 68% of the cases and differed in some respect in the remainder.
Conclusions:
Some external referrals did not correspond to the departmental agreed referral pathologists' list.
The turnaround times for the external referral cases are higher than desirable.
The changes recommended were: to refer the cases ASAP following provisional diagnoses, SOPs in place for immediate referral and addition of final report, computerised maintenance of Referral register and, changes to the agreed referral pathologists' list.

P156

Audit Of Reporting Of Important Prognostic Parameters In Resected Colorectal Carcinomas

{P} S Katti, O Rotimi
Leeds General Infirmary, Leeds, West Yorkshire, United Kingdom

Aims: We aimed to audit the reporting of important prognostic parameters in resected colorectal carcinomas and study the effect of preoperative treatment on prognostic parameters.
Methods: 306 cases of resected colorectal cancer reports were audited for the incidence of serosal involvement, extramural vascular invasion (EMVI) and the number of lymph nodes retrieved.
Results: T4 tumours constituted 27.45% of all cases. EMVI was seen in 48.62% of all cases increasing to 77.38% in T4 stage tumours. The average number of lymph nodes retrieved was 18.
Nodal disease was seen in 80% of cases where 17-20 nodes were retrieved, compared to 41% with 9-12 nodes. The average number of nodes retrieved in rectal carcinomas with and without preoperative treatment was 14 and 19 respectively. Nodal disease was present in 34% of preoperatively treated cases compared to 43% in untreated cases. When more than 20 nodes were retrieved, 67% of preoperatively treated cases showed nodal disease
Conclusions:
Retrieving the recommended 12 lymph nodes may lead to under staging of disease. Retrieval of 20 nodes is perhaps ideal.
A high node yield in preoperatively treated carcinomas correlates strongly with nodal disease.
Incidence of EMVI reporting is high, particularly in T4 tumours at our institute

P157

How often do histopathologists get it wrong: Do trainees make more errors?

{P} JP Dormer, S Coates, AH McGregor
Department of Histopathology, Leicester Royal Infirmary, LEICESTER, United Kingdom

Supplementary histopathological reports (SRs) are issued in various circumstances from reporting further work to correcting errors. Previous studies have shown a serious error rate of 0 to 1.2%. An audit was undertaken to assess the use of supplementary reports, to evaluate the error rate and ascertain whether more mistakes were made by consultants working alone than when supervising trainees.

All supplementary reports from August 2004 to July 2005 were reviewed. Most supplementary reports were used to report additional work (67% immunohistochemistry, 9% special stains, 5% additional blocks, 2% molecular tests). The error rate (proportion of cases where the diagnosis was amended) was 4.2% and no serious errors were identified. Two thirds of errors were identified as a result of Multidisciplinary Team (MDT) review.
The error rate for consultants was steady throughout the year, and in total a similar proportion of amended reports was produced by consultants alone compared with consultants supervising trainees. Interestingly, more errors were noted in cases involving trainees early in the training year than later possibly relating to the impact of a recent intake of junior trainees.
This study shows a low error rate with no serious errors identified and reinforces the importance of MDT meetings in clinical risk management.

P158

Assessment Of Virtual Microscopy By Histopathology Specialist Registrars

{P} BM Da Gama-Rose[1], SA Hales[2], D Johnston[3]
[1]. Specialist Registrar Rotation In Histopathology, North West Deanery, Manchester, United Kingdom, [2]. Countess Of Chester Hospital, Chester, United Kingdom, [3]. SlidePath, Dublin, Ireland

It has been suggested that Specialist Registrars (SpRs) during their training should independently report certain specimens. Participation in a Non-gynaecological EQA scheme would provide evidence of quality assurance for them. In 2004 only 8 out of the possible 47 SpRs within our region took part on a formal basis. The problems encountered with glass slide circulation are legion but timely circulation is a major problem particularly for SpRs who are not formally included in the circulation list.
The 2002/3 circulation has been developed by SlidePath into a virtual microscope web based circulation and sent to all SpRs in our region. They have recorded their answers and been given immediate access to the consensus consultant opinion with illustrations of follow up histology. A questionnaire was completed to evaluate the scheme.
Their results have been analysed to provide a comparison with those of the consultant body, which type of slide/case did best and their conclusion on the medium for educational purposes.

P159

Daily multihead review of cellular pathology diagnosis in a district hospital

{P} H El-Daly, C Womack, L Astall, H Lawton, M Gertenbach, T Hayes, M Harris
Histopathology Department, Peterborough Gerneral Hospital, Peterborough, United Kingdom

We describe a review of records from daily meetings of the medical staff in a district hospital cellular pathology department during 2005.
Meetings take place around a multiheaded microscope (hydra) and typically last between 20-30 minutes. The primary purpose is diagnostic including double reading of all gastrointestinal dysplasias and new malignancies and discussion of difficult and interesting cases. Most cases referred away for second opinion are reviewed at a hydra session. Trainees are expected to attend. All cases are recorded including consensus, decision for referral, or levels/special stains.
During 2005 hydra sessions were held on 194 days with an average 4.6 persons attending.
2754 cases were discussed including 1511 (55%) cancers, 80 cases were sent for external opinion and 185 cases had further investigations.
More skin cases (549) were discussed than any other group and less than half of these were malignant (228 cases). In other groups the proportion of malignant cases varied between 70 % (colon) to 83% (prostate).
The daily hydra meeting allows for timely peer review of malignant, difficult and interesting cases, has potential as an audit tool, is a training opportunity, fosters team working and is often extended to include discussion of management and other issues.

P160

Urgent histopathology specimens – an audit of 100 consecutive cases

P Bhagwat, L Panchal, {P} A D Ramsay
University Collge London, London, United Kingdom

All histopathology departments receive cases marked as urgent, but there is little data on this type of specimen. An ideal urgent case is one in which the diagnosis is likely to immediately affect patient management, where the pathology findings can be rapidly conveyed to the clinician, and where adequate clinical information and the reason for the urgency are provided. Using this as a standard, an audit of 100 consecutive urgent cases was carried out in a teaching hospital department in 2005.
We found that urgent cases formed 2.7% of our workload, and originated mainly from the gastroenterology (28%), breast (11%), ENT (10%) and maxillo-facial (10%) units. The most frequent reason for urgency was a presumed diagnosis of malignancy (80%), and malignant disease was confirmed in 64% of this group. Other reasons included "for MDT review" (4%), "infection?" (3%) and "?diagnosis" (3%). A contact telephone or bleep number was supplied in 42% of urgent cases, but in 51% of the requests no contact details were given.
In conclusion;
Most urgent specimens are clinically justified
The majority of urgent biopsies contain cancer
Over half of urgent requests lack contact details – this should be a mandatory requirement for an urgent case.

P161

Is Histopathology still the Gold Standard in staging of rectal cancer? – An audit comparison of preoperative staging by histopathological staging and magnetic resonance imaging

{P} F Puls, S Ghosh-Ray, A Abraham
Northampton General Hospital, Northampton, United Kingdom

Background
Preoperative staging by magnetic resonance imaging (MRI) has been introduced recently to identify patients likely to benefit from radiotherapy alone or in combination with chemotherapy before surgical resection of rectal cancer. The aim of the audit was to compare the agreement of preoperative MRI and histological staging in routine practice.
Methods
Tumour (T) and nodal (N) stage of preoperative MRI reports were retrospectively compared with histological staging of resection specimens in fifty-two consecutive patients who underwent anterior resection for biopsy proven rectal cancer between 1/2004 and 2/2006. Patients receiving preoperative radiotherapy or radiochemotherapy were excluded from the study.
Results
There was fair agreement between MRI and histopathological assessment of T stage (61,54%, unweighted kappa = 0,35). Observed agreement of nodal status (N0, N1, N2) was fairly poor (53,85%, unweighted kappa = 0.25). T4 stage in five patients with peritoneal involvement was not detected preoperatively. Sensitivity of MRI for positive nodal status (N1, N2) was 55.2%, positive predictive value was 69.6%.
Conclusion
Histopathological staging remains the gold standard in staging of rectal carcinoma. In a routine clinical setting preoperative MRI staging of rectal carcinoma is fairly accurate, while its value in prediction of nodal status is limited.

P162

Measuring Workload in Histopathology

{P} LR Wheatley, S Carvell, TA Sims, AH McGregor
University Hospitals of Leicester NHS Trust, Leicester, United Kingdom

Background: The Royal College of Pathologists has provided guidance on measuring workload for histopathologists. However, this has not been validated as a way to measure the overall workload in histopathology departments. The accurate measurement of workload is additionally important, as most departments have reported an increase in workload in recent years, not in terms of requests, but rather in the complexity of investigation and interpretation. The aim of this study is to compare different methods to measure workload in histopathology.
Methods: The departmental workload was measured by requests, blocks, slides and tests; and by different modifications of the RCPath workload guidelines to reflect additional requirements of prioritised turnaround and MDT review.
Results: Workload increased from 1999-2005 by all methods, but the magnitude of increase varied enormously: 4.5% increase in requests, 35% increase in blocks, 45% increase in slides and increase from 42-50% using weighted scores.
Discussion: In the era of payment by results histopathology departments must be much more astute in measuring workload because resource should follow work done. Requests are not an accurate measure of work: block and slide numbers are more accurate but may not reflect the additional burden of work that requires specialist techniques, prioritisation and MDT review.

P163

Internet Virtual Pathology - an update

{P} F A Lewis[2], G A Casali[1], M Waterhouse[1], D Turner[1], P Quirke[1]
[1]. University of Leeds, Leeds, United Kingdom, [2]. Leeds Teaching Hospitals NHS Trust, Leeds, United Kingdom

In August 2005 the slide digital scanning facility was upgraded to include an additional Aperio T2 automated slide scanner and three Aperio T3 semi-automated slide scanners. In addition, the Image Storage Server has been upgraded to 18Tb. This has enabled a potential scanning output of 350 digitised slides per day. Since the upgrade, an additional 14,000 slides have been scanned resulting in over 27,000 digitised slides now being stored on the image server.
Development of the public internet site has continued with significant additions to the EQA, Graduate and Undergraduate Education and Clinical Trials areas of the site with the inclusion of EM images, MRI scans, gross pathology images and metadata.
Analysis of the activity on the site has shown that it is accessed from around the world with USA and UK being the most active users. November 2005 was an exceptional month with over 38,000 hits of the site recorded and since January 2006 there has been an average of 20,000 hits per month with daily peaks of activity from 11.00 to 15.00 and from 19.00 to 22.00. Popular areas of the site have included the educational areas but analysis of the most visited areas shows that the EQA slides were the most viewed with Renal EQA, GI EQA and Liver EQA, accessed preferentially in that order. It is clear that this resource is proving to be valuable for the assessment of EQA slides.

P164

Thyroid Pathology – Is There A Need For Specialist Reporting?

{P} P Joseph, K Skordilis
Manchester Royal Infirmary, Manchester, United Kingdom

Aims:
To assess the average reporting time for thyroid specimens and the causes for delay, including the number of cases requiring a second opinion.
Method:
All thyroid specimens received in the histopathology department of a university teaching hospital, in the year 2004 were included in the study. The turn around times were calculated in working days. The reporting times for the various consultants, the cases requiring a second opinion and the time required for the same was noted.
Results:
Total number of cases was 109. The average reporting time was 7 days. 31 cases (28%) took more than 7 days to report. In 14(45%) of these additional work (special stains, immunohistochemistry and further sampling) was the cause of delay. A total of 14 cases (13%) were sent for a second opinion. The time required for the second opinion, ranged from 17 to 71 days, with and average of 28.4 days. In 5 cases the specialist report confirmed the initial provisional diagnosis. In 9/14 (64%) where the initial provisional report was "inconclusive", 6 turned out to be malignant after second opinion.
Conclusion:
The need for a regional specialist in endocrine pathology is highlighted.

P165

Fine Needle Aspiration Cytology Of Follicular Variant Of Papillary Carcinoma Of Thyroid

{P} Nigam Shah, Marina Perera
Manchester Royal Infirmary, Manchester/Lancashire, United Kingdom

The cytological diagnosis of "follicular variant of papillary carcinoma of the thyroid gland (FVPCT)" by fine needle aspiration (FNA) causes diagnostic problems. Most cases are misdiagnosed as follicular lesions. We tried to evaluate the cytological findings of this lesion and then compare it with other studies. Four cases of histologically proven FVPCT were identified from the centre. Out of them only, one was originally diagnosed as FVPCT on FNAC.

All cases on review showed abundant cellularity with micro-follicles, syncytial groups and monolayered branching sheets. Papillary structures were noted in two cases and showed nuclear grooves and intranuclear cytoplasmic inclusions (INCI). Dense colloid was seen in all cases within follicles or as globules and masses of variable shapes adjacent to the cells. The other studies (7 articles were reviewed) showed similar features in follicular epithelial cells and dense globular colloid. They showed nuclear grooves, INCI and papillae were present in variable amount, while psammoma bodies and squamoid cells were rare.

Our study suggest that FVPCT should be suspected in any thyroid FNAC having high cellularity with syncytial clusters, monolayered branching sheets, micro-follicles and dense globular colloid. Nuclear grooves, INCI, papillae and squamoid cells may not be present but if present carries a high diagnostic value.

P166

Genome wide expression microarray analysis of papillary thyroid carcinoma and benign thyroid tissue: emphasis on the follicular variant and potential biomarkers of malignancy.

{P} P Smyth[1], S Finn[1], S Cahill[1], C Streck[4], E O'Regan[2], M Toner[2], R Henfrey[4], J Sherlock[4], D Howells[4], C Timon[3], M Cullen[3], J O'Leary[1], O Sheils[1]
[1]. Trinity College, Dublin, Ireland, [2]. Dublin Dental School and Hospital, Dublin, Ireland, [3]. St James's Hospital, Dublin, Ireland, [4]. Applied Biosystems, CA, United States

Papillary thyroid carcinoma (PTC), the most common endocrine malignancy, comprises a variety of morphological subtypes all characterised by a specific nuclear appearance. The most common sub-variant of PTC is the so-called follicular variant (FVPTC), which is a particularly problematic lesion and can be challenging from a diagnostic viewpoint even in resected lesions.
We used the recently developed Applied Biosystems 1700 microarray system to interrogate a series of 25 thyroid lesions. The cohort comprised 11 benign lesions and conditions and 14 samples of PTC (6 with classic morphology and 8 with FV morphology). TaqMan® RT-PCR was used to validate the expression portfolios of 50 selected transcripts.
Using ANOVA analysis 236 genes were identified that distinguish benign from malignant groups. Our data corroborates potential biomarkers previously identified in the literature such as LGALS3, S100A11, LYN, BAX, and CD44. However, we have also identified numerous transcripts never previously implicated in thyroid carcinogenesis. Diminished expression of metallothioneins featured strongly among these and suggests a possible role for this family as tumour suppressors in PTC. 15 transcripts were significantly associated with FVPTC morphology. Surprisingly, these genes were associated with an extremely narrow repertoire of functions, including the MHC and cathepsin families.

P167

MicroRNA Profiling of Papillary Thyroid carcinoma cell lines.

{P} S Cahill[1], P Smyth[1], K Denning[1], R Flavin[1], JH Lee[1], SP Finn[1], EM Oregan[1], R Henfrey[2], J Sherlock[2], JJ O'Leary[1], OM Sheils[1]

[1]. Trinity college Dublin, Dublin, Ireland, [2]. Applied Biosystems,, Foster City, CA, United States., United States

Activating mutations in BRAF and RET/PTC rearrangements are frequent genetic changes in Papillary thyroid carcinoma (PTC). MicroRNAs (miRNAs) are a group of non-coding single stranded RNAs measuring approximately 22 nt in length that have been found to control cell growth, differentiation and apoptosis. MiRNAs negatively regulate their target genes by translational repression. Components of the miRNA machinery have been implicated in tumourigenesis. Furthermore, miRNA expression profiling correlates with various cancers, with these genes thought to act as both tumour suppressors and oncogenes. This study was undertaken to identify a miRNA signature for PTC's harbouring Ret rearrangements and BRAF mutations and to elucidate the role of miRNAs in the development of PTC.
Design:
Differential expression of 157 human miRNAs was analysed from a panel of 5 PTC cell lines (harbouring Ret rearrangements, BRAF mutations) and a normal thyroid cell line.
Results:
A unique miRNA expression signature differentiated between PTC cell lines with RET rearrangements and BRAF mutations, and PTC cell lines from normal thyroid cell lines.
A distinct miRNA expression profile exists in PTC cell lines with RET rearrangements and BRAF mutations. Several of these up / down regulated miRNAs may be involved in PTC pathogenesis. A further challenge is to identify the targets of these miRNAs. MiRNA profiling will assist in the elucidation of disease pathogenesis and identification biomarkers and targets.

P168

Expression microarray analysis of thyroid carcinoma cell lines harboring BRAF V600E mutations

S Cahill[1], P Smyth[1], K. Denning [1], J H Lee[1] , S P Finn[1], E O'Regan[2], M. Gallagher[3] , M Toner[2], C Streck[4], R Henfrey[4], J Sherlock[4], J J O'Leary[1] and O M Sheils[1].

{P} S Cahill[1], K Denning[1], P Smyth[1], R Flavin[1], JH Lee[1], S Finn[1], EM O'Regan[1], R Henfrey[2], J Sherlock[2], JJ O'Leary[1], OM Sheils[1]

[1]. Trinity College dublin, Dublin, Ireland, [2]. Applied Biosystems, Foster City C.A, United States

Background: The use of cDNA microarrays is a powerful method for the quantitative analysis of altered gene expression associated with pathogenesis of disease. The complete repertoire of genes and signalling pathways involved in the pathogenesis of thyroid disease remains poorly defined. Another shortcoming remains the inadequacy biomarkers of disease sub-categorisation and progression. This study was undertaken as a step toward identifying previously uncharacterised molecular mechanisms in thyroid carcinoma and to further understand the role of BRAF V600E mutation.
Design: Gene expression profiles for a panel of thyroid cell lines were compared using a whole genome microarray system from Applied Biosystems. This panel included cell lines characteristic of various papillary and anaplastic thyroid carcinomas harbouring BRAF mutations and a normal thyroid cell line. In addition this normal cell line was used to generate a modified cell line expressing myc epitope tagged ^{V600E}BRAF.
Result: Genes found to be differentially expressed were those involved in cell structure, motility, and the extracellular matrix. Genes involved in the MAPK signalling pathway, oncogenesis, apoptosis and transcription regulation where shown to be upregulated. Several cell cycle regulators and apoptosis associated genes were down-regulated. This experiment was carried out in an attempt to uncover the key cellular pathways and genes that are significantly associated with the BRAF oncogene in the early stages of tumorogenesis. This knowledge will assist in the identification of diagnostic biomarkers and targets

P169

Expression of hTERT, P53, Bcl-2 and PCNA in non Hodgkin's lymphoma

{P} DM He, Y Zhang, GX Liu

Institute of Hematology, Jinan University Medical College, Guangzhou, China

Purpose: To investigate telomerase reverse transcriptase (hTERT), P53, Bcl-2 and PCNA expression in non Hodgkin's lymphoma(NHL) and their clinical significance. **Methods:** The expression of hTERT, P53, Bcl-2 and PCNA protein were evaluated by immunohistochemistry in 35 cases of NHL and in 15 cases of normal lymphom node. **Results:** The expression of hTERT, P53, Bcl-2 and PCNA protein were detected in 71%(25/35), 43%(15/35), 60%(21/35) and 86%(30/35), respectively. Whereas in 15 normallymphom node,hTERT, P53, Bcl-2 and PCNA protein expression was only in 27%(4/15) 0%(0/15) , 0% 0/15 and 32% 6/15 , respectively. hTERT, P53, Bcl-2 and PCNA protein expression in NHL were significantly higher than in normal control(P<0.05).In 35 NHL, 20 cases were both hTERT and Bcl-2 protein expression.Both the expression of hTERT and Bcl-2 protein in the low grade NHL were significantly higher than in the high grade NHL. The expression of hTERT protein is correlated with Bcl-2(P<0.05). In 35 NHL, 25 cases were both hTERT and PCNA protein expression. The expression of hTERT protein is correlated with PCNA(P<0.05). **Conclusions:** The expressions of hTERT,P53,Bcl-2 and PCNA protein were markedly higher in non Hodgkin's lymphoma than normal control. hTERT, P53, Bcl-2 and PCNA is associated with the malignant degree of NHL. hTERT is positively associated with Bcl-2,PCNA.

P170

Podocalyxin-like protein 1 vascular expression in normal and malignant colon

BD Tarlow, L Rangell, {P} HK Koeppen

Genentech, Inc, South San Francisco, CA, United States

We identified Podocalyxin-like protein 1 (PODXL) as a potential tumour vascular marker by using laser capture microdissection (LCM) followed by microarray expression profiling of microdissected tissue. PODXL is known to be expressed in podocytes and vascular endothelium in the human kidney. Recently, PODXL was reported to be a distinct marker of tumour endothelium in hepatocellular carcinoma, but not expressed in normal liver endothelium. The expression pattern of PODXL in normal and malignant colon tissues has not been previously reported.
In companion experiments, we determined PODXL protein expression in tumour vasculature relative to normal vasculature. PODXL expression was detected immunohistochemically with rabbit anti-human PODXL antiserum in normal and malignant colon. Co-localization with CD31 was evaluated in frozen tissues by immunofluorescence.
Result: PODXL was expressed in vascular endothelium throughout the normal human colon. All PODXL positive vessels co-expressed CD31, though a subset of larger CD31+ vessels was negative for PODXL. PODXL staining was observed in tumor-associated vascular channels at similar levels as in normal vasculature.
Conclusion: PODXL is expressed in tumor-associated vasculature and normal colon vasculature.

P171

The Role Of HER Gene Expression In The Response Of Primary Ovarian Cultures To Pertuzumab (rhuMAB2C4)

{P} M J Alemkunnapuzha, AQ Ganjifrockwala, M Adams, NT Oxley, GD Hall, PA Burns
LIMM, University of Leeds, St. James's University Hospital, Leeds, West Yorkshire, United Kingdom

Ovarian cancer is the leading cause of death among women with gynaecological disease in Western countries. Molecular alterations associated with some ovarian cancers include overexpression of EGFR (HER1), HER2 or HER activating ligands. Pertuzumab is a monoclonal antibody that binds to HER2 blocking its association with activated HER1, HER3 or HER4 thus inhibiting the growth of tumour cells that require HER-ligand dependent signaling.
We have previously assessed the efficacy of pertuzumab on primary ovarian cultures stimulated with heregulin (HRG; a HER3/4 activating ligand). Fifty five carcinoma cell lines were cultured from primary ovarian tumour tissue, malignant ovarian ascites or pleural effusion from ovarian cancer patients. Response to HRG (1nM) and pertuzumab (50μg/ml) was assessed using the WST1 cell proliferation assay. The cell lines fell into five distinct groups with respect to whether they responded to HRG and the degree to which they responded to pertuzumab. Overall, addition of HRG stimulated proliferation in 60% (n=33) of cell lines, whereas pertuzumab inhibited (either partially or totally) HRG-induced proliferation in 48% of these cell lines (29% of total).
The aim of this project was to examine the relationship between response of primary cell lines to HRG and pertuzumab, and expression of the HER gene family in the 55 primary cell lines. To achieve this we used a semi-quantitative real-time PCR approach to examine the levels of mRNA and also constructed a tissue microarray (TMA) of all the cell lines with their matching primary tumour samples to examine the levels of the four HER genes. This poster will describe the results of these experiments.

P172

Adult Renal Tumours With TFE-3 Expression

{P} K Kalyanasundaram, S Chaudhri, DMA Wallace, A Doherty, N Deshmukh
University Hospital Birmingham, Birmingham, West Midlands, United Kingdom

Renal tumours showing the specific translocation Xp11.2 have been described in children and young adults. Two adult patients aged 63 and 45 years presented with renal tumours at an advanced stage. The former patient was found to have a liver metastasis postoperatively. The other patient had an ovarian mass at the time of diagnosis, which was also excised. Histologically, the renal tumours and the ovarian mass were composed of nests of clear cells with focal papillary architecture. The cells showed well-defined cell borders, clear/pale abundant eosinophilic cytoplasm with vesicular nuclei and prominent nucleoli (Furhman grade 3). On immunostaining with TFE-3, both the renal and the ovarian tumours showed intense nuclear staining.
These two cases belong to a group of renal tumours having in common t (X;1) (p11.2;q21.2) translocation. Three different gene fusions have been observed with Xp11.2 involving the PRCC gene (on chromosome 1q21.2), ASPL gene (17q25) and PSF (1p34). All of them have similar histological appearances and express distinctive TFE-3 nuclear staining. This is a specific and sensitive marker for labelling the reported group of tumours. Though these tumours have been categorized as a separate group, their clinical behaviour in adults is yet to be determined.

P173

Reporting acute inflammation in prostate needle biopsies: A note of caution

{P} M A Rahman, M Varma
University Hospital of Wales, Cardiff/South Wales, United Kingdom

Background:
Serum PSA is often elevated in patients presenting with clinical features of prostatitis but prostate needle biopsies are contraindicated in such patients. The clinical significance of inflammation observed in prostate needle biopsies is controversial. It is recommended that acute inflammation in prostate needle biopsies should be reported as it could explain a raised serum PSA. However, significant inflammation is sometimes observed in background biopsies of patients with prostatic adenocarcinoma. In this study we have sought to document the incidence of inflammation in background prostate in patients with limited prostatic adenocarcinoma.
Study Design:
Twenty consecutive cases of limited prostatic adenocarcinoma (less than 1mm length of tumour in only 1 core) were identified in prostate needle biopsies performed on asymptomatic patients enrolled in a PSA screening study. All the cores (with and without cancer) were reviewed and degree of acute and chronic inflammation graded separately as 0:negative; 1:<10%; 2:11-25%; 3:26-50% and 4:>50%.
Results:
18 (80%) of the 20 cases showed inflammation: 7 (35%) chronic, 2 (10%) acute and 7 (35%) acute and chronic. The inflammation, both acute on chronic, was either grade 1 or 2. None of the cases showed grade 3 or 4 inflammation.
Conclusion:
Acute inflammation was observed in prostate needle biopsies from almost half of the asmptomatic patients with limited prostatic cancer. Hence, caution must be exercised before attributing raised serum PSA levels to acute inflammation observed in prostate needle biopsies.

P174

Subclinical Recurrence of IgA Nephropathy following Renal Transplantation: Evidence from Early Protocol Biopsies

{P} ISD Roberts, N Price, E Sarkar, C Winearls
Oxford Radcliffe Hospitals, Oxford, United Kingdom

IgA nephropathy (IgAN) is the commonest recurrent disease, diagnosed in clinically-indicated biopsies following renal transplantation and may result in graft failure. Here we determine frequency and significance of early subclinical (histological) recurrent IgAN by retrospective immunostaining of 1-month protocol biopsies, performed irrespective of graft function.

Search of our unit's nephrology (1987-95) and pathology (1995-05) databases revealed 53 patients transplanted for IgAN or diagnosed with recurrent IgAN. Of 35 transplants with an adequate biopsy at 1-month, 13 developed clinical recurrence. Two recurred early, within 2 years of transplantation. Both these patients showed glomerular IgA in biopsies at 1 month, but not at implantation and 1 week. Ten patients developed late clinical recurrence at a median time of 72 months (range 40-219), none of these showing IgA deposits in the 1-month biopsies or later clinically-indicated biopsies during the first 3 years post-transplantation. One patient has histological recurrence in the 3-year protocol biopsy. The remaining 22 patients have not developed disease recurrence, after being followed for a median time of 148 months; all their 1-month protocol biopsies were negative for IgA. In patients showing early subclinical recurrence, IgA positivity was initially focal, involving only a proportion of glomeruli, a stage never detected in native IgAN.

Most clinical recurrent IgAN is late; however IgA-positivity in protocol biopsies at 1-month post-transplantation predicts early clinical recurrence. The time between histological and clinical recurrence is short. Finally, at least in early protocol biopsies, there is no evidence of histological recurrence without subsequent clinical recurrence.

P175

LOW INCIDENCE OF POLYOMA VIRUS (BKV) NEPHROPATHY

{P} S Nayak, C Glennie, LJ McWilliam
Department of Histopathology,Manchester Royal Infirmary, Manchester, United Kingdom

Introduction: Polyoma virus is increasingly recognised as a cause of interstitial nephritis in renal transplants. The incidence ranges from 1-9% in different countries and it is an important cause of graft dysfunction and loss. The histological appearances can mimick those of acute rejection and cause difficulties in biopsy interpretation. We have knowingly seen three cases in the last three years one of which was subtle. All were associated with inflammation and tubulitis and were positive for BKV immunohistochemically. Our incidence seems low(<1%)and we needed to consider whether we were missing cases.
Aims: To determine the background incidence of BKV in renal transplant biopsies showing any tubulointerstitial inflammation. Initially, as a pilot study to stain biopsies from a six month period.
Methods: 100 transplant biopsies were performed in the first six months of 2005. Of these 29 showed tubulointerstitial inflammation and these were stained for BKV using a standard immunoperoxidase technique.
Results: All the test cases were negative for BKV.
Conclusions: Initial indications are of a low incidence as suspected, which may be partly due to immunosuppressive protocol differences from other centers. Staining a larger cohort of biopsies, targeting those between 2-18 months post transplant is to be undertaken.

P176

AUDIT OF ELECTRON MICROSCOPY SAMPLING IN RENAL BIOPSIES

{P} S Ibrahim, P Rowland, LJ McWilliam
Department of Histopathology,Manchester Royal Infirmary, Manchester, United Kingdom

Introduction: Recommended best practice for native renal biopsies is that electron microscopy(EM)should be performed in all cases, in addition to light microscopy and immunohistochemistry. Use of all these modalities in renal biopsy diagnosis is imperative to ensure consistency of diagnoses with superb quality control.
Aims: To determine whether all native renal biopsies had Electron Microscopic sampling in accordance with departmental standard operating protocols(SOPs).
Methods: A retrospective audit of native renal biopsies received over a period of six months in 2005. Light microscopic and EM reports,worksheets and original request cards were reviewed.
Results: Of 106 native biopsies received 93 had EM performed. Of the 13 cases where EM was not done, in 12 there was a perfectly valid explanation for this which was mainly inadequacy of the tissue received. One high risk case did not have EM taken. In 15 cases where tissue was sampled for EM no glomeruli were present. We looked at whether if more pieces were taken for EM this increased the glomerular yield, but this was not the case.
Conclusions: We are following our SOPs for EM sampling properly and EM was carried out in 88% of cases. The main reason for deviance was inadequate biopsies.

P177

Clear-Cell Papillary Renal Cell Carcinoma Of End Stage Kidney – A Report Of A Newly Described Entity

{P} s sundararajan, s chaudhri
university hospital birmingham, birmingham, United Kingdom

Introduction:
Recent literature describes two sub-types of renal cell carcinoma (RCC) that appear unique to end stage renal disease (ESRD). We report a case of one of these entities – clear-cell papillary renal cell carcinoma of end stage kidney (CPRCC).
Case report:
A 54 year old female with known diabetic nephropathy in ESRD underwent a nephrectomy following detection of a mass in the right kidney. Gross examination, showed a solid-cystic tumour 20mm maximum dimension. Histologically, it had a predominantly papillary architecture and was composed of cuboidal cells with clear cytoplasm and low-grade nuclei. In places, there was polarisation of nuclei in a linear array away from the basement membrane. On immunostaining it was positive for CK7 but negative for AMCAR.
Discussion:
There is an increased incidence of RCC in patients with ESRD. Historically, the majority of renal cell neoplasms occurring in ESRD have been reported to be papillary RCC. However, recently two new entities have been described that together constitute the commonest tumour types in this setting - acquired cystic disease associated RCC and CPRCC. Apart from distinctive morphology, these show an immunoprofile that differs from both conventional clear-cell and papillary RCCs and a biologic behaviour less aggressive than the RCCs in sporadic or non-ESRD setting.

P178

Epithelioid angiomyolipoma in a renal graft. To transplant or not to transplant?

{P} GM Mitsopoulos[1], KS Skordilis[2]
[1]. Royal Sussex county hospital, Brighton, United Kingdom, [2]. Manchester Royal Infirmary, Manchester, United Kingdom

Epithelioid angiomyolipoma of the kidney is an exceedingly rare variant of angiomyolipoma, which, unlike the more typical variant may behave more aggressively.
We present a case of epithelioid angiomyolipoma, review the literature and suggest clinical management.
A 6 mm cortical nodule was identified on a renal graft at the time of transplantation. The 65 year old, non-heart beating, donor had no known history of tuberous sclerosis or any other lesions in any of his organs. A wedge excision of the tumour was submitted for histological examination. This revealed a well-circumscribed tumour composed of mature adipose cells, large vessels, hyalinised stroma and irregular trabeculae of epithelioid cells. The cells expressed HMB-45, Melan-A, Smooth Muscle Actin and focally Vimentin. Other markers such as CD10, EMA, Synaptophysin and Inhibin were negative. The diagnosis was that of an epithelioid angiomyolipoma.
The clinical advice to the transplant teams, the renal recipient and the other recipients concerned was that this is a tumour with a risk of metastasis but probably a relatively low risk. The patient proceeded with the transplant and had a good recovery. In view of the possible risk of metastasis or recurrence it was suggested that the patient should be followed –up by CT scans at 3, 6 and 12 months post-operatively.
Angiomyolipomas are mixed mesenchymal tumours belonging to the group of PEComas, tumours derived from perivascular epithelioid cells. The epithelioid variant is rare and may mimic malignant tumours such as renal cell carcinoma. Ongoing research into the molecular biology and clinical behaviour of this tumour may improve our ability to manage these lesions.

P179

The Effects of Intermediate Solvents and Resins on Glomerular Basement Membrane (GBM) Thickness

{P} K Edwards[1], C. J. von Ruhland[2], M. Morgan[3], D. F. R. Griffiths[3]
[1]. University of Wales Institute Cardiff, Cardiff, United Kingdom, [2]. Cardiff University, Cardiff, United Kingdom, [3]. University Hospital of Wales, Cardiff, United Kingdom

Establishing GBM thickness is important in the diagnosis of some renal diseases. It is often stated that thickness varies according to the processing method, making comparisons between departments difficult. There is no published information to support this statement. We aimed to determine if processing influenced GBM thickness.
Superficial cortex from glutaraldehyde perfusion fixed rat kidney was post-fixed in 2% OsO_4, dehydrated through graded ethanol, followed by intermediate solvent (propylene oxide (PO), xylene (Xyl), anhydrous acetone (Ac) or anhydrous ethanol (Et)), and infiltrated and embedded in epoxy resin (Araldite (Aral), TAAB embedding resin (TER) or TAAB low viscosity resin (TLV)). Thin sections were stained with uranyl acetate and lead citrate and examined in a Philips CM12 TEM at 80kV.
GBM thickness was measured by the orthogonal intercept method.
Compared with PO – Aral embedding, GBM was significantly thicker in Xyl-Aral, Ac-Aral, Xyl-TER, and significantly thinner in PO-TER, Ac-TER and Et-TER embedded tissue($p<0.05$ in all cases). No significant difference was seen with Xyl-TLV and Ac-TLV embedded tissue. Et-Aral, PO-TLV and Et-TLV embedding resulted in poor quality blocks.
The results suggest that the choice of both intermediate solvent and resin may influence shrinkage of the GBM during TEM processing.

P180

Benign Ovarian-Type Epithelial Tumours Of Testis And Paratestis

{P} S Malhotra, S Patil, L Sheehan, A Coup
Dept.of Histopathology,PathLinks,County Hospital, Lincoln, United Kingdom

Ovarian-type epithelial tumours (OTET) are believed to arise from Mullerian type epithelium entrapped within the testis. These rare tumours are identical to their ovarian counterparts. Most reported cases are serous or mucinous tumours of borderline malignancy with fewer serous carcinomas and single cases of endometrioid and clear cell adenocarcinomas.
We report two cases of benign OTETs arising in the testicular region.
A 49yr old man presented with left scrotal swelling. A large cystic lesion attached to the left epididymis and adherent to the testis was excised. Histology showed a cyst with a thin fibrous wall, lined by cuboidal ciliated epithelium. There were areas of cyst wall with architecture identical to an ovarian serous cystadenofibroma.
The second case was a 69yr old man undergoing repair of a right inguinal hernia. A cystic mass was found, apparently arising from the testis. Microscopically, this was lined by mucinous epithelium.
In both cases, immunohistochemistry was positive for BerEP4, CK7, CA125, ER and PR.
In conclusion, OTETs of the testis and paratestis are histologically and immunophenotypically identical to their ovarian counterparts, supporting a Mullerian origin.
We raise the possibility that benign OTETs, particularly of serous type are being overlooked and are reported as epididymal cysts.

P181

Prostatic Invasion By TCC In Cystoprostatectomies For Primary Bladder Cancer And Incidental Prostatic Adenocarcinoma.

{P} H M Abdelsalam, M Wallace, A Doherty, S Chaudhri, N Deshmukh
University Hospital Birmingham, Birmingham, United Kingdom

Pathological staging of primary bladder transitional carcinoma requires careful histological examination of the prostate for invasive or in situ TCC as well as for the presence or absence of primary prostatic adenocarcinoma. We reviewed the histological findings in 94 male patients who underwent cystoprostatectomy between January 1996 and March 2005 for primary bladder transitional cell carcinoma.
43 (45.7%) out of 94 patients showed either involvement of the prostate by TCC or prostatic adenocarcinoma. 16 (17%) patients had involvement of the prostate by transitional cell carcinoma; 8 of which were carcinoma in situ only involving the prostatic ducts or urethra. The other 8 cases with prostatic stromal invasion had direct invasion of TCC through the bladder wall in 7 patients. Only one patient had indirect stromal invasion from a prostatic duct.
Incidental prostatic adenocarcinoma was diagnosed in 33 (36%) out of 92 patients (2 patients had prostatic adenocarcinoma diagnosed before surgery). Clinically significant prostate cancer was diagnosed in 12 out of 33, with 7 cases having more than one criterion for clinical significance.
In conclusion, this study emphasizes that prostatic neoplasia is a common finding in cystoprostatectomy specimens for primary bladder TCC and highlight the importance of complete prostatic removal.

P182

Microstaging of Prostate Cancer: Biopsy Core Length of Tumour is the Only Variable That Can Identify Clinically Insignificant Tumours

R H Hurle[1], A Hawizy[2], P Linardatou-Novak[1], H G Kynaston[2], D Cochlin[2], {P} D F R Griffiths[1]
[1]. University of Cardiff, Cardiff, United Kingdom, [2]. Cardiff and Vale NHS Trust, Cardiff, United Kingdom

There are currently no agreed criteria for preoperative identification of small prostate cancers (tumour volume ≤ 0.5ml) and many are over treated. In theory accurate sampling of the gland by a biopsy protocol should allow assessment of tumour volume.
Analysis of the diagnostic biopsy in 223 consecutive prostate cancer patients having radical prostatectomy (RP) was carried out to determine how well biopsy findings can predict tumour size. All patients had previously had protocol biopsies performed by a radiologist with a special interest in the biopsy technique. Biopsy specimens had been processed for histology in separate cassettes; on average 15mm of core was examined from each sample. Number of positive cores, maximum length of adenocarcinoma in any core (MLA), presentation PSA; clinical and MRI stage and RP tumour volume was recorded.
45 cases had only a single core positive and an MLA of $\leq$4mm, of these 23 had clinically insignificant tumours giving this criterion a positive predictive value of 51% and a negative predictive value of 93%. In multivariate binary logistic regression with PSA, MRI stage and clinical stage this biopsy finding was the only variable to predict a clinically insignificant tumour (relative risk: 12 {95% CIs 5 – 29}
With consistent biopsy technique and meticulous histological processing biopsy findings have the potential to predict tumour size.

P183

How Should the Length of Prostate Cancer In Needle Cores Be Measured?

{P} S M Howarth[2], D F R Griffiths[1]
[1]. Cardiff University, Cardiff, United Kingdom, [2]. Cardiff and Vale NHS Trust, Cardiff, United Kingdom

The length of tumour in a needle biopsy core correlates with the tumour volume in subsequent radical prostatectomy (RP) specimens. However there is no agreed way to measure tumour length in a core. There are good theoretical reasons for using the dimension between the outer margins of tumour in the core involved – the maximum length (ML); although some pathologists are reluctant to report two small separate foci as a single long length and prefer to sum the lengths of separate foci- the sum lengths (SL).

The aim of the study is to determine the measurement which best correlates with tumour size and hence gives the most appropriate information to clinicians. 35 of a series of 272 RPs had a single core positive in prior protocol biopsies. The length of tumour was measured in the positive core by both ML and SL methods and the strength of correlation with RP tumour volume tested by a Spearman's rank test.

There was a difference between the two measurements in 10 cases (mean difference of 2.2mm, range 0.4 to 5.1). Measurements by both methods correlated strongly with the tumour volume; the correlation with ML was substantially more significant than the SL, P=0.004 vrs P= 0.02 respectively.

ML is simpler and correlates better with tumour volume; it is recommended as the method to report tumour length.

P184

Wilms' tumour may develop from impaired differentiation of the ureteric bud as well

{P} Z Stupar, G Kovacs
Laboratory of Molecular Oncology, Medical Faculty, University of Heidelberg, Heidelberg, Germany

It is generally acknowledged that Wilms' tumour arises from pluripotential cells of the metanephric blastema. During the analysis of SLIT2 and its receptors ROBO1 and ROBO2 in conventional renal cell carcinomas and also other types of kidney tumours by real time RT-PCR we have detected an increased expression of these genes in embryonal/fetal kidneys and Wilms' tumours. By immunohistochemical analysis we were able to localize the ROBO1 and SLIT2 proteins to the ureteric bud and developing collecting duct system in embryonal/fetal kidneys. A positive staining for ROBO1 and SLIT2 proteins was detected in 16 of the 27 Wilms' tumours of mixed type. Circumscribed proliferation of embryonal tubular structures which were surrounded by blastemal or stromally differentiated cells resembling the condensation of blastemal cells around the tip of ureteric bud in normal developing kidneys were stained positively with both antibodies. No expression was seen in the 7 monophasic blastemal and 3 purely epithelial Wilms' tumours. Our data suggest that the impared differentiation not only of the blastemal mesenchyme but also of the ureteric bud may be involved in the development of Wilms' tumours.

P185

Expression Signature of End Stage Kidney

A Nagy, {P} G Kovacs
Laboratory of Molecular Oncology, Medical Faculty, University of Heidelberg, Heidelberg, Germany

End stage renal disease (ESRD) is associated with hyperplastic and cystic remodelling of the kidneys (ARCD) and increased rate of kidney tumours, especially of papillary renal cell cancer (RCC). Using the Affymetrix HG U133 oligoarray, we have established the gene expression signature of ESRD/ARCD kidneys and compared to those of fetal and adult kidneys and of distinct types of childhood and adult renal cell tumours. The correspondence analysis revealed 5 sample clusters improving that ESRD/ARCD has a unique transcription profile, well separated from the other clusters of normal and tumorous samples. Under stringent conditions the overexpression of 69 genes was selected, 47 of them were confirmed by quantitative RT-PCR. Eighteen genes were also expressed in papillary RCCs and further 8 in other types of tumours. Immunhistological analysis localized the expression of selected genes GPR87, CSF2, SCEL, KRT19 to hyperplastic tubular structures, cysts and small epithelial cell clusters in ESRD/ARCD kidneys. The SCEL and KRT19 were also expressed in small papillary precursor lesions and papillary RCC suggesting their role in the tumorigenesis. The unique expression signature, e.g. the specific molecular environment may explain the high frequency of papillary RCCs in ESRD/ARCD kidneys.

P186

Sertoliform Cystadenoma of the Rete Testis. Case Report of a Rare Benign Tumour of the Testis

AM Sinclair, T Gunendran, RD Napier-Hemy, {P} H Denley
Manchester Royal Infirmary, Manchester, United Kingdom

Testicular cancer is the most common solid malignancy occurring in young men between the ages of 15 and 35 years, with approximately 2000 cases per year in the UK. If a young man presents with a testicular lesion then the most likely diagnosis is therefore a malignant tumour and radical orchidectomy is the recommended treatment. A Sertoliform cystadenoma of the rete testis is a very rare testicular tumour, this poster presenting only the fifth ever case. Although rare the diagnosis is essential to make, as, unlike the majority of testicular tumours, a Sertoliform cystadenoma of the rete testis is benign.

The diagnosis is difficult to make pre-operatively and its position at the rete testis does not permit a partial orchidectomy. However, post-operatively its benign nature has major implications in that, if this diagnosis can be confidently made, the patient does not need adjuvant treatment or long term follow up.

The poster will show the ultrasound appearances of a mass at the rete testis and pathological slides identifying features of the tumour that enabled us to consider this rare diagnosis. The poster will then demonstrate the immunohistochemical features that enabled the diagnosis to be confidently made.

P187

Malignancies after renal transplantation

{P} E A Husain, I Junaid, S I Baithun
Barts and The London NHS Trust, London, United Kingdom

We examined rates of malignancies among 681 recipients of renal transplants at Bart's and the London hospitals performed between 1984 and 2005. Malignancies developed in 58 patients (8.5%), which is significantly higher than that of the general population. Many patients tended to have more than one tumour. The most prevalent neoplasms were non-melanoma skin cancers, which tended to be multiple and recurrent. The next in frequency was transitional cell carcinoma of the bladder and renal pelvis and it was characterised by higher grade and stage at presentation. Cancer rates for each individual malignancy was higher in renal transplant recipients compared to the general population with a relative incidence ranging from 1.2 in lung cancer to 1835 in Kaposi's sarcoma. This high frequency of various malignant tumours makes cancer prevention a major target following transplantation. Modulation of immunosuppressive therapy and avoiding exposure to various oncogenic factors are the most important preventive measures. The aggressive nature and bad prognosis of malignancies in these patients necessitates close follow up for their early detection.

Malignancy	No	Malignancy	No
Non-melanoma skin cancer	32	Cancer lip and oral cavity	3
Transitional cell carcinoma	6	Renal cell carcinoma	1
Breast carcinoma	5	Oesophageal carcinoma	1
PTLD	5	Lung carcinoma	1
Kaposi's sarcoma	5	Cervical cancer	1
Malignant lymphoma	5	Merkel cell carcinoma	1
Prostatic carcinoma	3	Leiomyosarcoma	2
Testicular cancer	2	Dermatofibrosarcoma	1

P188

Audit Of Gleason Grading Of Prostatic Carcinoma In Needle Core Biopsies

{P} K Sawalem, S Verma
Trafford General Hospital, Manchester, United Kingdom

The Gleason grading system is the most widely used grading system for prostate carcinoma. Histopathological grade is a predictor of biological behaviour of cancer and influences patient management. There is inter-observer variation in Gleason grading of prostatic carcinoma. Reproducibility has been shown to improve with teaching methods.
An audit was undertaken to measure the inter-observer variation in Gleason grading of prostatic carcinoma in needle core biopsies, amongst general histopathologists of varying experiences, in a DGH. 22 consecutive and retrospective cases of prostatic carcinoma diagnosed on needle core biopsies were collected. 10 pathologists reviewed each case and performed Gleason grading and scoring. The data was analysed to measure the inter-observer variation in the scores. The simple kappa value was found to be 0.18 ($p<0.0005$), indicating poor agreement.
Educational measures were recommended to improve reproducibility of Gleason grading in needle core biopsies. These included
Clear knowledge of definitions of different Gleason patterns of carcinoma by all pathologists involved in reporting prostate needle core biopsies.
Use of guidelines in reporting prostate cancer e.g. RCPath minimum dataset on prostate cancer.
Use of reference images for Gleason grading.
Web-based tutorial for Gleason grading.
Re-audit after measures have been implemented.

P189

Prostatic Needle Core Biopsy for diagnosis of adenocarcinoma: Are Three Initial Levels Essential? –An Audit

{P} Stephen McGrath, Sudha Desai
Hope Hospital, Manchester, United Kingdom

Needle core biopsy of the prostate gland is commonly employed when there is clinical or biochemical suspicion of adenocarcinoma. Few articles have discussed what is the most suitable method of processing these specimens. In some instances our own "3 initial levels" protocol appeared to consume a large proportion of the biopsy tissue and subsequent requests for immunohistochemistry staining in equivocal cases were rendered non-informative. Accordingly, we decided to audit (a) the workload generated by biopsies diagnosed as containing carcinoma, (b) the variation in Gleason score between sequential levels and (c) embedding techniques. Positive biopsies from 37 patients (92.5% of all unequivocally malignant specimens received over a six month period) were audited. Over 90% of the cases required no further levels and no immunohistochemistry for diagnosis. All cases contained carcinoma on the first level and in most cases the Gleason score remained unchanged between levels. The results suggest that the vast majority of positive biopsies will include representative malignant foci in the first level and that only negative or equivocal cases need multiple levels with/without immunohistochemistry (in order to exclude adenocarcinoma). Processing each biopsy initially at only one level may preserve important diagnostic material and facilitate the examination of both extra H&E levels and immunohistochemistry studies.

P190

Evaluation of Minichromosomal Maintenance Proteins (MCM2, 5 & 7) as Biomarkers of Proliferation in Prostatic Hyperplasia and Adenocarcinoma

J Watkins[1], TA Bailey[1], AC Woodman[1], {P} AJ Molyneux[2]
[1]. Cranfield BioMedical Centre, Cranfield University, Silsoe, United Kingdom, [2]. Department of Cellular Pathology, Northampton General Hospital, Northampton, United Kingdom

Minichromosome maintainence proteins (MCMs) are essential regulatory proteins in DNA replication and are down regulated in quiescence and cell differentiation. Their overexpression may be a useful marker for cell cycle deregulation. The expression of three MCM proteins (MCM2, 5 and 7) was assessed immunohistochemically on prostate tissue sections from 74 patients, comprising prostatic hyperplasia, BPH (32 cases) and prostatic adenocarcinoma (42 cases, Gleason scores ranging from 4 to 10). Nuclear staining profiles were recorded qualitatively and semi-quantitatively using a standardised scoring index. Benign and malignant MCM staining profiles were compared. MCM staining scores were also compared with adenocarcinoma Gleason scores. MCM staining in BPH was almost entirely confined to the basal cell compartment and was very low or absent in luminal epithelial cells (<4%). In contrast, all MCM markers showed a significant increase in luminal epithelial staining in the adenocarcinoma group ($p=0.0001$). MCM5 and MCM7 also showed a progressive increase in staining index with increasing Gleason score, with greater sensitivity than MCM2 for Gleason score 5-8 carcinomas.
This study confirms the value of MCM proteins as biomarkers of abnormal proliferation on prostate tissue sections. The results also suggest a relationship between Gleason score and progressive cell cycle deregulation.

P191

Metastatic Carcinoma Of Prostate Masquerading As A Primary Caecal Tumour

{P} M Kabeer, S Kulkarni, G Maskell, R Hohle, J Mathew
[1]. Department of Surgery, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [2]. Department of Clinical Imaging, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [3]. Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom

Aim: To describe a case of metastatic prostatic carcinoma masquerading as caecal cancer.
Case Report: A 72 year-old man presented with a clinically palpable caecal mass and a firm nodule on his thigh. CT scan showed a caecal mass with nodal involvement as well as bony metastasis. He was being treated (Zoladex) for prostatic cancer diagnosed 6 years previously. On admission his PSA was 224µg/L but despite this the MDT consensus was, that whilst not excluding a metastatic lesion, the clinical and imaging appearances were strongly suggestive of a primary caecal tumour with probable regional metastasis.
The patient subsequently underwent a right hemi-colectomy. The histology of the specimen showed a poorly differentiated adenocarcinoma.
Immunohistochemistry showed that the tumour was PSA positive. The patient is currently undergoing adjuvant therapy and the nodule on his thigh is under investigation.
Conclusion: Although up to 20% of patients have metastasis on presentation, metastasis of prostatic carcinoma to the bowel is very rare: so far only three cases have been reported in literature. Palliative treatment remains the mainstay of therapy for metastatic prostate cancer and hormonal therapy represents the standard. Surgery, radiotherapy, chemotherapy and secondary hormone therapy are the options for hormone refractory cases.
Metastatic prostatic carcinoma to the bowel is a rare occurrence and presents a challenging diagnosis.

P192

Reproducibility Of Activity And Chronicity Index Scoring In Lupus Nephritis And Association With Clinical Outcomes

{P} M Patel, L Crompton, L McWilliam, H Denley
Manchester Royal Infirmary, Manchester, United Kingdom

Lupus nephritis (LN) is a major complication of SLE. Previous studies evaluated the prognostic significance of active or chronic lesions on renal biopsy. However, these are subject to inter- and intra-observer differences, thereby limiting reproducibility. This study examined whether activity (AI) and chronicity indices (CI) were reproducible and predictive of outcome in LN.

55 LN patients were studied. Each biopsy was reviewed by two histopathologists independently, blinded to clinical history and original biopsy report. The AI and CI were assessed using published methods (Austin et al, 1984). The primary renal outcome measure was chronic renal failure (GFR <50% predicted/SLICC Damage Index 2) or development of ESRD (SDI 3). Scores rarely differed by >1 point and discrepancies were resolved by employing the higher grade.

There was a significant correlation between CI and serum creatinine, GFR, 24hr protein excretion and serum albumin *($P<0.0001$,* Spearman's). The CI was not associated with overall SDI but was with renal SDI scores. The probability of developing CRF was significantly higher in patients with higher CI *($P<0.0001$,* Kaplan-Meir).

Therefore AI and CI scoring is reproducible and useful in LN. High CI correlates with increasing creatinine and proteinuria and is strongly predictive of poor renal outcome.

P193

Quantification and Phenotype of Factor XIIIa Positive Cells in the Renal Interstitium and Proposals for their Role in Tubulointerstitial Fibrosis

{P} V Elliot[1], R King[1], SK Campbell[2], JE Collins[2], PS Bass[1]
[1]. Southampton University Trust Hospital, Southampton, HANTS, United Kingdom, [2]. University of Southampton, Southampton, HANTS, United Kingdom

Introduction
Fibrosis is a common final pathway leading to end stage renal failure. Cells expressing the transglutaminase Factor XIIIa (FXIIIa) have been identified in fibrotic tissue. FXIIIa cross links proteins such as fibronectin and collagen and cells expressing FXIIIa may play a role in inflammation and repair.
Objectives
To assess the prevalence of FXIIIa positive cells in renal disease
To determine the phenotype of cells expressing FXIIIa
Methodology
Renal cell expression of FXIIIa and their phenotype was examined in paraffin embedded renal biopsies from cases of membranous glomerulonephritis, minimal change disease and IgA nephropathy. Normal renal tissue from nephrectomy specimens acted as control (n=20/group). Antibody expression was visualised using the Vectastain ABC kit. Cellular distribution of staining was noted and positive cells were quantified. Serial sections were mapped to assess co-localisation.
Results
FXIIIa positive cells were identified in the interstitium in normal and diseased kidneys. There was significant up-regulation in all diseased groups compared with normal. Mapping of serial sections indicated that various cell types may express FXIIIa.
Conclusions
Multiple cell types may express FXIIIa which may indicate a state of cellular activation. FXIIIa may also play an important role in tubulointerstitial fibrosis.

P194

Fine Needle Aspirates Of The Thyroid: Ultrasound Versus Non-Ultrasound Guided

{P} P Joseph, N Singh, R Howitt
Southampton University Hospitals, Southampton, United Kingdom

Aims:
This study has been undertaken to assess the adequacy of thyroid fine needle aspirates and to compare ultrasound guided (USG) versus non-ultrasound guided aspirates (Non-USG).
Method:
All thyroid aspirates received in the cytopathology department of a university teaching hospital, over a four-year period from 2001 were included in the study. All identified cases were evaluated for the method of aspiration (USG / Non-USG) and adequacy of the specimen. The criterion for adequacy used was a minimum of 6 groups, each with 10 or more cells.
Results:
A total of 334 specimens were received in the department. Of these 209 were Non-USG specimens and had 65 inadequate specimens (inadequacy rate: 31.10%) while the remaining 125 specimens were USG specimens and had 19 inadequate specimens (inadequacy rate 15.20%).
Conclusion:
There is significant difference in the inadequacy rate between USG and Non-USG thyroid aspirates.
Recommendations:
To reduce the number of inadequate specimens, this information should be brought to the notice of the clinicians and the use of USS guided aspirates should be advocated.

P195

Genetic And Epigenetic Alteration Profiles For p16 In Head And Neck Squamous Cell Carcinoma In Young Adults

{P} EM O'Regan[2], CY Fan[4], ME Toner[2], M Ring[3], SP Finn[1], C Timon[1], P Smyth[1], S Cahill[1], O Sheils[1], JJ O'Leary[1]
[1]. Department of Histopathology Research, University of Dublin, Trinity College, Dublin, Ireland, [2]. Department of Oral Pathology, Dublin Dental School and Hospital, Dublin, Ireland, [3]. Dept of Histopathology, Coombe Womens Hospital, Dublin, Ireland, [4]. Dept of Pathology and Otolaryngology, Arkansas Cancer Research Centre, Little Rock, Arkansas, 72205, United States

Background: Recently, there has been a worldwide increase in incidence of HNSCC in young adults (under 40 years old), many of them never smokers and females. Functional inactivation of p16 is a common event in classic HNSCC. The purpose of this study is to evaluate inactivation and expression patterns of p16/INK4A in HNSCC with a particular focus on the not so typical HNSCC patient (the young adult nonsmokers).
Design: 25 samples of HNSCC (10 < 40 yrs old, 15 > 40 yrs old) were collected. CGH microarray, methylation-specific PCR and immunohistochemistry were used to evaluate the genetic and epigenetic profile of p16 (CDKN2A).
Result: 48% of the samples showed p16 inactivation (either by deletion or methylation). Deletion of p16 was only detected in the older cohort (46%), while methylation of p16 was detected in 30% of the young adults and 13% of the older group. Strong p16 staining was evident in equal numbers in the young cohort and in the older cohort (20%).
Conclusion: Our results indicate that inactivation of the p16 gene is a frequent event in HNSCC. While deletion is the main mechanism of inactivation in male smokers, methylation is a more common event in female nonsmokers. This suggests that specific modes of inactivation of p16 in HNSCC are related to specific patient profiles. This most likely reflects different aetiologies or exposure patterns, which need to be further explored.

P196

Caspase-3 Expression In Head And Neck Squamous Cell Carcinoma

M. Hafidh, A.S. Jones, {P} T.R. Helliwell
University of Liverpool, Liverpool, United Kingdom

This study has evaluated the pattern of expression of cleaved caspase-3, a key protease involved in the initiation of apoptosis in head and neck squamous carcinoma, and examined the association with other apoptosis-related markers and prognosis. A tissue microarray was created from archived wax blocks of 200 primary squamous carcinomas (83 larynx, 72 pharynx and 45 oral cavity). 75 cases also had tissue from nodal metastases. Immunocytochemical labelling was performed for caspase-3, p53, mdm2, survivin, bcl-2 and bax proteins.

Caspase-3 labelling occurs in both the nucleus and cytoplasm of a small proportion of malignant cells, and in apoptotic nuclear debris. Nuclear labelling is present in a mean of 3.79% cells in laryngeal carcinomas, 2.53% cells in pharyngeal carcinomas and 2.09% cells in oral carcinomas. Expression is similar in primary carcinomas and their nodal metastases. The % cells showing nuclear caspase-3 expression correlates with poor differentiation and nodal metastasis. Nuclear caspase-3 expression is positively correlated with the expression of mdm-2, bax and survivin, and negatively correlated with involucrin expression. Nuclear caspase-3 expression may be a useful measure of apoptotic activity in head and neck squamous carcinoma.

P197

Suppression Of Cutaneous Fatty Acid Binding Protein Reduces Proliferation in Squamous Carcinoma Cells *In Vitro*

W. Gomaa[1], Y. Ke[1], I.C. Paterson[2], {P} T.R. Helliwell[1]
[1]. University of Liverpool, Liverpool, United Kingdom, [2]. University of Bristol, Bristol, United Kingdom

Human cutaneous fatty acid binding protein (C-FABP) is involved in the differentiation of normal keratinocytes and is over-expressed in a subset of human head and neck squamous cell carcinomas and their nodal metastases. This study has examined the effect of suppression of C-FABP expression on proliferation of human oral squamous carcinomas cell lines. Oral squamous carcinoma lines were tested for their C-FABP expression by western blotting. The RNA interference technique was used to suppress C-FABP expression in the cell line with the highest C-FABP expression. The clone with the highest level of protein suppression was compared with parental cells and cells only transfected with vector in an in vitro proliferation assay.
RNAi resulted in a clone of transfectant cells with 65% reduction of C-FABP protein expression. C-FABP suppression was associated with a 2.5 fold reduction in the rate of proliferation of the transfectant clone compared with parental and vector controls. These in vitro observations confirm earlier suggestions that C-FABP may have a significant role in the differentiation and proliferation of squamous carcinoma cells in vivo.

P198

Follicular Dendritic Cell Sarcoma: A Rare Tumour Presenting with an Abdominal Mass

SE Low[1], {P} CM Manson[1], L Menasce[2]
[1]. Dept of Histopathology, Warrington Hospital, Warrington, United Kingdom, [2]. Dept of Histopathology, Christie Hospital, Manchester, United Kingdom

Follicular dendritic cell (FDC) sarcoma is a rare neoplasm of follicular dendritic cells, with the majority having a lymph node origin. Only a third located extranodally. We report a case of FDC sarcoma arising within the abdominal soft tissues.
Occasional cases are associated with the hyaline vascular variant of Castleman disease arising from dysplastic follicular dendritic cells. Those that resemble inflammatory pseudotumour are associated with the Epstein Barr virus and differ from conventional FDC sarcomas.
Dendritic markers (CD21, CD23 and CD35) are positive. The tumour cells are also positive for vimentin and EMA, with variable positivity for S100 and CD68. Staining for CD1a, lysozyme, CD34, CD3, CD79a, CD30, HMB45 and cytokeratin is negative. The small lymphocytes can be either T or B cells.
As FDC markers are often not routinely included in antibody panels, awareness is important, as it can be confused with other tumours especially when occurring extranodally. A high index of suspicion of FDC sarcoma is required when faced with cytokeratin negative pleomorphic tumours, thymomatous and meningiomatous tumours located outside the mediastinum and dura respectively, and other unusual looking tumours. In the presence of scattered intratumoral small lymphocytes, the diagnosis of FDC sarcoma should always be considered.

P199

Follicular lymphoma with both t(14;18) and Burkitt translocations at presentation: an adverse prognostic parameter identified by karyotypic analysis?

{P} LJ Christie[1], NM Kernohan[1], DA Levison[1], M Sales[2], P Batstone[3], D Meiklejohn[4], JR Goodlad[5]
[1]. Department of Pathology and Neuroscience, University of Dundee, Dundee, United Kingdom, [2]. Department of Cytogenetics, Ninewells Hospital and Medical School, Dundee, United Kingdom, [3]. Department of Cytogenetics, Raigmore Hospital, Inverness, United Kingdom, [4]. Department of Haematology, Ninewells Hospital and Medical School, Dundee, United Kingdom, [5]. Department of Pathology, Western General Hospital, Edinburgh, United Kingdom

Follicular lymphoma (FL) is a common form of low grade B-cell non-Hodgkin's lymphoma (NHL). Although this form of lymphoma often pursues an indolent course in some cases it may behave in a more aggressive manner. Clinical and histological parameters have been shown to correlate with an adverse prognosis but a number of cytogenetic abnormalities may also be associated with aggressive disease. Although the t(14;18) in itself does not affect prognosis in cases of FL, secondary abnormalities that occur in a complex polyploid karyotype may identify cases with a poor prognosis. It is unusual to find both a t(14;18) and a Burkitt translocation in the same tumour, those cases in which it has been described being examples of high grade B cell NHL (either de novo or transformed FL) or B cell acute lymphoblastic lymphoma. In this report 4 cases of FL are described in which both a t(14;18) and a Burkitt translocation were identified at presentation. These cases, and a further 3 already described in the literature, indicate that Burkitt type translocations in FL appear to be an adverse prognostic parameter and suggest that routine cytogenetic studies in cases of lymphoma may define clinically important subtypes of disease.

P200

Genetic Analysis Of CD5+ Diffuse Large B-Cell Lymphoma

{P} MA Catherwood[2], L Venkatraman[1], P Kettle[2]
[1]. Royal Victoria Hospital, Belfast, United Kingdom, [2]. Belfast City Hospital, Belfast, United Kingdom

Background and aim: Microarrays of diffuse large B-cell lymphoma (DLBCL) have shown 2 molecularly distinct groups, germinal centre (GCL) and activated B-cell (ABC) like DLBCL which include de novo CD5+DLBCL. CD5 positivity alone may not confer a poor prognosis as previously reported. The extent of somatic hypermutation (SHM) in the immunoglobulin heavy chain gene (IgH) can help identify germinal centre origin.
Patients and methods: We identified 4 cases of CD5+ DLBCL (2 nodal and 2 extra nodal) in two male and two female patients. Histology and immunohistochemistry was reviewed and PCR for IgH rearrangements were performed to determine gene usage and degree of SHM.
Results: All cases displayed centroblastic morphology. The immunophenotype was CD20+, CD5+, bcl2+, cyclin D1- and high MIB-1. The breast lymphoma and a nodal lymphoma were CD10 negative indicating a non-germinal centre phenotype while CD10 was positive in the other nodal lymphoma and the penile lymphoma. SHM was variable, but greater in the GCL DLBCL with no preferential gene usage. The 2 patients with non-germinal centre phenotype died within 6 months of diagnosis. The other 2 patients are alive at 24 months follow up.
Conclusion: We believe that CD5+DLBCL can be of either germinal centre or non-germinal centre origin. The GCL CD5+DLBCL have a higher rate of somatic hypermutation and better prognosis.

P201

Clinicopathological And Molecular Study Of Gastric MALT Lymphoma Non-responsive To *Helicobacter Pylori* Eradication

{P} L Venkatraman[1], MA Catherwood[2], AL Moore[3], M El-Agnaf[3], TCM Morris[2]
[1]. Royal Victoria Hospital, Belfast, Northern Ireland, United Kingdom, [2]. Belfast City Hospital, Belfast, Northern Ireland, United Kingdom, [3]. Ulster Community Hospitals and Trust, Dundonald, Northern Ireland, United Kingdom

Background and Aims: H.*pylori* eradication is associated with Gastric MALT lymphoma regression in 60-90% of cases. The translocation t(11;18)(q21;q21) resulting in API2-MALT1 fusion is found in 20-30% of gastric MALT and predicts unresponsiveness to H.*pylori*. We present the clinicopathological and molecular findings of 11 gastric MALT patients non- responsive to anti-helicobacter therapy.
Patients and methods: Clinical and histopathological findings were reviewed in 11 patients with gastric MALT. 10 patients were stage IE and 1 was IIE. Clonality was assessed by DNA-PCR for Immunoglobulin (Ig) gene rearrangements and RT-PCR was used to detect t(11;18).
Results: In addition to *H. pylori* eradication, 10 patients received chemotherapy, 1 patient received chemoradiotherapy and 1 patient had partial gastrectomy. 7 patients had concurrent *H.pylori* gastritis. Ig PCR was positive in 7/7 patients in the initial biopsy. t (11;18) was positive in 5/11 patients. Follow-up ranged from 3 to 84 months. 8/11 (77%) patients (including 3 with t(11;18) achieved continuous complete remission (CR). 1 patient relapsed 36 months after CR, received chemotherapy and remains in CR 14 months later. 1 patient had partial remission and 1 patient died of progressive disease.
Conclusion: Patients with early stage gastric MALT lymphoma not responding to Helicobacter eradication therapy may respond to alternative treatment and remain in complete remission regardless of the presence of t(11:18) abnormality.

P202

Immunophenotypes Of Lymphomas In Nigerians

{P} K.A. ADELUSOLA, O ROTIMI, M.A. DUROSINMI
[1]. OBAFEMI AWOLOWO UNIVERSITY, ILE-IFE/OSUN, Nigeria, [2]. ALGERNON FIRTH INSTITUTE OF PATHOLOGY, LEEDS GENERAL INFIRMARY, LEEDS, United Kingdom, [3]. OBAFEMI AWOLOWO UNIVERSITY, ILE-IFE/OSUN, Nigeria

Introduction
The burden of lymphoma on the health care system in Nigeria is enormous. Immunophenotyping is essential for correct classification, treatment and prognosis but this is not a routine practice in Nigeria. This study is on immunophenotyping of lymphomas seen in our institution.
Method
Forty nine (35 males and 14 females with age range 4-7 years) consecutive cases of lymphoma originally diagnosed by H&E only were immunophenotyped using the indirect immunoperoxidase method. Disease staging was by the Ann Arbor system.
Results
The peak age incidence was 21-30 years (24%). There were 31 cases of DLBCL (6 were originally diagnosed by H&E as HL), 6 T-cell lymphoma, 5 follicular lymphoma, 5 HL, 1 ALCL and 1 Burkitt's lymphoma. Lymphadenopathy was the most common presenting sign (67.3%) and the disease was extranodal in 16 cases (33%), most cases (n=6) occurring in the GIT. All the patients were staged IIIB-IVB except one (IIB).
Conclusion
Immunophenotyping identified 6 misdiagnosed cases, underscoring the need for routine use of this method in the diagnosis of lymphoma in Nigeria. We recommend that this should be a routine practice in Nigeria, nay Africa, as in other parts of the world.

P203

The Significance of Mismatch between Framework 2 and Framework 3 IgH PCR Analyses of Clonality in B-Cell Neoplasia

{P} J Le Quesne, R Stitson, K West
Leicester Royal Infirmary, Leicester, United Kingdom

Molecular detection of clonality is a key tool in the diagnosis of B-cell lymphoma. In our centre, possible B-cell neoplasms are routinely tested for clonality by PCR using primers directed across the framework 2 and 3 regions of the IgH gene. In the presence of a clonally derived B-cell population, a single re-arranged IgH is expected, giving rise to a strong band with both sets of primers.
However, it is noted that often only one primer set gives rise to a band, making interpretation difficult. Therefore, this investigation was carried out to identify the significance of mismatch, and why it might occur.
2 years of cases (169) were collated. Associations were sought between mismatch, type of specimen, B-cell clonality (as judged after all investigations) and delay between taking and processing specimens.
Framework 2 alone was found to be 66% specific and 78% sensitive.
Framework 3 alone was found to be 91% specific and 50% sensitive.
Correlations between formalin-fixation of specimens and mismatch, and between the age of paraffin blocks used as a source of DNA and mismatch were observed.
In conclusion, the two framework PCRs are of complementary specificity/sensitivity, and mismatch may be minimised by maximising DNA quality.

P204

HODGKINS DISEASE – The great mimic!

{P} NS Mungalsingh, C Sen
Southmead Hospital, Bristol, United Kingdom

INTRODUCTION
Although the cytological features of Hodgkins disease (HD) have been well documented, it accounts for the most of the false negative diagnoses on aspiration cytology of lymphomas.
MATERIAL and METHODS
We present three cases of histologically proven Hodgkins disease, which had a false negative diagnosis on aspiration cytology.
Case 1:Cytological diagnosis: Granulomatous lymphadenitis.
In the presence of granulomata and a paucity of typical Reed-Sternberg (RS) cells, HD can be misdiagnosed as granulomatous lymphadenitis.
Case 2:Cytological diagnosis: Reactive lymph node.
Unless a high index of suspicion is maintained, Reed Sternberg cells may be overlooked within the reactive background of lymphoid cells that is seen in HD
Case 3: Cytological diagnosis: Metastatic carcinoma
When numerous RS cells are present, they can be mistaken for malignant epithelial cells due to the presence of large nuclei and prominent nucleoli.
CONCLUSION
The cytological diagnosis of HD is challenging. The cytological appearances can mimic low grade NonHodgkins lymphoma, granulomatous lymphadenitis, reactive lymphadenitis and metastatic carcinoma. All lymph node aspirates should be screened for RS cells and when present, RS cells should be carefully scrutinised to ensure that they are indeed RS cells!

P205

Standards, Guidelines And Placental Examinations.

{P} CM Ho-Yen, SV Nayak, C Platt
Department of Histopathology, St Thomas' Hospital, London, United Kingdom

In the United Kingdom one model for selecting placentas for examination is the Association of Clinical Pathologists (ACP) guidelines. In the United States the College of American Pathologists (CAP) have consensual guidelines.

The purpose of this audit was to assess the adequacy of clinical data given in specimen request forms and the adherence to the ACP guidelines for placenta examination.

Our Pathnet database was searched for 300 consecutive placentas between 2004 and 2005. The clinical data in each placenta report was retrieved, recorded and matched with the ACP guidelines for placenta examination.

83% of reports contained adequate clinical history. Where clinical data was adequate according to ACP guidelines, 63% of placentas would have undergone macroscopic and microscopic examination, 3% would have had a macroscopic description only and 9% would have been stored. 25% of placentas did not follow an ACP guideline for submission.

In summary: We have not adopted ACP guidelines for submission of placentas but our results suggest adoption may change workload. We have found that in a minority of cases adequate clinical information is not received with placentas. Our results are comparable to a previous audit of these guidelines. In our opinion, there should be a national standard for placenta examinations.

P206

A female infant with histiocytoid cardiomyopathy and Microphthalmia with linear skin defects (MLS) syndrome

{P} M AL-ADNANI, M MALONE, NJ SEBIRE, M ASHWORTH
Great Ormond Street Hospital for CHildren, LOndon, United Kingdom

Histiocytoid cardiomyopathy is a rare disorder of early childhood identified by the presence of characteristic histiocyte-like cells within the myocardium. It usually affects children under the age of two, with a female predominance, and presents with arrhythmia or sudden death. It can occur in isolation or as part of a syndrome such as Microphthalmia with linear skin defects syndrome (MLS). To our knowledge, there are only two cases of histiocytoid cardiomyopathy associated with MLS syndrome previously reported in the English literature.
Case Report:
A female infant with MLS syndrome who had episodes of tachycardia in utero treated with Digoxin. Tetralogy of Fallot was diagnosed after birth. She died suddenly at home aged six weeks. At autopsy, MLS syndrome with Tetralogy of Fallot was confirmed. The enlarged heart showed mild endocardial fibroelastosis of the right atrium but the myocardium appeared normal. The other organs showed no significant abnormality.
Histologically, the heart showed histiocytoid cardiomyopathy; there were multiple nodules of histiocytoid cells beneath the endocardium of both ventricles and around the atrio-ventricular node and bundle of His. The sino-atrial node was normal. The thyroid showed small nodules of histiocytoid cells. Death was attributed to arrhythmia secondary to histiocytoid cardiomyopathy.

P207

Paediatric Primary Orbital Rhabdomyosarcomas – Subtype Analysis

{P} DJ Fowler, S Dharmaraj, A Upponi-Patil, I Russell-Eggitt, NJ Sebire
Great Ormond Street Hospital for Children, London, United Kingdom

Introduction: Small round cell tumours often have similar morphological features and require immunohistochemistry and molecular techniques for accurate diagnosis. Rhabdomyosarcoma is the most common SRCT found in the orbit in children & may have serious local effects on vision. Light microscopy, immunohistochemistry and molecular techniques are now used to subclassify RMS for treatment, alveolar RMS (ARMS) having a worse prognosis compared with embryonal RMS (ERMS).
Methods: Histopathological features of all orbital RMS at GOSH (1988-2006) were collated. Diagnosis was made on a combination of light microscopic appearances, immunohistochemistry (desmin, myogenin) and molecular techniques (conventional and quantitative realtime PCR for translocations involving PAX3/PAX7 and FKHR) depending on year.
Results: There were 13 cases of paediatric orbital RMS (median age: 2.5 range: 1 month – 9 years): 5 (38%) ARMS and 8 (62%) ERMS.
Discussion: Immunohistochemistry can help in differentiating between RMS subtypes since nuclear myogenin staining is more widespread and intense in ARMS. In contrast, most ERMS show strong cytoplasmic staining for desmin, but only a minority of tumour cells express strong nuclear myogenin and no PAX/FKHR fusion transcripts are detected by conventional or real time PCR. In this small series alveolar subtype comprised 38% of RMS, which is greater than would be expected (approximately 20%) from previous reports (SIOP & IRS). Subtyping, including molecular data, is mandatory for all rhabdomyosarcomas, and will result in more accurate diagnosis leading to improved stratification & therapeutics.

P208

Neonatal Mastocytosis: A Case of Systemic Aggressive Mast Cell Disease due to C-KIT Mutation

{P} DJ Fowler, NJ Sebire, C Turnbull, JI Harper, M Malone
Great Ormond Street Hospital for Children, London, United Kingdom

Introduction: The differential diagnosis of neonatal skin nodules includes congenital infections, leukaemia, metastatic tumours, histiocytic lesions and mastocytosis. Cutaneous mastocytosis is variable & presents with nodules or large macules/plaques or diffuse skin infiltration. Neonatal cutaneous mastocytosis usually spontaneously resolves but in this case there was systemic aggressive involvement, anaphylaxis & sudden infant death.
Case: A 6-day old neonate presented with hepatosplenomegaly, coagulopathy and a widespread nodular infiltrate over the trunk and limbs (sparing palms and soles) with a history of blistering at birth. Skin biopsy showed diffuse dense band-like infiltration of the dermis by a bland monomorphic infiltrate of mast cell tryptase-positive uniform large CD117 (c-kit) positive cells with abundant cytoplasm and folded nuclei. The patient died on d36 following probable anaphylactic cardiorespiratory arrest. Autopsy showed massive infiltrates of mast cells in skin, liver, spleen, kidney, lungs, thymus, lymph nodes & bone marrow. DNA analysis showed KIT D816V mutation, which is reported in systemic mastocytosis.
Discussion: The C-KIT D816V missense mutation strongly activates KIT by altering the enzymatic site of the KIT protein & has only been reported as an acquired somatic change, not as a germline change. Familial mastocytosis with GISTs are reported rarely but are associated with germline mutations that activate KIT by affecting the regulatory domain rather than the enzymatic site. Kinases with enzymatic site mutations show different inhibitor sensitivity profiles than regulatory-type mutations, which is of potential pharmacological importance. Classification of mast cell diseases should include molecular genetics to be prognostically & therapeutically useful in future.

P209

Expression of P_{53} in BCC tissues and adjacent non tumoral epidermis from sun-exposed areas of head and neck regions

{P} Parvin Rajabi Dehnavi, Nooshin Afshar Moghadam, Mohammad Ali Rajabi
Isfahan university, Isfahan, Iran

Background and Objective: P53 mutations are common in a variety of human tumors. Paradoxical findings have been reported about the frequency of P53 mutation in basal cell carcinoma, which point out the necessity of an exact study on this subject. The intensity of P53 expression would also be a helpful key to make a more exact prognosis.
Methods: This descriptive – analytical study was performed retrospectively over a 5-year period on 150 basal cell carcinoma cases in the pathology department of Alzahra hospital in Isfahan (Iran).
Paraffin blocks stained immunohistochemically for P53 .The intensity of immunoreactivity was graded. Age of the patients was recorded as well.
Result: Positive P53 immunoreactivity was observed in 123 basal cell carcinoma (82%) and in 117 adjacent normal epidermal tissues (78%) (P=0.38). Frequency of severe immunoreactivity in tumoral tissue and adjacent normal epidermis were 46% and 32% respectively (P=0.046).
Conclusion: No significant differences was detected between P53 immnoreactivity in tumoral tissue and adjacent non tumoral epidermis. Intensity of P53 immnoreactivity was greater in tumoral specimens. Comparison of mean ages showed a significant differences between P53 expressing and non-expressing groups.

P210

The Expression of Oestrogen Receptor Beta1 (ERB1) in Squamous Cell Carcinoma of the Skin

{P} D Pathak[1], V Speirs[2], A M Shaaban[3]
[1]. Department of Histopathology, Leeds General Infirmary, Leeds, United Kingdom, [2]. Breast Research Group, Leeds Institute of Molecular Medicine, St James's University Hospital, Leeds, United Kingdom, [3]. Leeds General Infirmary, Leeds, United Kingdom

Introduction: Oestrogen receptor beta (ERB) has recently been identified in the dysplasia supports an extended follow up interval.
breast as well as in various tissues. Several isoforms to ERB have been described, the most prevalent of which is ERB1. The development of reliable antibodies has allowed the study of the gene at protein level.

Materials and Methods: In the present study, using the monoclonal antibody PPG5/10, ERB1 expression has been analysed in 100 paraffin-wax embedded sections of squamous cell carcinomas of the skin. The percentage of expression, intensity were analysed and the Allred score was calculated. The expression was correlated with demographic data and the known histological prognosticators.

Results: The majority of cases expressed the protein. Allred score of >4 was seen in 92% cases. The median percentage of nuclear expression was 75% (range 2 – 98 % and interquartile range 60 – 90%). A trend towards higher expression (score >4) was noted in well and moderately differentiated tumours (P=0.07) when compared with poorly differentiated carcinomas. Cytoplamic expression was significantly higher (P=0.001) in well and moderately differentiated tumours. There was no significant correlation of expression with age, gender, thickness of the tumour, lymphovascular invasion or state of margins.

Conclusions: Our data show high expression of ERB1 in squamous cell carcinoma of the skin and suggest that ERB1 might have prognostic significance in these tumours. To our knowledge this is the first report of ERB expression in squamous cell carcinoma of skin.

Abstracts

Oral

O1

Biopsy technique and outcome in long segment Barrett's oesophagus

J.E. Abela, {P} J.J. Going, M. McKernan, R.C. Stuart
University of Glasgow, Glasgow, United Kingdom

Background. Management of cancer risk in patients with Barrett's oesophagus remains controversial. *Method.* Separate cohorts of patients with long-segment Barrett's oesophagus were followed in a prospective, non-randomised study. Group A: (n 178), 4-quadrant biopsies were taken systematically. Group B: (n 183) biopsies were taken at random. Detection and treatment of high-grade dysplasia ± adenocarcinoma constituted the primary end-point. Prevalent adenocarcinomas diagnosed up to 6 months after index endoscopy were excluded. *Results*. Patient age, metaplastic segment length and follow-up were comparable. Prevalent low and high grade dysplasia were identified in 32, 5 group A patients but only 2, 0 group B patients. Incident low and high grade dysplasia were identified in 4, 4 group A patients and 12, 1 group B patients. In group A the annual end-point incidence was 1.1% per patient-year, with 8 high-grade dysplasia patients and one with intramucosal adenocarcinoma receiving curative treatment. In contrast 3 group B patients presented with incurable invasive adenocarcinoma, giving an end-point incidence of 0.6% per patient-year. *Conclusions*. Quadrantic biopsy much more reliably detects dysplasia and early adenocarcinoma in Barrett's oesophagus than random sampling. In the absence of dysplasia following systematic biopsy the low incidence of new dysplasia supports an extended follow up interval.

O2

The Clonal Origin of Human tumours: is Familial Adenomatous Polyposis (FAP) different?

{P} SJ Leedham[1], LC Maia[1], O Seiber[1], SL Preston[1], SAC McDonald[1], IPA Tomlinson[1], MR Novelli[1], NA Wright[2]
[1]. Cancer Research UK, London, United Kingdom, [2]. Barts and the London School of Medicine, London, United Kingdom

We teach our students that human tumours are monoclonal in origin. However, our previous work (*Science* 1996; 272:1187) has shown that 76% of microadenomas in FAP are polyclonal, a result confirmed in mice (Dove W *PNAS* 1997; 94:1392; 2005; 102:6960). We have analysed a series of adenomas for clonality in FAP of different types by detecting the site of the hit in the second allele of the *Apc* gene, using contiguous microdissected crypts. We find that in cases of classical FAP the lesions are polyclonal, whereas in attenuated FAP, where the lesions are much fewer, adenomas are clonal for the same mutation in the second allele. These results raise several questions: (i) are the earliest lesions – the microadenomas - in FAP polyclonal from their onset? (ii) as lesions grow, do they revert to monoclonality as one clone outgrows the rest? (iii) Can these results be explained on the basis of very early collision of crypts with a second *Apc* mutation? If the latter, the proportion involved would indicate some form of active recruitment of *Apc* -/- crypts, possibly through epithelio-mesenchymal interactions. Whatever the mechanism, this evidence of early co-operativity between the early neoplastic crypts is intriguing and warrants closer study.

O3

Chromosome 20q Amplification in Colorectal Adenoma to Carcinoma progression

{P} B. Carvalho, C. Postma, S. Mongera, B. Ylstra
VUMC- VU University Medical Center, Amsterdam, Netherlands

About only only 5% of colorectal adenomas ever progress to cancer, and gains at chromosomes 8q22-q23, 13q21-q31 and 20q13 are associated with this progression (Hermsen et al. Gastroenterol 2002;123:1109-1119). We aimed to further delimit the 20q amplicon and unravel the genes that drive this amplicon.
Array-CGH was performed on 41 paired samples from both adenoma and carcinoma compartments of malignant polyps.. An independent set of 73 frozen tumours (34 adenomas and 39 carcinomas) was also analysed by array-CGH and by expression microarrays, to validate candidate oncogenes.
The analysis of both the learning set and validation set showed the same regions to be involved. We observed 8q, 13q and 20q gains in the carcinomas and in adenomas that had proven to progress to carcinoma (malignant polyps), in much higher frequency than in non-progressed adenomas. Focusing on 20q, we were able to delimit three smallest regions of overlap (20q11.2, 20q13.13-q13.2 and 20q13.3) spanning segments of 3 – 6 Mb. Microarray expression data provided multiple candidate genes mapping at 20q which are significantly overexpressed in tumours with 20q gain, including DNMT3B (20q11.2) and ZNF217 (20q13.3).

O4

Characterization of Side Populations of Human Gastrointestinal Cell Lines

{P} J Burkert, WR Otto, D Davies, K Allen, J Paris, NA Wright
Cancer Reseach UK, London, United Kingdom

A population of cells known as the Side Population (SP) can efflux the nucleic acid binding dye Hoechst 33342, and shows stem cell features. SP cells have been discovered in haematopoietic malignancies and solid tumours of the brain, breast and lung, and may represent cancer stem cells (CSC). We have identified SP cells in several human gastrointestinal tumour cell lines and are investigating whether they may be CSCs. Six tumour cell lines (5 colorectal; 1 gastric) showed a distinct SP phenotype, comprising between 0.3% and 17% of the population. Flow sorted SP cells showed clonal self-renewal and differentiation *in vitro,* and the ability to generate tumours in nude mice *in vivo* has been invetigated. By multicolour FACS analysis, SPs were tested for putative stem cell markers CD34, CD44, CD133, cKIT and BCRP1, which were also confirmed by RT-PCR. SP clones were tested by immunohistochemistry for intracellular stem cell markers Musashi-1, Hes-1, Oct-4, β-catenin, Bcl-2, and Nanog. Further differences of the subpopulations with regards to differentiation potential and growth characteristics in long-term cultures are being investigated. These studies may reveal useful information regarding the isolation and therapeutic targeting of the CSC population.

O5

Audit of management of inadequate smears

{P} I BABARINSA[1], J MATHEW[2], A ST JOHN[2], A OLADIPO[1]
[1]. Department of Gynaecology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom, [2]. Department of Histopathology, Royal Cornwall Hospital, Truro, Cornwall, United Kingdom

Introduction: Inadequate cervical cytology constitute about 9.3% of all smears. It reflects the skill of the smear taker, adequacy of smear fixing, proficiency of the cytoscreener/cytopathologist or a combination of these factors.
The cervical screening programme (NHSCSP) sets standards against which all participating services are benchmarked. The guidelines stipulates a review of practice if any laboratory performs outside a given inadequate range (5.8-12.9%).
Aim: To ascertain our compliance with national guidelines.
Method: Retrospective review of patients with inadequate cervical cytology between August 2004 and May 2005 was undertaken. Compliance with set standards were benchmarked against each patient referred to the colposcopy clinic.
Results:53 patients were identified during the review period, however seven patients were excluded from the audit as they were referred after only one reported inadequate smear. None of the patients had surgical treatment for their referral cytology; representing 100% compliance with one of the standards. 95.2% of the patients were referred after 3 consecutive inadequate cytology, comparing favourably with recommend-ations.
52.14%.complied with being referred within three months of third inadequate cytology. Majority of inadequate cytology reverted back to normal within 24 months of the report.
Conclusion :Compliance with the national standards was demonstrated in our practice which resulted in curtailing against over-treatment, better use of resources (clinic attendance, histological review) alleviation of anxiety for patients and improved delivery of care.

O6

The Heat Sink Effect May Cause Inadequate Tissue Ablation: A Histological Evaluation Of Microwave, Radiofrequency and Cryoablation In The Rat Liver.

{P} N Bhardwaj, A Strickland, L Atanesyan, F Ahmad, K West, D Lloyd
University Hospital Leicester Royal Infirmary, Leicester, United Kingdom

BACKGROUND. Complete cell death is necessary for any ablative technique to be successful. Microwave tissue ablation (MTA), radiofrequency (RF) and cryotherapy (CT) are three means of destroying tumours. Unlike MTA, RF and cryotherapy rely on conduction of a thermal insult for their cytotoxicity, potentially affecting lesion uniformity.
AIMS/OBJECTIVES. To compare the uniformity of demarcation of lesions induced by MTA, RF and CT in the rat liver.
METHODS. Twenty-one rats were assigned to the three ablative modalities.
RESULTS. All specimens exhibited macroscopically well-demarcated lesions. Microscopically, the most clearly delineated lesions followed MTA with no viable hepatocytes or peri-vascular survival. All vessels and bile ducts had undergone complete necrosis. CT specimens showed more prominent inflammation at the lesional edge than MTA and peri-vascular hepatocyte survival within the lesions. RF showed hepatocyte survival in the ablated area and conspicuous peri-vascular hepatocyte survival with evidence of a very irregular lesion edge
CONCLUSION. Unlike RF and CT, MTA induced microscopically well-demarcated lesions, with no intralesional hepatocyte survival. Intralesional cell survival in RF and CT may be due to the relatively prolonged treatment times needed, allowing thermal energy to dissipate via blood flow. This is known as the heat sink effect.

O7

Functional analysis of disease associated mutations in SEPT9

{P} PA Hall, S McDade, P Hyland, L Chatham, SEH Russell
Centre for Cancer Research & Cell Biology, Queens University Belfast, Belfast, Northern Ireland, BT97AB, United Kingdom

Disease-associated mutations can provide insights into the functions of cellular proteins, and rare genetic diseases can have lessons that can be generalised to more common scenarios. We have reported that SEPT9 is over-expressed in neoplasia with a dramatic alteration in the expression of the SEPT9_v4 and v4* transcripts: v4* being predominant in tumours. SEPT9_v4 and v4* are distinct transcripts that encode the same polypeptide but differ only in their 5'UTRs. In hereditary neuralgic amyotrophic (HNA), mutations have been described in exon 3 of SEPT9, a region present in the 5'UTR of both SEPT9_v4 and v4*. In a comprehensive series of experiments utilising reporter constructs, site directed mutagenesis and deletion mapping, we have shown that the unique 5' UTR regions of SEPT9_v4 and V4* can profoundly influence the efficiency of the translation of these messages. We found an IRES (internal ribosome entry site) in the common regions of the 5'UTR and short in-frame open reading frames in the v4 message modulate this activity. A range of cellular stresses, including DNA damage and hypoxia, alter the translational efficiency of the v4 but not the v4* message. We have engineered the mutations seen in HNA and show that these alter the translational efficiency of SEPT9_v4 and v4*. Crucially the stress induced modulation of SEPT9_v4 translation is lost. The consequence of this is deregulated SEPT9_v4 protein expression. SEPT9_v1 isoform binds to HIF1alpha and modulates its activity, but this binding is modulated by SEPT9_v4 protein. We have engineered the exon 3 HNA mutations in SEPT9_v1 and investigated how this modulates HIF1alpha binding. These and our previous data places translational control of SEPT9_v4 protein as a regulator of septin protein complexes and modifiers of HIF1alpha, and provide profound mechanistic insights into septin biology and how septins are deregulated in disease.

O8

DC-SIGN association with the Th2 environment of lepromatous leprosy lesions: cause or effect?

{P} EJ Soilleux[1], EN Sarno[3], MO Hernandez[3], E Moseley[7], J Horsley[7], UG Lopes[4], MJ Goddard[7], SL Vowler[5], N Coleman[2], RJ Shattock[6], EP Sampaio[3]
[1]. Nuffield Department of Clinical Laboratory Sciences, John Radcliffe Hospital, Oxford, Oxford, Oxfordshire, United Kingdom, [2]. Medical Research Council Cancer Cell Unit, Hutchison/M.R.C. Research Centre, Cambridge, Cambridgeshire, United Kingdom, [3]. Oswaldo Cruz Institute, FIOCRUZ, Manguinhos, Rio de Janeiro, Brazil, [4]. Instituto de Biofisica Carlos Chagas Filho, Federal University of Rio de Janeiro, Ilha do Fundao, Rio de Janeiro, Brazil, [5]. Cambridge University Centre for Applied Medical Statistics, Department of Public Health and Primary Care, Cambridge, Cambridgeshire, United Kingdom, [6]. Department of Cellular and Molecular Medicine, Infectious Disease, St George's Hospital Medical School, London, United Kingdom, [7]. Department of Histopathology, Papworth Hospital, Papworth Everard, Cambridgeshire, United Kingdom

The clinical spectrum of leprosy is related to patients´ immune responses. Non-responsiveness towards *M. leprae* seems to correlate with a Th2 cytokine profile. The reason for such a polarised immune response remains unclear. The C-type lectin, DC-SIGN (dendritic cell-specific, ICAM-grabbing non-integrin), which is known to bind HIV and a number of other pathogens, is expressed by subsets of dendritic cells and macrophages and has previously been associated with Th2 responses. Here we show abundant DC-SIGN expression in lepromatous, but not borderline tuberculoid, leprosy, in both HIV-positive and HIV-negative patients. Moreover, we demonstrate that DC-SIGN can act as an entry receptor for *M. leprae*, as it does for *M. tuberculosis*, through the cell wall component lipoarabinomannan. DC-SIGN is expressed on virtually all *M. leprae*-containing cells, providing further evidence for its role as a receptor. DC-SIGN may therefore be induced on macrophages in lepromatous leprosy and may then contribute to mycobacterial entry into these cells.

O9

Laser capture microdissection and bioinformatics profiling to identify tumour vascular markers

BD Tarlow, LA Strickland, Z Modrusan, TD Wu, {P} HK Koeppen
Genentech, Inc, South San Francisco, CA, United States

BACKGROUND: Tumour-associated vasculature has been demonstrated to be qualitatively different from normal vasculature. Gene expression changes in tumour endothelium have been difficult to study because these cells comprise only a small fraction of all cells in a heterogeneous tissue. Here, we enriched endothelial cells from human colon tumours with laser capture microdissection (LCM) to identify a tumour-vascular gene expression signature.
METHODS: Vasculature in malignant human colon tissues was identified using immunofluorescent labelling for CD146 followed by LCM with the SL-μCUT system (MMI). Gene expression was analyzed with Agilent microarrays. We generated a ranked list of molecules preferentially expressed in the tumour vasculature.
Next, we took a bioinformatics approach by utilizing a gene expression database of benign and malignant colon to identify genes whose expression correlated with vascular density. Genes expressed in a pattern that correlated with vascular density in the malignant colon but not benign colon, and discovered in LCM profiling, generated a tumour-vascular gene expression signature.
RESULTS:
Utilizing a combination of LCM and bioinformatics, we identified pan-endothelial markers (CD31, von Willebrand Factor, PDGFR-b) and tumour vasculature markers (SPARC, TEM7, Thrombospondin-1). We demonstrated the feasibility of utilizing LCM and bioinformatics profiling to identify molecules preferentially expressed in tumour vasculature.

O10

Measurement of Lymphoma Gene Signatures by Real-time PCR in Globally Amplified PolyA cDNA from Paraffin Embedded Tissue

{P} J Goodchild[1], E Sakhinia[1], C Glennie[1], L Menasce[2], JA Radford[2], JA Hoyland[1], RJ Byers[1]
[1]. University of Manchester, Manchester, United Kingdom, [2]. Christie Hospital, Manchester, United Kingdom

Microarray gene expression profiling has identified gene signatures, or "*Indicator*" genes, predictive of outcome in diffuse large B-cell lymphoma (DLBCL) and follicular lymphoma (FL). In order to evaluate use of these for diagnosis tool we have demonstrated use of a simple, practical *polyA PCR* based method for analysis of *Indicator* profiles in frozen samples of DLBCL and FL which we here tested use of in formalin fixed paraffin embedded tissue (FFPET).

PolyA cDNAs were prepared from 40 FFPET lymph nodes (18 FL, 22 DLBCL) and analysed by quantitative real-time PCR for expression of 36 *Indicator* genes, each of which had been measured previously in parallel frozen samples. Expression levels were normalised using the mean of four housekeeping genes (IF2-b, Gap, Rbs9 and beta actin).

Housekeeping genes were detectable at high levels in all 40 cases, though the 36 *Indicator* genes were expressed at a lower level. Despite this there was a statistically significant difference in gene expression between DLBCL and FL for HSP27 ($p<0.02$).

The results demonstrate ability to quantify gene expression level of prognostic genes in FFPET, and identified HSP27 as a diagnostic discriminator for DLBCL and FL, suggesting practical extension of the technique to FFPET for diagnostic purposes.

O11

Cytokine Involvement In The Pathogenesis Of Disc Degeneration: IL-1 Or TNF Alpha?

{P} CL Le Maitre, AJ Freemont, JA Hoyland
University of Manchester, Mancheter, United Kingdom

Intervertebral disc (IVD) degeneration is a major cause of low back pain. Cytokines (particularly IL-1 and TNF) have been implicated in its pathogenesis. However, to date no studies have investigated their expression concurrently in IVD degeneration.
Using real time PCR we investigated the gene expression of IL-1 and TNF and their related receptors in 80 human IVDs. Additionally immunohistochemistry was used to localise the cytokines and their receptors in 40 human IVDs.
IL-1 gene expression was observed in a greater proportion of IVDs than TNF (79% v/s 59%). Degenerate and prolapsed discs displayed higher levels of both cytokines than non-degenerate discs, but IL-1 was seen at higher levels than TNF. In addition an increase in both gene expression and protein production was observed for the IL-1 receptor in degenerate compared to non-degenerate discs but no increase was seen for the TNF receptor
We have shown that both cytokines are present in human IVDs. However, IL-1 appears to be expressed at higher levels and in more discs especially during disc degeneration. This suggests that although both cytokines may be involved in the pathogenesis of disc degeneration, IL-1 may have a greater role than TNF.

O12

Accelerated Cellular Senescence In Human Intervertebral Disc Degeneration

{P} CL Le Maitre, AJ Freemont, JA Hoyland
University of Manchester, Manchester, United Kingdom

Low back pain is a common, debilitating and economically important disorder. Current evidence implicates intervertebral disc (IVD) degeneration as a major cause, but its pathogenesis is poorly understood. Many of the characteristic features of disc degeneration mimic those seen during aging, but occur at an accelerated rate. This study investigates the involvement of cellular senescence in disc degeneration.
Immunohistochemical investigation for $P16^{INK4A}$ in normal & degenerate human IVD was performed. Cells from normal and degenerate IVDs, were used to assess cell replication potential, telomere length and gene expression for $P16^{INK4A}$, MMP13 and ADAMTS 5 gene expression.
Normal IVDs showed an age-related increase in cellular expression of $P16^{INK4A}$ and a decrease in telomere length. Degenerate IVD cells, even from young patients, exhibited increased expression of $P16^{INK4A}$, reduced telomere length and a decrease in replication potential. A direct correlation was observed for gene expression of $P16^{INK4A}$ and the matrix degrading enzymes MMP 13 and ADAMTS 5.
This study has shown that degenerate IVDs show accelerated cell senescence compared to non-degenerate discs. Furthermore the senescent phenotype was linked to characteristic features of ageing and degeneration of the IVD.

O13

Diagnostic Accuracy Of Conventional Smears And ThinPrep Samples In Cervical Screening

{P} Bijal Shah, Durgesh Rana, Janet Marshall, Mina Desai
Manchester Royal Infirmary, Manchester, Lancashire, United Kingdom

Liquid Based Cytology has been recommended in England and Wales as the primary means of processing samples in cervical screening programme. We conducted this study to compare the positive predictive value (PPV) of conventional smears (CS) and ThinPrep samples (TP) reported as high-grade squamous dyskaryosis and glandular neoplasia from September '01 to August '05. The performance of both techniques was compared with histological results.

The total number of CS and TP were 373,990 (86.8%) and 48,216 (11.2%) respectively. Inadequate rate for CS and TP was 10.68% and 1.64% respectively. PPV of moderate and severe dyskaryosis including invasive carcinoma in CS was 71.7% and 90.4%; while for TP, it was 65.8% and 89.4% respectively. PPV of glandular neoplasia for CGIN/Adenocarcinoma was 66.7% and 64.3% in CS and TP respectively. PPV of glandular neoplasia for CIN2 and more in CS and TP was 82.7% and 85.7% respectively. The PPV for a diagnosis of CIN2 or worse in CS and TP was 79.3% and 73.3% respectively. They are within acceptable range of the U.K. (65-87%). The difference in PPV for varied categories in both systems is statistically insignificant.

In general, interpretation requires less time and inadequate rate is significantly reduced with TP.

O14

Stromal Nodules And Vessel Wall Thickening In TURP Specimens Are Associated With Failure Of Alpha-Blocker Treatment

{P} M-A Tran-Dang[1], R. D. Smith[2], B Khoubehi[2], A Patel[2], M. M. Walker[2]
[1]. Royal Berkshire Hospital, Reading, United Kingdom, [2]. St Mary's Hospital, London, United Kingdom

Patients on alpha-blockers for benign prostatic hyperplasia (BPH) may have a finite period of benefit before they suffer disease progression, eventually requiring surgery (TURP). Prominent stromal nodules and vessel wall thickening have been noted in TURP specimens taken from patients on alpha-blockers and are proposed to be pathological indicators of failure of medical treatment. Alpha-blocker treatment and retention status were obtained for 60 men with BPH confirmed at TURP. The number of stromal nodules/section and the degree of vessel wall thickening were recorded blinded to the clinical detail. Patients on alpha-blockers had significantly more stromal nodules/section than untreated men ($p=0.04$ Mann-Whitney U-Test). Vessel wall thickening was present in 33/35 men taking alpha-blockers and in 15/25 men who were not ($p=0.01$, Fisher's Exact Test) and in 16/16 men who had acute urinary retention (AUR) on alpha-blockade compared with 8/22 men in retention who were not ($p<0.0001$, Fisher's Exact test). These data support the conclusion that stromal nodules and vessel wall thickening are pathological features of BPH significantly associated with alpha-blocker treatment. The prevalence of vessel wall thickening in men developing AUR on alpha-blockers may be associated with stromal nodule overgrowth, thereby suggesting a possible mechanism for the failure of medical management.

O15

Hadfield's Procedure – Pathology review And Appropriateness Of Surgical Treatment

{P} a naveed, b benatar, a benatar, s Ellenbogen
tameside genreal hospital, manchetser, United Kingdom

Hadfield's procedure involves the excision of the major duct system. It is indicated in patients with multiple duct nipple discharge, mamillary fistula and recurrent subareolar breast abscesses. The aim was to evaluate the appropriateness of surgical treatment by correlating the results of excision specimen with preoperative diagnosis. The pathology of the 35 cases was reviewed for periductal mastitis. ductal ectasia, duct papilloma and malignancy. Patient satisfaction was assessed with a postal questionnaire. The clinical and the histological conclusions were established. It was concluded clinically that Hatfield's procedure is associated with a high rate of complications. The histology concluded that the histological correlation between the excision specimen and the preoperative diagnosis was seen in less than half of the patients and DCIS found in one patient. The procedure can be offered to patients in whom one of the above pathology is strongly suspected. Many patients attending clinics can be reassured and considered for expectant treatment.

O16

The area of involved lymph nodes in colorectal carcinoma. Lessons for the radiologist and pathologist

{P} ML BAXANDALL, F LEWIS, G CASALI, P GUILLOU, H THORPE, J WALKER, D JAYNE, AMH SMITH, RM HEATH, JM BROWN, P QUIRKE
Leeds University, leeds, United Kingdom

INTRODUCTION: Radiologists are improving their techniques for imaging colorectal cancer. They have a minimum detection limit of 3x3mm. We have analysed over 2600 lymph nodes in 213 cases of colorectal carcinoma and quantified the area of involved and uninvolved nodes. This data is also important in understanding the importance of finding small lymph nodes for pathologists.

METHOD: The areas of 2673 lymph nodes were measured on digital files using the aperio scanning system on 213 cases from the MRC Classic trial. Median age 71.2 (32-94). Node count for each case was noted and area of lymph node calculated by an alogram pixel count.

RESULTS:126 nodes were positive for cancer (median 20.43mm² range 2.11-170mm²). 2547 were negative (median 8.02 mm² range 0.3-141.3mm²) $p < 0.001$ Rank-Wilcoxon non parametric analysis.20% of positive nodes are smaller than 9.0mm².The larger the node the more likely it was to be cancer positive.

Table 1 probability of cancer positive node by size

Size of lymph node(mm²)	Probability of tumour positivity
0-2.11	0%
2.12-5	1.9%
2.13-30.8	11.9%
30.9-83	33.3%
83.1-120	77.7%
120.1-170.9	100%

CONCLUSION The larger the node the more likely it is to be positive for tumour. However the smallest positive node detected was 2.11mm².It is these small positive nodes which may be missed and lead to under staging by radiologists and pathologists.

O17

Cervical Stromal Sarcoma: A Rare Neoplasm

{P} Preeti Jaiswal, Al Samarrai
RCOG, London, United Kingdom

Endocervical stromal sarcoma is an exceedingly rare neoplasm. With incidence of 0.4% of cervical malignancies. Histologically these tumours are composed of fascicles of spindle shaped cells.
We present a case report of 80 years old patient presenting with postmenopausal bleeding. On clinical examination, large fungating mass was found on cervix and provisional diagnosis of cervical cancer was made. She had past history of intracavitary radiotherapy for cancer cervix 20 years back.
Biopsy of the lesion showed prolapsed leiomyoma showing changes due to ulceration. Vaginal hysterectomy was attempted but uterus could not be felt and cervical stump was removed. The mass was anterior to cervix and when it was released, it was found to be invading the bladder.
The histology showed endocervical stromal sarcoma with pleomorphism and prominent mitotic activity. MRI revealed 5cm x 4cm x 3cm abnormality of lower uterine body and cervix. Anteriorly indistinguishable from bladder but free posteriorly. She is being referred to tertiary centre in London for further treatment.
The rarity of these tumours has contributed to paucity of information regarding management and prognosis of patients with cervical sarcomas.

O18

Application Of A Modified Histological Activity Index In Primary Biliary Cirrhosis

{P} A Awasthi[1], A Smith[2], TW Warnes[2], RFT McMahon[3]
[1]. Department of Histopathology, Manchester Royal Infirmary, Manchester, United Kingdom, [2]. The Liver Unit, Manchester Royal Infirmary, Manchester, United Kingdom, [3]. University of Manchester, Manchester, United Kingdom

Primary biliary cirrhosis (PBC) is a chronic liver disease of autoimmune aetiology affecting middle aged women with positive antimitochondrial antibodies, characterised by bile duct centred inflammation, granulomas, bile duct loss and fibrosis. The histological features have traditionally been assessed by the Ludwig grading system, which includes both features of necroinflammatory activity (portal and periportal hepatitis) and fibrosis stage. In other chronic liver diseases such as autoimmune chronic hepatitis and chronic viral hepatitis B and C, the necroinflammatory (grade) and fibrotic (stage) components have been separated using histological activity indices such as the Ishak system. In this clinicopathological study of 112 patients with 180 biopsies suitable for fibrosis staging and 172 for necroinflammatory grading, the Ishak system was applied as well as the conventional Ludwig stage. Additional features with particular relevance to PBC were also included, including the presence of granulomas, the total number of portal tracts and bile ducts (with portal tract/bile duct ratio), the presence of copper associated protein, sinusoidal fibrosis and dilatation, and venular changes. Ludwig stage 4 (n=33) correlated well with Ishak staging (4-6), while Ludwig stage 3 (n=94) ranged more widely from Ishak 2-5. Orcein-positivity indicative of chronic cholestasis correlated positively with portal and interface hepatitis and negatively with the bile duct-portal tract ratio. These results suggest that a modified Ishak grading and staging system can be applied to a chronic cholestatic condition such as PBC and provides a baseline for comparative studies relating to clinical parameters of this disease such as wedged hepatic vein pressures and dynamic studies of liver function.

O19

A Comparison Of Cervical Screening Histories In Women Developing Intraepithelial Neoplasia And Invasive Carcinoma

{P} I Morrison, A Deery
St George's Healthcare NHS Trust, London, United Kingdom

Introduction: Our aims were to investigate smear histories in cases of high-grade intraepithelial neoplasia and invasive carcinoma, determine smear accuracy retrospectively by histological outcome and assess differences in cytological detection of squamous and glandular disease.
Methods: Women diagnosed with high-grade intraepithelial neoplasia or invasive carcinoma on histological examination had their smear histories analysed. These patients were selected over a five-year period at a large teaching hospital.
Results:
74% diagnosed with cervical intraepithelial neoplasia grade 3 (CIN3) had a complete smear history. 80% were referred with a single smear of cytological pattern 7, 4 or 5.
46% diagnosed with squamous cell carcinoma had a complete smear history. 56% were referred with a single smear of cytological pattern 7,4 or 5.
76% diagnosed with adenocarcinoma in situ (AIS) had a complete smear history. 45% were referred with a single smear of cytological pattern 6.
67% diagnosed with adenocarcinoma had a complete smear history. 33% were referred with a single smear of cytological pattern 6.
Discussion: More women are presenting with cancer and a complete smear history, a reversal of previous trends. The cytological identification of glandular neoplasia and combined squamous and glandular neoplasia needs improving. Further education to aid recognition of normal and abnormal glandular appearances is necessary.

O20

Development of an *in vitro* Model of Prostate Carcinoma Metastasis to Bone

{P} J Roulson[1], MD Brown[1], TD Allen[1], JH Shanks[2], NW Clarke[2]
[1]. Paterson Institute for Cancer Research, Manchester, United Kingdom, [2]. Christie Hospital, Manchester, United Kingdom

Prostate carcinoma (CaP) is the most common cancer in men in the UK. Malignant cells spread to bone via peripheral blood and must cross the endothelial lining of vessels to enter the marrow. Both benign and malignant prostate epithelial cells (PEC) bind preferentially to bone marrow endothelium (BME) and grow in co-culture with bone marrow stroma. However, only malignant PEC are able to migrate across BME, indicating this is the critical step in bone metastasis, and understanding this process may lead to the development of antimetastatic therapies.

In order to study the physical and molecular interactions between PEC and BME we have developed an *in vitro* model of endothelial transmigration utilising the human bone metastasis derived PEC cell line (PC-3) and a human BME cell line (BMEC). Using this system, we have observed PC-3 cells completing migration across BME in 249 +/- 48 minutes, with virtually all PC-3 cells completing invasion. Our model provides a useful *in vitro* system for studying the interactions between PEC and BME. This model will be used to examine the role of endothelial junctional proteins during transmigration, and involvement of cell adhesion molecules in binding and signalling between PEC and BME.

O21

Analysis of MMP Single Nucleotide Polymorphisms in relation to breast cancer progression

{P} S Hughes[1], D Holliday[1], J Shaw[2], S Duffy[3], I Hart[1], L Jones[1]
[1]. Centre for Tumour Biology, Institute of Cancer and CR-UK Clinical Centre, Bart's and The London, Queen Mary's School of Medicine & Dentistry, London, United Kingdom, [2]. Department of Molecular Medicine and Cancer Studies, Robert Kilpatrick Building, University of Leicester, Leicester, United Kingdom, [3]. Cancer Research UK Centre for Epidemiology, Wolfson Institute of Preventive Medicine, London, United Kingdom

Matrix Metalloproteinases (MMPs) have an established role in cancer. We have evaluated the association between polymorphisms in the promoter regions of MMP-1 (1G/2G -1607 bp), MMP-3 (5A/6A -1171 bp), MMP7 (A/G -181 bp), MMP9 (C/T -1562 bp), MMP12 (A/G -82 bp) and MMP-13 (A/G -77 bp) and metastasis of breast cancer in 88 lymph node negative and 70 lymph node positive patients. All genes except MMP13 showed Hardy-Weinberg equilibrium. Data were analysed by logistic regression. To increase statistical power, trends in risk of node positive disease with number of copies of the 'unfavourable' genotype were tested for and the effect of having any copies of the unfavourable genotype compared to none. For MMP9, there was a significantly increased risk of node positive disease for genotype CT ($p=0.03$). For MMP1 there was a borderline significant increase in risk with number of copies of genotype the G allele ($p=0.06$). For either one or two copies of G compared to no copies, the result was not significant but suggestive (OR=1.84, $p=0.2$). There were no significant associations with risk of node positive disease for MMP3, MMP7, MMP13 or MMP12. However, for MMP7, the results look suggestive, when AG and AA were combined their odds ratio was 1.77 ($p=0.3$).

O22

Breast Cancer Profiling: A Minimised Panel of Predictive Markers for Survival

{P} N Thompson[1], S Leigh-Bramwell[1], DG Powe[1], G Ball[2], E Rakha[1], J Williams[1], EC Paish[1], IO Ellis[1]
[1]. Nottingham University, Nottingham, United Kingdom, [2]. Nottingham Trent University, Nottingham, United Kingdom

Aim: Breast cancer is a heterogeneous disease but recent studies have succeeded in segregating patients into groups according to their survival outcome using gene expression. Here, we sought to find a minimised panel of antibodies to predict patient survival.
Method: Tissue microarrays comprising 400 successive breast tumours were immunostained using a panel of antibodies selected for their relevance in predicting cancer progression. These included ER, PR, Her-2, Mib-1, Bcl-2, MUC-1, CD24, E-cadherin, and MMP-11. H-score morphometry was used to compare tumour staining to 10 normal breast controls and subjects were grouped according to survival using hierarchical clustering. Furthermore, the drivers for group membership were determined using bioinformatics.
Results: Five biomarkers showed a significant difference, four did not, in expression between cancer and normal tissue. Cluster analysis identified 4 patient groups, separated by differences in overall survival, conferring significant differences in tumour grade.
Conclusion: This pilot study suggests that immunophenotyping of routinely-processed paraffin breast tumours could be used to predict patient survival. Bioinformatics will be used to further refinine the design of a minimal predictive panel of antibodies as an adjunct to conventional tumour grading. Future studies will assess the panel for predicting tumour recurrence.

O23

EGFR amplification and lack of activating mutations in metaplastic breast carcinomas

{P} JS Reis Filho[1], C Pinheiro[3], MBK Lambros[1], F Milanezi[2], S Carvalho[2], K Savage[1], PT Simpson[4], C Jones[5], S Swift[1], A Mackay[1], RM Reis[3], JL Hornick[6], EM Pereira[7], F Baltazar[3], CDM Fletcher[6], A Ashworth[1], SR Lakhani[4], FC Schmitt[2]
[1]. The Breakthrough Breast Cancer Research Centre, Institute of Cancer Research, London, United Kingdom, [2]. IPATIMUP – Institute of Molecular Pathology and Immunology of Porto University and Medical Faculty of Porto University, Porto, Portugal, [3]. Life and Health Sciences Research Institute (ICVS), School of Health Sciences, University of Minho, Braga, Portugal, [4]. Molecular & Cellular Pathology, Mayne Medical School, University of Queensland, Brisbane, Australia, [5]. Section of Paediatric Oncology, Institute of Cancer Research, Sutton, United Kingdom, [6]. Department of Pathology, Brigham and Women's Hospital, Boston, United States, [7]. Laboratório Salomão & Zoppi, Sao Paulo, Brazil

Metaplastic breast carcinomas (MBCs) are reported to harbour epidermal growth factor receptor overexpression (EGFR) in ~80% of the cases, however EGFR gene amplification is the underlying genetic mechanism in around one third of these. 47 MBCs were analysed using immunohistochemistry with antibodies against EGFR, chromogenic *in situ* hybridisation with probes for EGFR gene, microarray comparative genomic hybridisation (aCGH) analysis and direct gene sequencing of exons 18-21 to define i) the prevalence of EGFR ovexpression and amplification, ii) to provide a detailed mapping of EGFR amplicon (7p11.2), and iii) to investigate whether EGFR activating mutations would be an alternative genetic mechanism for EGFR overexpression in MBCs. 32 cases showed EGFR overexpression and of these 34% harboured EGFR gene amplification. EGFR amplification showed a statistically significant association with EGFR overexpression and was restricted to carcinomas with homologous metaplasia. The minimal region of amplification on 7p11.2 as defined by aCGH encompassed 3 genes: EGFR, LANCL2 and SEC61G. No EGFR activating mutations were identified. Our results confirm that MBCs frequently harbour EGFR overexpression, which is driven by gene amplification in ~33% of the cases. EGFR activating mutations are unlikely to be an alternative genetic mechanism for EGFR overexpression in MBCs.

O24

Comprehensive molecular genetic analysis reveals FGFR1 as a potential therapeutic target for lobular breast carcinomas

{P} JS Reis FIlho[1], PT Simpson[3], N Turner[1], MBK Lambros[1], C Jones[4], A Mackay[1], A Grigoriadis[1], D Sarrio[5], K Savage[1], T Dexter[1], M Iravani[1], K Fenwick[1], B Weber[6], D Hardisson[7], FC Schmitt[2], J Palacios[5], SR Lakhani[3], A Ashworth[1]
[1]. The Breakthrough Breast Cancer Research Centre, Institute of Cancer Research, London, United Kingdom, [2]. IPATIMUP – Institute of Molecular Pathology and Immunology, University of Porto, Porto, Portugal, [3]. Molecular & Cellular Pathology, Mayne Medical School, University of Queensland, Brisbane, Australia, [4]. Section of Paediatric Oncology, Institute of Cancer Research, Sutton, United Kingdom, [5]. Centro Nacional de Investigaciones Oncológicas, Madrid, Spain, [6]. University of Pennsylvania, Philadelphia, PA, United States, [7]. Department of Pathology, La Paz Hospital, Madrid, Spain

Classic lobular carcinomas (CLCs) account for 10% of all breast cancers. At the genetic level, CLCs show recurrent physical loss of chromosome16q coupled with lack of E-cadherin (CDH1) expression. We subjected thirteen cases of CLC to a comprehensive molecular genetic analysis including immunohistochemistry for E-cadherin, oestrogen (ER) and progesterone receptors, HER2 and p53; high-resolution comparative genomic hybridisation (HR-CGH); microarray-based CGH (aCGH); and in situ hybridisation (ISH) for CCND1 and FGFR1. All cases lacked expression of CDH1, p53 and HER2 and all but one case was positive for ER. HR-CGH revealed recurrent gains on 1q and losses on 16q (both, 85%). aCGH showed a good agreement with but higher resolution and sensitivity than HR-CGH. Gains at 11q13 and 8p12-p11.2 were identified in 7 and 6 cases, respectively, and were validated with ISH. aCGH and gene expression of profile data analyses of the cell lines MDA-MB-134 and ZR-75-1, which harbour distinct 8p12-p11.2 gains, identified FGFR1 as a putative driver of this amplicon in MDA-MB-134. Inhibition of FGFR1 using siRNA or a small molecule inhibitor demonstrated that FGFR1 signalling contributes to the survival of MDA-MB-134 cells. Our findings suggest that receptor FGFR1 inhibitors may be useful as therapeutics in a subset of CLCs.

O25

BRCA1 gene promoter methylation in metaplastic breast carcinomas

{P} JS Reis Filho[1], N Turner[1], FC Schmitt[2], A Tutt[1], A Ashworth[1]
[1]. The Breakthrough Breast Cancer Research Centre, Institute of Cancer Research, London, United Kingdom, [2]. IPATIMUP – Institute of Molecular Pathology and Immunology, University of Porto, Porto, Portugal

Breast carcinomas arising in BRCA1 germline mutation carriers frequently harbour a basal-like phenotype. However, somatic mutations of BRCA1 gene have proven to be remarkably rare in sporadic breast carcinomas. Metaplastic carcinomas of the breast (MBCs), tumours characterised by an admixture of adenocarcinoma with metaplastic elements, harbour a basal-like immunophenotype in approx 90% of the cases. Given the association between basal-like phenotype and BRCA1 pathway inactivation, and the lack of BRCA1 somatic mutations in sporadic breast cancer, we sought to investigate whether BRCA1 gene would be epigenetically inactivated in MBCs. DNA was extracted from representative formalin-fixed, paraffin embedded sections of 27 MBCs. The methylation status of the BRCA1 promoter was assessed by methylation specific PCR (MSP) and results confirmed by sequencing the BRCA1 promoter on bisulfite treated DNA. MSP revealed a high incidence of BRCA1 methylation in metaplastic cancers (63%, 17/27; 95%CI 42%-81%), which was confirmed by bisulfite sequencing in 16/18 samples. Our results suggest that epigenetic inactivation of BRCA1 is likely to play an important role in the biology of MBCs. These findings may prove useful in defining more effective chemotherapy regimens for these chemo-resistant neoplasms.

O26

Cyclin D1 protein overexpression and gene amplification in breast carcinomas: an immunohistochemical and CISH analysis

{P} JS Reis Filho, K Savage, MBK Lambros, M James, D Steele, RL Jones, M Dowsett
The Breakthrough Breast Cancer Research Centre, Institute of Cancer Research, London, United Kingdom

Conflicting results on the prevalence of cyclin D1 (CCND1) overexpression and its correlation with *CCND1* amplification and outcome of breast cancer patients have been reported. Due to limited sensitivity and specificity of most antibodies against CCND1, evaluation of CCND1 immuno-expression is reported to be problematic. We analysed CCND1 expression and amplification and their correlations with clinicopathlogical features and outcome in a cohort of 245 breast cancer patients treated with anthracycline-based adjuvant chemotherapy. Immunohistochemistry for CCND1 was performed using the SP4 rabbit monoclonal antibody; results were scored according to the Allred scoring system. Chromogenic *in situ* hybridisation was carried out using the *CCND1* SpotLight probe. CCND1 expression and amplification were found in 67.4% and 14.5% of the cases, respectively. CCND1 overexpression was strongly correlated with gene amplification and positivity for hormone receptor expression. An inverse correlation was observed between an immunohistochemical panel of 'basal-like' markers and both CCND1 overexpression and amplification. On univariate analysis CCND1 expression showed a correlation with longer overall survival. Neither CCND1 overexpression nor amplification were independent prognostic factors for disease-free or overall survival. Our results confirm the association between CCND1 overexpression and positivity for hormone receptors and the lack of *CCND1* amplification in basal-like breast carcinomas.

O27

Bone Marrow Contributes to Podocyte Regeneration and Amelioration of Renal Disease in Alport's Syndrome

{P} E I Prodromidi[1], R Poulsom[2], R Jeffery[2], C A Roufosse[3], T Hunt[2], C D Pusey[1], H T Cook[3]
[1]. Renal Section, Imperial College London, Hammersmith Campus, London, United Kingdom, [2]. Histopathology Unit, Cancer Research UK, London Research Institute, London, United Kingdom, [3]. Department of Histopathology, Imperial College London, Hammersmith Campus, London, United Kingdom

In a model of autosomally inherited Alport's syndrome, mice that lack the α3 chain of collagen IV *(Col4α3$^{-/-}$)*, develop progressive glomerular damage leading to renal failure. The proposed mechanism is that podocytes fail to synthesise normal glomerular basement membrane (GBM), so the collagen IV network is unstable and easily degraded. We used this model to study whether bone marrow (BM) transplantation can rectify this podocyte defect by correcting the deficiency in Col4α3. Female C57BL/6 *Col4α3$^{-/-}$* (-/-) mice were transplanted with whole BM from male wild type (+/+) mice. Control female -/- mice received BM from male -/- littermates. Renal tissues were examined 20 weeks post-transplant. Serum urea and creatinine levels were significantly lower ($p<0.05$) in recipients of +/+ BM compared with those of -/- BM. Glomerular scarring and interstitial fibrosis, as determined by PAS histology and Picro-Sirius Red staining respectively, were also significantly decreased ($p<0.05$). Donor-derived cells were detected by *in situ* hybridisation (ISH) for the Y chromosome, and fluorescence and confocal microscopy indicated that some showed an apparent podocyte phenotype in mice transplanted with +/+ BM. Glomeruli of these mice showed small foci of staining for α3(IV) protein by immunofluorescence. α3(IV) mRNA was detectable by RT-PCR and ISH in some mice transplanted with +/+ BM but not -/- BM. Our data show that improved renal function in *Col4α3$^{-/-}$* mice results from BM transplantation from +/+ donors and the mechanism by which this occurs may in part involve generation of podocytes in which the gene defect has been corrected.

O28

TOP2A Amplification in Prostate Cancer is associated with *HER-2* Amplification, Androgen Resistance & Decreased Survival

{P} AJ Murphy[2], CA Hughes[2], C Barrett[1], H Magee[1], B Loftus[1], J O'Leary[2], O Sheils[2]
[1]. Adelaide and Meath Hospital, Tallaght, Dublin, Ireland, [2]. Trinity College, Dublin, Ireland

Background: *TOP2A* encodes topoisomerase IIα (TopoIIα), an enzyme involved in DNA replication. *HER-2*, located on the same amplicon as *TOP2A*, has been reported to be co-amplified with *TOP2A* in breast and bladder cancer. Studies of prostate cancer (PCa) have shown overexpression of TopoIIα and Her-2 in high grade androgen resistant tumours. Both genes have not previously been compared in the same tumours in one study.
Design: Using tissue microarrays, we carried out immunohistochemistry and FISH for *HER-2* and *TOP2A* in 100 58 PCa's (localized and advanced) and 42 BPH cases. Her-2 staining was scored from 0 to 3+. Percentage nuclei staining for TopoIIα was recorded with overexpression defined as ≥ 5% tumor cells staining. Amplification was defined as a ratio of target gene signals to centromere 17 signals of greater than 1.5.
Results: 31% of 58 advanced PCa's showed TopoIIα overexpression; 16% overexpressed Her-2. 75% of Her-2 positive cases showed concurrent TopoIIα overexpression. 26% of advanced cancers showed *TOP2A* amplification; 13% showed *HER-2* amplification; four of these (66%) showed *TOP2A* co-amplification. No localized BPH or PCa case showed *HER-2* amplification, TopoIIα overexpression or *TOP2A* amplification. Amplification of either gene was associated with high tumour stage, Gleason score and androgen resistence. Using multivariate analysis, *TOP2A* amplification was related to survival.
Conclusion: TopoIIα expression in advanced PCa is usually due to *TOP2A* amplification, which may occur with or without *HER-2* amplification. *TOP2A* amplification is associated with reduced survival in advanced PCa. Therapies directed against TopoIIα (and possibly Her-2) in such patients may improve survival.

O29

Urine Cytology Screening for Polyoma Virus Nephropathy following Renal Transplantation: Review of a Working Service

TP Thamboo, K Jeffery, P Friend, {P} ISD Roberts
Oxford Radcliffe Hospitals, Oxford, United Kingdom

Polyoma virus nephropathy (PVN) associated with graft dysfunction has a high incidence of irreversible injury and graft failure. Here we review the first year of a monthly urine cytology screening service, introduced on our unit to identify patients at risk of PVN, at an early, potentially treatable, stage.
Urine samples (n=392) were received from 97 of 108 transplant recipients in 2005. Of 56 patients with follow-up >6 months, 20% and 9% had significant (>10 decoy cells/cytospin) and non-significant positive cytology respectively. In this group, the first positive urine samples occurred at a median of 3 months post-transplantation and patients with significantly positive samples had higher 3 and 6-month serum creatinine levels than patients with negative urine cytology ($p<0.01$). Three patients with a significant positive urine cytology had a subsequent positive plasma BK virus PCR, performed according to protocol following 2 consecutive months of significant viruria. Three of 97 patients had biopsy-proven PVN, all in the third month, 1-6 weeks after first positive urine samples.
We conclude that significant PV viruria is common following renal transplantation, with onset usually within the first 3 months. Viruria is associated with worse graft function at 3 and 6 months. The time between urine positivity and clinical PVN is short. More frequent early urine screening and more rapid investigation and intervention is required to achieve clinical benefit.

O30

Assessment of the Cardiff nephrectomy cut-up protocol with total blocking of the renal sinus: Impact on Tumour Staging and Practical Issues.

{P} EJ Soilleux, ISD Roberts
Department of Histopathology, John Radcliffe Hospital, Oxford, Oxfordshire, United Kingdom

Objective: To evaluate the effects on the detection of vascular invasion and workload following introduction of a new standard dissection protocol for examining nephrectomy specimens for renal cell carcinoma (RCC), which was devised to identify more effectively the major prognostic determinants.

Method: The incidence of vascular invasion and the number of blocks of tissue taken per tumour were compared before and after introduction of the new protocol, using 192 consecutive nephrectomy specimens for RCC submitted to our unit.

Results: The Cardiff protocol increased the percentage of tumours staged as T3b (renal sinus or hilar vein invasion) from 37.7% to 55.7% cases ($p <0.001$) with an increase from 9.1% to 21.7% staged as T3b due to renal sinus vein invasion alone ($p <0.01$). There was a small, but significant, permanent increase in workload, from an average of 11.7 to 13.4 blocks per case ($p <0.001$).

Conclusions: This protocol is suitable for use in routine practice to evaluate pathological prognostic determinants important for clinical management, while causing only a small increase in workload.

O31

YY1 Expression Predicts Survival in Follicular and Diffuse Large B-cell Lymphoma

E Sakhinia[1], C Glennie[1], JA Hoyland[1], L Menasce[2], JA Radford[2], {P} RJ Byers[1]
[1]. University of Manchester, Manchester, United Kingdom, [2]. Christie Hospital, Manchester, United Kingdom

Recent gene expression profiling has identified gene signatures predictive of outcome, *Indicator* genes, for diffuse large B-cell lymphoma (DLBCL) and follicular lymphoma (FL). However, measurement of *Indicator* genes in routine practice remains difficult.

We applied real-time PCR, to polyA cDNAs prepared from 122 archived human frozen lymph nodes. Reverse transcription and polyA RT-PCR was performed on extracted total RNA and resultant cDNA probed for 36 candidate *Indicator* genes, by real-time PCR with quantification against human DNA, and the normalisation to the mean of four housekeeping genes.

Ten genes showed statistically significant different expression between FL and DLBCL, including Cyclin B, COL3A1, NPM3, H731, PKC.B1, OVGL, ZFPC150, HLA-DQ-a, and XPB. Of these, cyclin b, a cell cycle gene, NPM3, nucleolar phosphoprotein, and COL3A1were higher in DLBCL. Six genes showed statistically significant higher expression in the neoplastic nodes compared to reactive nodes, namely PKCB-1, BCL-6, EAR2, ZFX, Cyclin B, YY.1. High levels of YY.1 were associated with a shorter survival interval in both FL and DLBCL by Kaplan-meir survival analysis.

The method is simple, sensitive and robust, allowing translation into routine use and may be used as a platform for clinical measurement of prognostic gene signatures.

O32

Different Expression Pattern of Vascular Endothelial Growth Factor-A (VEGF-A) in Diffuse Large B-Cell Lymphoma (DLBCL) and Hodgkin's Lymphoma (HL)

{P} L A Sanders[1], C Burton[2], S Chalasani[1], G Frantz[1], A Jack[2], H Koeppen[1]
[1]. Department of Pathology, Genentech Inc., South San Francisco, CA, United States, [2]. Haematological Malignancy Diagnostic Service, Leeds General Infirmary, Leeds, United Kingdom

VEGF and microvascular density (MVD) are indicators of poor prognosis for many solid tumours; this observation is less well established for malignant lymphomas. Malignant and immune cells are potential sources of endothelial mitogens, including VEGF. We determined VEGF expression pattern and degree of vascularity in DLBCL and HL.
Expression of VEGF-A protein and RNA in formalin fixed, paraffin embedded tissues of DLBCL (n = 10 germinal center, n = 10 activated B-cell) and HL (n = 17 nodular sclerosis) was performed by immunohistochemistry (IHC) and in-situ hybridization (ISH). Vascularity was determined by IHC staining for VEGF-receptor 2.
In DLBCL, VEGF was seen in rare vessels and activated macrophages in all cases, regardless of subtype. In HL, VEGF was observed in vessels and stroma in all cases, and in Reed-Sternberg cells in 6/17 cases. VEGF-receptor 2 expression was limited to vascular cells in all cases (DLBCL and HL). HL tissue showed greater vascularity than DLBCL.
Lack of VEGF expression in DLBCL, regardless of subtype, suggests absence of prognostic relevance in this malignancy. While VEGF was expressed by Reed-Sternberg cells in cases of HL the majority of VEGF was produced by associated reactive stromal or inflammatory cells. Our results emphasize the role of stroma as a source of VEGF that should be investigated further in haematological malignancies.

O33

A Novel Picropolychrome Technique for the Selective Staining of Basement Membranes

{P} K Asaad, DT Sharpe, S Huq, S Stevenson, A Adekunle, IL Naylor
University of Bradford, Bradford, United Kingdom

We present a new histological technique that may be helpful to selectively stain cytoplasm, nuclei, collagen and basement membranes. Based on Herovici's technique, an established, easily reproducible picropolychrome method that selectively stains cytoplasm, nuclei and collagen.

Normal samples of human skin (17) and rat kidney (4) were formalin-fixed and wax-embedded. Solutions and staining protocol were as for Herovici's technique except that sirius red replaced acid fuschin. As a control the Sirius red and acid fuschin were omitted. Samples also underwent PAS staining for comparison.

The PAS technique resulted in dark pink staining of basement membranes with pale pink staining of other structures. Our technique resulted in dark brown staining of nuclei, greenish cytoplasm and collagen fibres showed as thin blue and thick green fibres. Basement membranes were purple in 19 of the 21 samples. Negative controls showed nuclear and cytoplasmic staining as before, with green and blue staining collagen fibres and no differential staining of the basement membrane.

Replacing acid fuschin with sirius red in Herovici's technique alters the pattern of collagen fibre staining. In addition it allows differential staining of basement membrane. The most likely explanation is that the Sirius red binds preferentially to type IV collagen in the presence of methyl blue and picric acid.

O34

Molecular genetic analysis suggests a developmental origin for malignant myoepithelial carcinomas of the salivary gland from pleomorphic adenomas

{P} MB Lambros[1], D Tan[1], E Arriola[2], K Fenwick[1], N Tamber[1], A Mackay[1], K Savage[1], A Ashworth[1], S Di Palma[3], JS Reis Filho[1]
[1]. The Breakthrough Breast Cancer Research Centre, Institute of Cancer Research, London, United Kingdom, [2]. Academic Department of Biochemistry and Breast Unit, Royal Marsden Hospital, London, United Kingdom, [3]. Royal Surrey County Hospital, University of Surrey, Guildford, United Kingdom

The origin of salivary gland myoepithelial carcinomas (MCs) is unclear. There is circumstantial evidence to suggest that ~50% of MCs arise from their benign counterparts (ie, pleomorphic adenomas (PAs)/ benign myoepitheliomas). We present a case of a patient with a parotid tumour composed of 2 juxtaposed but distinct nodules, comprising a MC and a PA. Tiling-path array comparative genomic hybridisation (TP-aCGH) with ~32k bacterial artificial chromosome clones was performed to identify genetic changes in both components and determine whether there was evidence for genetic evolution from the PA to MC components. TP-aCGH revealed regions with unbalanced genomic changes common to both lesions, including gains in 2p14-15, 7p15.1, 7p21.1, 9p22.3, 11q21, 13q14.2-14.3, 13q32.2, 18q21.32, and 18q23, and loss of Xq26.2. Distinct and more frequent copy number changes were found in the MCC than the PA, including amplification of 8p11 (FGFR1), 8q23.3 (TPRS1), 8q24 (MYC), 9q23 (NFIB), 10p12.1 (ABI1), 10q26.12-q26.3 (FGFR2). Gains of 8p11 (FGFR1) and 8q24 (MYC) were validated by chromogenic in-situ hybridisation. These results suggest that at least some MCs of the salivary glands may originate from associated benign lesions. Furthermore, disease progression from PA to MC appears to be mirrored at the genetic level by amplification of key oncogenes.

O35

Implementation of the 2005 Coroner's Rules Amendments – A survey of practice in England and Wales

{P} R J Delaney, I S D Roberts
John Radcliffe Hospital, Oxford, United Kingdom

On June 1st 2005 amendments to the Coroners Rules 1984 were introduced in England & Wales. These principally cover the retention of tissues from autopsies and their subsequent disposal. This study assesses regional variations in the interpretation of the amendments and their impact on our local autopsy practice.

Questionnaires were circulated to pathologists in 120 Coronial jurisdictions, addressing conditions under which histological material could be retained. A local review of autopsy practice was conducted before and after the introduction of the amendments. Questionnaires were returned from 71 Coronial jurisdictions.

Thirty-five Coroners (49%) have provided written guidelines on their interpretation of the amendments. In 52 jurisdictions (73%), pathologists are authorised to retain material to confirm/refine causes of death from natural causes. In 77% of jurisdictions, Coroner's Officers are responsible for obtaining instructions from next of kin on subsequent retention, use or disposal of retained tissues. In our department there has been a reduction in the proportion of cases in which histology is taken but an increase in the proportion of cases in which a histology report is issued.

This study demonstrates considerable regional variation in interpretation of the 2005 Coroner's Rules Amendments. These variations have potentially important implications for clinical practice.

O36

Immunohistochemical Nuclear Positivity For WT1 In Acute Myeloid Leukaemia

{P} M AL-ADNANI, S WILLIAMS, J ANDERSON, M ASHWORTH, M MALONE, NJ SEBIRE
Great Ormond Street Hospital for Children, London, United Kingdom

Background: Several studies have previously reported that acute myeloid leukeamia (AML) may express WT1 detected by RT-PCR and /or northern blotting. The diagnostic utility of WT1 expression in AML using immunohistochemistry has not previously been reported.

Methods: Paraffin-embedded tissue sections from 55 AML, 12 ALL and 10 normal bone marrow specimens were immunostained for WT1 (anti-N terminus antibody).

Results: 22/55 AML cases (40%) demonstrated nuclear immunopositivity for WT1, including 20/47 bone marrow trephines and 2/4 granulocytic sarcomas. All the ALL and normal bone marrow specimens were negative.

Conclusion: A significant proportion of AML express nuclear immunostaining for WT1, a finding that has only been described previously in Wilms' tumour and desmoplastic small round cell tumour. This finding is important for the correct interpretation of immunohistochemical findings in the diagnosis of 'small round cell' tumours, especially in cases of extramedullary deposits of AML, in which traditional myeloid markers may be negative.

O37

National Histopathology Training Schools (NHTS) – Recruitment Experiences, Issues and Challenges!

S Cossins, **{P}** A McGregor, K West, J Appleyard, D Bailey, J Brinklow, T Collins, PJ Gallagher, C Gore, NR Griffin, S Hill, R Liebmann, M Young

[1]. University Hospitals of Leicester NHS Trust, Leicester, United Kingdom, [2]. Leeds Teaching Hospitals NHS Trust, Leeds, United Kingdom

As the Histopathology Training Schools in England have expanded in number/size, recruitment has become increasingly complex. Questions have been asked as to whether the criteria for selection are fit for purpose.

Using structured scoring systems allows a correlation of scores at each stage. We were able to determine which questions added most value and eliminate or change redundant questions. The application form contains mostly "hard" questions and robust scoring allows a fair, reproducible comparison between short-listed and appointed applicants. The interview requires a more dynamic interaction with flexible questioning.

With more than 700 applicants to approximately 100 posts, the marking of applications requires a large cohort (24) of individuals. The total number of forms is divided into 6 - each being scored by 4 individuals, from different centres. Because the questions are relatively firm with predictable answers, consistency of scoring is maintained. The interview covers less firm areas and requires a smaller panel of individuals (3 sub-panels of 4 interviewers) who score every candidate. Expert Human Resources advice suggests that the approach used is optimal for a recruitment process of this size.

Ongoing review and comparison with results from previous years allows continual refinement and improvement to minimise the risk of reducing standards.

O38

Routine examination of multiple levels from gastro-intestinal biopsies: a resource management approach

C Chambers[2], A McGregor[1], **{P}** KP West[1]

[1]. Dept of Histopathology, University Hospitals of Leicester, Leicester, United Kingdom, [2]. Leicester Medical School, University of Leicester, Leicester, United Kingdom

Examination of three or more levels from gastrointestinal biopsies is generally considered best practice. However, there is little published evidence to support this. This study assessed the diagnostic information that could be obtained from examination of a single deep section compared to the examination of three levels. Fifty gastro-oesophageal, 50 duodenal and 50 colorectal biopsies were studied. The third level from each case was reviewed by two gastrointestinal histopathologists and the diagnoses were compared with the original diagnosis made after examining three levels. Diagnostic discrepancies were graded 1 = not significant, 2 = potentially significant, 3 = probably significant.

Duodenal biopsies – 49/50 no discrepancies, 1/50 grade 1 discrepancy

Gastro-oesophageal biopsies – 41/50 no discrepancies, 9/50 grade 1 discrepancies.

Colorectal biopsies – 41/50 no discrepancies, 9/50 grade 1 discrepancies.

Discrepancies most often concerned normal versus mild inflammation and could not be attributed to non-examination of levels.

We estimate that approximately 15% of biopsies would still require levels (eg dysplasia surveillance) but cutting one deep level instead of three standard levels from the remainder would save approximately 50 technical working days per year.

In a cash-constrained service this approach deserves further consideration.

O39

Positive Predictive Value Of Cervical Smears Reported As Mild Dyskaryosis

{P} Bijal Shah, Janet Marshall, Durgesh Rana, Mina Desai

Manchester Royal Infirmary, Manchester, Lancashire, United Kingdom

Recent national guidelines (NHSCSP Document 20) recommend the referral of patients after one test showing mild dyskaryosis. We evaluated the usefulness of this guideline and determined the positive predictive value (PPV) of conventional smears (CS) and ThinPrep samples (TP) reported as the first occurrence of mild dyskaryosis from January '05 to June '05.

There were 1016 cases, which were reported as mild dyskaryosis. Out of them, 51.1% (519 cases) were first report of mild dyskaryosis: 61.8% (321 cases) and 38.2% (198 cases) were CS and TP respectively. Of these, 181 CS (56.4%) and 120 TP (60.6%) had histological follow up. The results showed that 54.1% CS and 56.7% TP had a low-grade outcome, 26.0% CS and 25.8% TP had a high-grade outcome and 19.9% CS and 17.5% TP had a normal outcome.

The PPV of mild dyskaryosis for CIN1 or worse result was 53.0% and 50.0% in CS and TP respectively. The PPV of mild dyskaryosis for CIN1 only was 27.1% and 24.2% in CS and TP respectively. This difference is statistically insignificant. This result endorses usefulness of colposcopic referral, however, this guideline should not alter the management of women enrolled in HPV studies to improve understanding of management of mild cytological abnormalities.

O40

Direct Observation of Practical Skills in Early Histopathology Training: A Pilot Study of Competency Assessment

{P} KP West

Dept of Histopathology, University Hospitals of Leicester, Leicester, United Kingdom

Histopathology training is very closely supervised in comparison to many specialties but in the past there has been little formal assessment of competence. In the future, under the aegis of the Postgraduate Medical Education and Training Board (PMETB), there will have to be documented evidence of the acquisition of multiple competences during training.

This study used a structured form to assess nine aspects of surgical cut-up including cleanliness, safety, communication and block selection. An overall rating for the procedure was then given. Each item was assessed on a scale of 1-6 where 4 was satisfactory for a year 1 trainee and 3 was borderline. Trainees and assessors also gave a score out of 10 indicating their level of satisfaction with the assessment.

Upto six assessments were conducted for five trainees. The mean overall scores for trainees ranged from 4.2 - 5.0. The only score of 3 given was the result of non-disposal of sharps. Mean trainee and assessor scores for satisfaction with the assessments were 8.9 and 8.6 respectively.

This small pilot study suggests that the direct observation of practical skills can be used to assess competence in surgical cut-up. Its applicability to other aspects of histopathology/autopsy should be explored.

Abstracts

Speakers

S1

New Strategies in Cervical Screening

{P} Henry C Kitchener
University of Manchester, Academic Unit of Obstetrics & Gynaecology, Manchester, United Kingdom

Cervical screening has been one of the successes of modern preventative medicine. It has saved hundreds of thousands of lives worldwide, including at least 1,000 lives/year in the United Kingdom. Since the introduction of the NHS Cervical Screening Programme in 1988, deaths from cervical cancer in the United Kingdom have been halved and fewer than 1,000 women per year die from this disease.

The essential role of human papillomavirus in cervical carcinogenesis has changed the way we think about the disease. It can now be regarded epidemiologically as a rare complication of a common infection which as two key implications. The first of these is that HPV testing can be clinically valuable and prophylactic HPV vaccination could move control from a secondary prevention to a primary prevention.

Other technological advances in cytology itself are changing practice. Liquid based is replacing conventional as a more convenient, efficient means of undertaking cytological testing. This also presents the means for undertaking HPV testing should it be required. Automation in cytology is becoming feasible and its potential is currently being evaluated in comparison with 'manually read' cytology.

Cervical screening will change in the future in two respects:
Cytology will become combined with HPV testing to achieve more effective screening, triage and follow-up strategies.
Prophylactic vaccination will eventually result in a major reduction in the incidence of cervical neoplasia and major adjustments will be required in the use of cytology and HPV testing in secondary prevention for those not protected by vaccination.

S2

Molecular markers in cytology – scope for the future

{P} J.J. O'Leary
Trinity College Dublin, Ireland, Ireland

Cervical cancer, a potentially preventable disease, remains the second most common malignancy in women worldwide. Current screening protocols rely on the PAP smear test, which has a reported false negative rate of 15-50%. Advances in automation of cervical cytology have resulted in improved cell preservation techniques and overall high quality cellular material, suitable for molecular analysis. The current focus has been primarily on the use of molecular biomarkers as adjuncts to existing screening procedures.
These biomarkers include human papillomavirus and host cell regulatory molecules, including minichromosome maintenance proteins, Cdc6, a cell division cycle protein, cyclins, BIRC5 and p16(INK4A) tumour suppressor protein. Developments in microarray technologies and their application to the study of cervical cancer have greatly expanded the list of differentially regulated genes and proteins known to be involved in cervical cancer.
The recent discovery of micro RNAs may offer additional insights into the complex pathobiology of cervical pre-cancer and cancer. Combined microarray and miRNA analyses will help to unravel the pathogenesis of HPV infection and dysplastic progression and ultimately improve treatment of CIN and cervical cancer.

S3

Our shape comes in extracellular modules. Their role in biology and pathology.

{P} JE Scott
Manchester University, Manchester, United Kingdom

Animal shapes are maintained by extracellular matrices (ECMs) of connective tissues. ECM shapes depend on keeping collagen fibrils in the right places, held by frequent and specifically located proteoglycan (PG) bridges, which are present even in remote animals, e.g. echinoderms. The PGs carry anionic glycosaminoglycan (AGAG) strings that span the interfibrillar spaces. These strings are aggregated antiparallel chains of dermochondan or keratan sulphates, stabilised by hydrophobic and hydrogen bonds Their length equals the distance between the fibrils. I called these structures 'shape modules' since they repeat regularly and hold together varied shapes of ECMs.
Shape module bridges are elastic because their specific AGAG/AGAG interactions break under stress and reform when the stress is removed, and/or they contain an elastic sugar, L-iduronate. Direct proof was obtained by stretching individual AGAG molecules.
This model was tested against biomechanical data. Cartilages are built of shape modules, which explains their anisotropic responses (along and at right angles to shape module axes) to compressive and tensile stresses. Degradation of shape modules in osteoarthrosis distorts these responses. Thus, inability to bind together collagen fibrils results in imbibition of water, fissuring etc. as fibrils drift apart under the swelling pressure of aggrecan and decoran.

S4

The connective tissue stem cell: lineage, plasticity and function

{P} JA Hoyland
Manchester University, Manchester, United Kingdom

Connective tissue stem cells are adult mesenchymal stem cells (MSCs) that usually reside in the stromal compartment of bone marrow. Under appropriate stimuli these multipotent cells exhibit cellular plasticity having the capacity to differentiate along osteogenic, chondrogenic, adipogenic and myogenic lineages. Recent evidence has shown that these cells differentiate along the neurogenic pathway and can also give rise to an endothelial cell phenotype. The molecular basis for mesenchymal stem cell plasticity is not fully understood but cellular senescence has been shown to affect differentiation potential and will be discussed.
Other sources of adult stem cells with mesenchymal potential have been identified including periosteum, trabecular bone, adipose tissue, skeletal muscle, cartilage/synovium and intervertebral disc raising new questions about their role and local differentiation cues. Stem cells in adult tissue could serve as reservoirs of reparative cells ready to mobilise and differentiate in response to certain stimuli or disease. However in some diseases the inability to repair may be due to the fact that stem cell populations are depleted or functionally altered. For example, evidence suggests that osteoblast dysfunction in Osteoporosis may be due to an imbalance in mesenchymal stem cell differentiation/plasticity leading to decreased osteogenesis and increased adipogenesis. Data to show that MSCs from patients with Osteoporosis are functionally altered favouring adipogenesis will be presented

S5

Difference of skin malignancy between Japan and UK

{P} H Sasano, Y Kariya
Tohoku University, Sendai, Miyagi, Japan

The incidence of breast, colon and prostate cancer has markedly increased in Japan for the last 20 years which is generally considered to be due to "Westernization" of life style of Japanese population. The incidence of both melanoma and non-melanoma skin cancers has increased in the Western countries possibly because of aging, increased outdoor activities and ozone depletion which increases the amount of harmful solar ultraviolet radiation. It is therefore interesting to note whether these increments detected in these cancers above could be also detected in skin malignancies among Japanese. In Japan, the incidence of melanoma and squamous cell carcinoma has not necessarily increased but their incidences have been demonstrated to be clearly correlated with the amounts of sun light exposure because the incidence of Okinawa, southernmost islands of Japan is highest in both melanoma and non-melanoma skin malignancies. However, the incidence of actinic keratosis, precursor lesions of squamous cell carcinoma and basal cell carcinoma has increased all over Japan, which may be also due to increased outdoor activities and aging of Japanese population as in the Western countries .

S6

Pyothorax-associated Lymphoma-A Distincive Type of Lymphoma which is common in Japan but rare in Western countries

{P} Katsuyuki Aozasa, Shin-ichi Nakatsuka, Tetsuya Takakuwa
Dept Pathol, Osaka Univ Graduate Sch Med, Suita, Osaka, Japan

Pythorax-associated lymphoma(PAL) is non-Hodgkin's lymphoma that develops from chronic pyothorax resulted from artificial pneumothorax for the treatment of lung tuberculosis or tuberculous pleuritis. This disease was originally described by Dr Aozasa as a distinctive clinicopathologic entity in 1987, and now listed as the disease entity in the WHO classification of Tumors, Pathology Genetics, Tumors of the lung , Pleura, Thymus and Heart(2004).Histologically PAL usually shows a diffuse proliferation of large cells of B-cell type, diffuse large B-cell lymphoma(DLBL). A Gene expression profile of PAL is distinct from nodal DLBL in its higher expression level of interferon-inducible genes. PAL is strongly associated with Epstein-Barr virus (EBV) infection with expression of EBV latent genes of EBNA-2 and/or LMP-1 together with EBNA-1. PAL is distinct entity both in its clinicopathologic presentation as well as its gene expression profile.

S7

OSPE Assessment and Year 1 Run-through Training

{P} AC Bateman
Southampton University Hospitals NHS Trust, Southampton, United Kingdom

The inception of run-through training in histopathology has driven the development of a new gateway mechanism towards the end of Year 1, with successful trainees gaining a National Training Number and progressing to Year 2. This will be achieved with the Deanery-supported End-of-Year Review (EYR), of which the new Royal College of Pathologists' (RCPath) Year 1 Assessment (Y1A), in the form of an Objective Structured Practical Examination (OSPE), forms an important component. OSCE (Objective Structured Clinical Assessment) assessment is widely used in undergraduate medical schools and was trialled for Year 1 histopathology trainees in recent years, but formally administered by the RCPath only from 2006. The Y1A is designed to test skills and applied knowledge and is mapped to the new RCPath Histopathology Curriculum using a blueprint. The Y1A is currently held at month eight of Year 1 (with opportunity for a resit in month 10) and comprises 15 stations (two of which are rest stations) at each of which the candidates will spend 12 minutes (i.e. total time three hours). The questions are selected from a bank created by the RCPath Year 1 Assessment Panel and will usually include at least one station with a double headed microscope and one testing communication skills – each with an external examiner. All questions are marked using a structured markscheme and all except those with external examiners are double marked. The pass mark for each question and therefore for the whole Y1A is determined using the borderline group method. Success at the Y1A will be a critical requirement for progression to Year 2 – but progression is also dependent upon the other elements of the EYR including the Educational Supervisor's report, the RCPath Teaching and Learning Record and a multi-source feedback tool.

S8

MRCPath in Histopathology Part 1: Why the Change

{P} EW Benbow
University of Manchester, Manchester, United Kingdom

Professional examinations need to be fit for purpose. They need to be valid, testing the knowledge the candidates will need in their future working lives. They need to be reproducible, so that results are consistent, and discriminatory, so that good candidates pass and poor candidates fail. They need to test the capacity to think and synthesise, rather than merely regurgitate memorised facts. Examinations drive learning, so great breadth of coverage is needed. Most of all, they must be fair. These are clearly very desirable characteristics, but traditional examinations fail on most or all of these counts. And now, not only must we develop examinations with these characteristics; we need to be able to demonstrate to the Postgraduate Medical Examination and Training Board (PMETB) that we've done so.
Essays papers can be written to have many of these desired characteristics, but it is difficult to do, and near-impossible to demonstrate. Multiple choice question (MCQ) papers can be written that have few of these characteristics, if done badly. A team of experienced, trained examiners writes the new MRCPath in Histopathology Part 1, with several stages of quality control, meeting the demands of both justice and the PMETB with flying colours.

S9

Part 2 MRCPath Examination

{P} RFT McMahon
Division of Laboratory and Regenerative Medicine, University of Manchester, Manchester, United Kingdom

Recent changes to the Part 2 examination include a move to modularisation with separation of the autopsy and histopathology components. In the histopathology section, the number of centres has been reduced to 4 or 5 arranged geographically, accommodating up to 20 candidates per centre. This format has now been run on 3 occasions. Along with an overall reduction in centres, the material for the surgical histology, cytopathology, macroscopic pathology and objective structured pathology examinations (OSPEs) components are produced centrally, so that all candidates deal with the same cases simultaneously. Local centres provide the material for frozen sections and long cases. In the surgical histology section, cases are identified according to an agreed blueprint and marking criteria are developed that are applied in a central marking exercise. Cytopathology cases are similarly identified by a dedicated group, also providing criteria for marking in a centralised setting. Candidates meet with 3 pairs of examiners over the 2 days of the examination to discuss their findings in the macros, frozen sections and OSPEs. Each centre uses a combination of 3 external examiners and 5 internal examiners. Following each running of the new format, there have been opportunities for detailed written feedback by all examiners and meetings at which those aspects of the assessments requiring modification have been identified. The effects of these changes have been to standardise the process, in accordance with PMETB recommendations, with the introduction of centralised marking applying criterion based systems. Evaluation and feedback has been positive and is extremely sensitive to the needs of candidates, while ensuring that appropriate objective standards are maintained.

S10

Functional Pathology of Breast Cancer

{P} AM Neville
Ludwig Institute for Cancer Research, London and New York

Thirty years ago, members of the Ludwig Institute for Cancer Research presented to the Society their early observations on the nature and causation of osteolytic bone metastases in patients with breast cancer. This Doniach Lecture will focus on subsequent progress with emphasis being given to those functionally active factors now known to be produced directly or indirectly by breast cancer that facilitate the development of not only metastases to bone but also other sites. The potential therapeutic implications of these discoveries will be discussed together with the role of the pathologist in guiding therapeutic intervention.

S11

Mechanisms Underlying Chromosomal Instability In Gastrointestinal Cancer

{P} H Grabsch
Leeds Institute of Molecular Medicine, University of Leeds, Leeds, United Kingdom

Chromosomal instability [CIN] is a characteristic feature of gastric [GC] and colorectal cancer [CRC]. Many genes triggering a CIN phenotype have been identified in yeast, but investigations of these genes/proteins in GC and CRC are lacking. The aim of this study was to a establish a thorough understanding of the biological basis of CIN in GC and CRC by (i) investigating key genes/proteins involved in cell cycle regulation and DNA repair pathways and (ii) identifying new markers associated with CIN using whole genome RNA expression arrays and array CGH.
The expression of key proteins involved in the mitotic spindle checkpoint, cell cycle regulation, DNA double strand break [DSB] repair, DNA mismatch repair [MMR] and cell proliferation was studied by immunohistochemistry in large series of GC and CRC. Data from array CGH and array RNA expression from GC were analysed and compared to CRC data. DNA ploidy and frequency of CGH events was measured to assess CIN and the association with clinicopathological data and patient survival was investigated.
In both, GC and CRC, the expression pattern of DNA DSB repair proteins is related to that of MMR proteins supporting the hypothesis of a functional interaction between these two DNA repair pathways in GI cancer. However, different DNA DSB repair proteins are predictors of survival in GC and CRC which may explain differences in response to therapy. While the RNA expression profile and the CGH profile are clearly different between GC and CRC, the association of these profiles with clinicopathological variables including patient survival is less obvious. Surprisingly, the expression pattern of key proteins of pathways presumably responsible for generating CIN is rarely related to DNA ploidy or number of CGH events in GI cancer.

S12

The Pathological Society: Future

{P} DA Levison
University of Dundee, Dundee, United Kingdom

The immediate future of the Society has been mapped out in the document produced last year – The Way Forward. Our mission of "understanding disease" will be pursued energetically through our support of research, training and education in Pathology. I see the Society as leading the rejuvenation of Academic Pathology through our support and organisation of Scientific Meetings, a greater and more active role in mentoring, training and inspiring trainees, more studentships and Clinical Fellowships, and raising the international stature of the Society. We will continue to encourage an editorial policy which will keep the Journal of Pathology at the top of the world pathology journal rankings. Membership will continue to increase if we continue to involve the Membership in the decision-making processes and to provide members with what they want. We will play a key role in striving to make the concept of a National Pathology Week a reality with participation of all the main British Pathology Societies and relevant specialist organisations. We will develop further our outward looking position by building international interactions and collaborations. We will convince people that the Society stands for serious fun.

S13

The Future of Pathology – Tissue Banking

{P} C Womack
AstraZeneca, Alderley Park, Cheshire, United Kingdom

Through primary legislation, the Human Tissue Authority and Codes of Practice, the 2004 Human Tissue Act should provide clarity about the future taking, storage, use and disposal of tissue. Under the Act, tissue banking will be regulated through licensing.

Where tissue is banked for transplantation, the new environment incorporates the EU Tissues and Cells Directive. Pathologists in all disciplines are involved in these activities but there is scope for additional involvement from histopathologists providing autopsy diagnostic support, tissue retrieval, standard setting and training.
The new environment, which includes parallel streamlining of the NHS Research Ethics Service, should alleviate the human tissue based research paralysis that followed the high media profile organ retention affairs…in theory. Clear rules about prospective tissue collection and access to millions of archived samples should ensure that tissue supply is not a rate-limiting step in biomedical research. Research tissue banks will be licensed for that purpose. Pathologists should play a pivotal role in collecting, storing and using tissue for research purposes.
Even with clear rules and regulations, there are challenges ahead: increased costs, changing public and professional perceptions, tissue quality, access to associated clinical data, data processing and collaboration between and within public and private sectors.

S14

Molecular Pathology

{P} F A Lewis[1], P Quirke[2]
[1]. Leeds Teaching Hospitals NHS Trust, Leeds, United Kingdom, [2]. University of Leeds, Leeds, United Kingdom

Molecular pathology is an essential service, which within histopathology has grown slowly and haphazardly within the NHS. In microbiology and haematology the technology has 'killer' applications that supplant standard techniques and strongly drives the introduction of these services. Histopathology has been slow to develop because of the lack of killer applications, the greater complexity of solid tumours, the chronic shortage of pathologists leading to a reduced research base and an entrenchment of the workforce into coping with routine workload, the greater difficulty of dealing with paraffin embedded material (FFPE) and the poor quality of much molecular pathology research.

This is rapidly changing as the first 'killer' technique of Her-2 testing is upon us and has been quickly introduced into practice. Other tests will follow. The pathology work force has been mended and is promptly delivering Consultants to the microscope. The turnaround will lead to increased competition for good posts and thus a resurgence in academic pathology leaves hope for the creation of a new generation of molecular pathologists. FFPE may be more flexible than previously thought with larger fragments recoverable. High quality RNA is also accessible from some blocks. Unfortunately we still publish too many single centre 100 patient studies that are of no value in informing patient care.

Although a scientific role of a molecular pathology clinical scientist has been created, a proper molecular pathology training programme for SHOs and SpRs needs to be introduced. As with the genetics network, molecular pathology laboratories need to be created to inform the community of what tests exist, to QC them and to press for service support. In research, molecular studies must be embedded in clinical trials to prove that they are superior to other methods or that they are additive to gold standard pathology.

To avoid losing our traditional role as the foundation of medicine we must act quickly as radiologists are increasingly encroaching on our staging methods and functional radiology may replace our techniques, the commercial sector are selling "one off" tests at high prices, geneticists are cherry picking high value tests to support their service developments. Without molecular pathology histopathologists are not delivering optimal care to patients and we are losing out on the excitement of the race to explain and treat disease.

ABSTRACT REVIEWERS

Dr MJ Arends, Cambridge

Dr I Buley, South Devon Healthcare NHS Trust

Prof AD Burt, Newcastle-upon-Tyne

Dr SS Cross, Sheffield

Prof AM Flanagan, London

Prof S Fleming, Dundee

Prof CS Foster, Liverpool

Prof AJ Freemont, Manchester

Prof PN Furness, Leicester

Prof KC Gatter, Oxford

Dr JJ Going, Glasgow

Dr JR Gosney, Liverpool

Dr SJ Gould, Oxford

Dr TR Helliwell, Liverpool

Prof CS Herrington, St Andrews

Prof M Ilyas, Nottingham

Dr N Kirkham, Newcastle-upon-Tyne

Prof JA Lowe, Nottingham

Prof SB Lucas, London

Prof JE Martin, London

Dr RFT McMahon, Manchester

Dr A-M McNicol, Glasgow

Prof GI Murray, Aberdeen

Dr KA Oien, Glasgow

Prof JJ O'Leary, Dublin

Dr P Ramani, Bristol

Dr ISD Roberts, Oxford

Dr TP Rollason, Birmingham

Dr N Rooney, Bristol

Prof GN Rutty, Leicester

Dr MN Sheppard, London

Dr DN Slater, Sheffield

Prof RA Walker, Leicester

Prof M Wells, Sheffield