Journal of Anatomy

J. Anat. (2017) **231**, pp445–468 doi: 10.1111/joa.12637

Anatomical Society Winter meeting, King's College London, London 2016

UNDERSTANDING ANATOMY THROUGH EMBRYOLOGY

S1 Enteric nervous system development: understanding disease and therapy

A. J. Burns

Stem Cells and Regenerative Medicine, UCL Great Ormond Street Institute of Child Health, London, UK

Normal function of the gastrointestinal (GI) tract requires the coordinated interaction of the neurons and glial cells of the enteric nervous system (ENS), interstitial cells of Cajal, and smooth muscle cells. The ENS, the intrinsic innervation of the GI tract, is entirely derived from neural crest cells. These precursor cells undergo extensive migration, proliferation and differentiation in order to colonize the entire length of the GI tract and form the ENS. Defects in these processes can give rise to congenital gut motility disorders such as Hirschsprung disease (HSCR), where variable lengths of the hindgut remain aganglionic, resulting in tonic contraction and functional bowel obstruction. Although HSCR is the best characterised paediatric enteric neuropathy, the aetiopathogenesis of other neuromuscular diseases affecting the GI tract is less well understood.

Our research focuses on investigating the genetic, molecular and cellular factors controlling ENS development from neural crest-derived precursors in chick and mouse. The information gleaned from these studies not only helps to unravel the mechanisms underlying normal ENS formation, but allows insight into abnormal ENS development (congenital disease), and accelerates the development of novel approaches, such as stem cell-based transplantations, for their treatment.

S4 The role of embryonic hypoxia and effect of prolyl-hydroxylase inhibitors in development of craniofacial structures in chick embryos

A. Kumar and N. Itasaki

Faculty of Health Sciences, University of Bristol, Bristol, UK

During embryogenesis, neural crest cells (NCCs) arise from the neural tube by epithelial-mesenchymal transition (EMT) and differentiate into various cell types. There are two populations of NCCs, cranial and trunk. In the cranial region, many NCCs contribute towards facial bones and cartilages, providing the skeletal basis for the face, maxilla, mandibular and neck structures. A deficit of cranial NCCs results in congenital craniofacial hypoplasia.

In adult tissue hypoxia, hypoxia-inducible factor-1α (HIF-1α) permits cell adaptation to a hypoxic environment by promoting angiogenesis and anaerobic glycolysis, thus aiding tissue recovery. However, in normoxia, HIF-1α is readily degraded via oxygen-dependent prolyl-hydroxylases (PHDs). Because of this, chemical compounds that stabilise HIF-1α, such as PHD inhibitors, are used for stroke therapies. Other functions of HIF-1α include promotion of EMT and metastasis in tumour and up-regulation of chondrogenesis, both of which are promoted in a hypoxic microenvironment.

Given that embryos are naturally hypoxic, our group has recently shown that induction of NCCs by EMT is upregulated by HIF-1α-stabilising PHD inhibitors in chick embryos cultured *ex ovo*. We replicated this effect *in ovo*; the increase of NCCs was examined using *Sox10* as a marker and RNA *in situ* hybridisation, at embryonic day 2. We then investigated the effect of PHD inhibitors at later stages of embryogenesis, up to embryonic day 10, to determine whether specific cell fate is increased and whether there is any impact on the development of other structures. Through cartilage staining, we found advanced development of the otic capsule, a cartilaginous structure encapsulating the inner ear, presumably due to hyperplasia caused by the increase of EMT and promoted chondrogenesis by PHD inhibitors. This was further investigated via utilising a micro-CT scanner, allowing us to produce three-dimensional models and animations of the otic capsule and analyse them in detail. This analysis revealed that PHD inhibitors changed the structure of the otic capsule, which in turn, altered the inner ear within it. This suggests that PHD inhibitors have the ability to upregulate the otic capsule formation and in the future could be applied to embryos that are destined to have a hypoplastic otic capsule.

We would like to thank Dr Tom Davies, Faculty of Life Sciences, University of Bristol, for his help in obtaining and analysing micro-CT scans.

We would also like to thank the Anatomical Society for their support and funding, without which this research would not have been possible.

S5 Going with your gut: using enteric nervous system stem cells for spinal cord injury repair, utilising the chick as a model organism

B. Jevans, N. Thapar and A. J. Burns

Stem Cells and Regenerative Medicine, UCL GOS Institute of Child Health, London, UK

Spinal cord injury (SCI) causes paralysis, multisystem impairment and reduced life expectancy; there is as yet no cure. Stem cell treatment can potentially replace lost neurons, promote axonal regeneration and limit scar formation. However, an optimal stem cell source has yet to be found. Our work explores the use of enteric nervous system stem cells (ENSSCs) isolated from the enteric nervous system (ENS) of the gastrointestinal tract.

ENSSCs can be harvested easily via endoscopy, raising the possibility of autologous cell transplantation for a range of neural disorders. This project will evaluate ENSSCs as a cell therapy for SCI by assessing survival, spread and differentiation of ENSSCs transplanted into the chick embryo spinal cord (SC).
GFP labelled ENSSCs were generated via chimeric grafting. Briefly, neural tubes of GFP^+ embryonic day (E) 1.5 embryos were grafted into age-matched WT embryos. At E14 the intestines were harvested, and GFP^+ ENSSCs were isolated by FACS and cultured to form neurospheres. For co-culture experiments and qPCR comparison, SC cultures were generated from age-matched WT embryos and labelled with mCherry lentivirus. For transplantations, a region of the neural tube was ablated in E1.5 embryos and a neurosphere grafted into the injury site. Embryos were harvested at timed intervals and analysed via cryosectioning and immunofluorescence. All procedures involving live embryos were in accordance with the UK Animal (Scientific Procedures) Act.
Isolated GFP^+ ENSSCs formed extensive cell connections when co-cultured with SC-derived cells, implying potential for *in vivo* integration of ENSSCs within the SC following transplantation. qPCR analysis revealed ENSSC neurospheres contained $TuJ1^+$ neurons, $S100^+$ glia and $Sox10^+$ stem cells, and expressed key neuronal subtype genes at levels comparable to those of SC neurospheres. Following transplantation into the SC of E1.5 chick embryos, GFP^+ ENSSCs were found up to 8 days later (latest time point examined), predominantly within the white matter. ENSSCs formed bridging connections within the SC injury zone and aligned along the anterior/posterior axis. Transplanted ENSSCs were immunopositive for the neuronal marker TuJ1.
This pilot study provides vital data supporting the use of ENSSCs for SCI, and informs future work of ENSSC transplantation into an adult rodent model of SCI. Acknowledgements: Ayad Eddaoudi, Stephanie Canning, FACS facility, UCL GOS-ICH.

S6 How to shape Meckel's cartilage: lessons from the mouse

M. Burton and N. Anthwal

Department of Craniofacial Development and Stem Cell Biology, King's College, London, Guy's Hospital, London, UK

The dentary (lower jaw bone) is an important feature of the human facial skeleton. Functionally, it enables us to chew food and communicate, and it carries the dentition. Central to this functionality is the shape of the mandible and, therefore, the morphogenesis of the developing mandible is closely regulated. Part of this regulation comes through Meckel's cartilage (MC). MC is a transient rod-like structure within the developing mandible that acts as a template for the condensing dentary. We show that the shape of MC is regulated by a group of signalling molecules, transforming growth factor beta (TGF-β). When the TGF-β signalling pathway is perturbed, the linear morphology of MC is disrupted. In the absence of TGF-β signalling MC displays a bending response, resulting in a sigmoidal morphology. Furthermore, we find that the cellular activity of both chondrocytes and perichondrocytes of MC are regulated by TGF-β. This suggests that TGF-β helps maintain the rod-like morphology of the developing MC through regulating cellular activity. Overall, we demonstrate that TGF-β signalling is key in controlling the morphogenesis of the dentary and ultimately its shape.
Experimental animals were cared for and killed according to UK Home Office licence and regulations.

S7 An investigation of the anatomy of the infrapatellar fat pad: a cadaveric study

J. Leese and D. C. Davies

Human Anatomy Unit, Department of Surgery and Cancer, Imperial College London, London, UK

The infrapatellar fat pad (IFP) is an extrasynovial, intracapsular, adipose body that occupies the space in the knee joint formed by the inferior pole of the patella, the femoral condyles, tibial plateau and patellar tendon. The IFP has been suggested to play a role in joint lubrication, shock absorption, proprioception, the progression of osteoarthritis and anterior knee pain syndrome. However, not enough is known about the anatomy of the IFP to underpin any of these suggested functions. Forty-three knees from 11 male and 15 female embalmed cadavers (mean age 84 years; range 55–97 years) were investigated. The cadavers were donated and the study was performed in compliance with the provisions of the UK Human Tissue Act (2004). The quadriceps tendon and the medial and lateral patellar retinacula were dissected from the patella, which was then reflected antero-distally. The IFP was carefully excised and details of its morphology and attachments to components of the knee joint were recorded, together with the presence of articular surface pathology on the patella and femoral condyles. The principal novel findings of the current study were that 81% of IFPs were attached to the superior pole of the patella by supero-medial extensions and 65% were attached by supero-lateral extensions; the supero-medial extensions were larger than the supero-lateral extensions. The superior extensions of the IFP were always attached anteriorly to the patellar retinacula and in four individuals the extensions formed a full loop around the superior pole of the patella. Significantly fewer knees with patella ($P = 0.001$) and femoral ($P = 0.002$) articular surface osteophytes exhibited superior IFP extensions and these extensions were significantly shorter in knees with patella ($P = 0.000$) and femoral ($P = 0.006$) osteophytes than in those without. In addition, the percentage attachment of the IFP to the medial meniscus was significantly shorter in knees with patella ($P = 0.023$) and femoral ($P = 0.050$) osteophytes than in those without osteophytes. This demonstration of a relationship between IFP morphology and knee joint pathology supports a functional role for the IFP that requires further investigation.

S8 An anatomical study of geographic and sex differences in the location and number of the zygomaticofacial foramen using cadaveric specimens

A. Ferro,[1] S. Basyuni,[1] C. Brassett[2] and V. Santhanam[3]

[1]*Hughes Hall, University of Cambridge, Cambridge, UK;* [2]*Department of Physiology, Development and Neuroscience, Downing Site, Cambridge, UK and* [3]*Addenbrooke's Hospital, Cambridge University Hospitals NHS Foundation Trust, Cambridge, UK*

The zygomaticofacial foramen (ZFF) is an opening in the zygoma that transmits the zygomaticofacial neurovascular bundle. Identification of the ZFF is important in surgical procedures to avoid injuring these structures. Although anatomical variation in the ZFF is recognised, there are no comprehensive studies of ZFF anatomy in different populations. This study aims accurately to map the location of the ZFF and determine geographic and sex differences in 429 skulls (858 zygomas) from nine different geographic locations. On each zygoma, the number of ZFF was recorded and patency was confirmed using a wire probe. To produce consistent landmarks for measurement, a cross-line laser was projected onto each skull, with Line 1 aligning with the Frankfurt horizontal plane and Line 2 aligning vertically with the posterior margin of the zygomaticofrontal suture. Measurements were taken using digital callipers. Statistical analysis was performed using R version 3.3.1, with two-way ANOVA for comparison of foramina location between geographic populations and sides, and between sexes and sides. Log-linear analysis was used for comparison of foramina number, with the Chi-square test for follow-up comparisons. Results showed that the number of ZFF per zygoma ranged from 0 to 4, with one foramen being commonest (49.8%). Although there was no sex difference in the number of foramina per zygoma, the number of foramina differed significantly between geographic populations. With regard to location, the mean distance of the ZFF from the orbital margin was significantly longer in males ($P < 0.001$), but ZFF location with respect to Lines 1 and 2 was similar between sexes ($P = 0.419$ and $P = 0.493$). However, ZFF location, both with respect to the orbital margin and to Lines 1 and 2, differed significantly between geographic populations. Irrespective of geographic population or sex, 81% of ZFF on the left zygoma, and 83% on the right, were found within a circle of 15-mm diameter. In conclusion, anatomical variation does exist in the location and number of ZFF between sexes and between different geographical populations. In addition, the landmarks created using a cross-line laser may have clinical application, and this measurement technique may be used in future studies to improve data reproducibility.
No ethical approval was required for this study.

S10 Understanding anatomy: lessons from abnormal mouse embryo development?

T. Mohun

The Francis Crick Institute, London, UK

If an appreciation of the embryological origins of anatomical structures and their developmental history can help our understanding of normal anatomy, then the same is also likely to be true for abnormal embryo development. Studying the nature of the malformations that occur during gestation can identify critical steps in the morphogenesis of individual anatomical features, steps that can be disrupted by a range of genetic or environmental influences. This in turn may clarify our ideas about the sequence of changes that build the mature, adult anatomy.
From systematic efforts to investigate gene function by targeted gene mutation in the mouse, it is now clear that a remarkable one-third of all the genes in the mouse genome are essential for normal embryonic development and survival beyond birth. 'Deciphering the Mechanisms of Developmental Disorders (DMDD)' is a collaborative international research programme funded by the Wellcome Trust to study the nature of such 'embryonic lethal' gene knockouts and investigate their potential as models of congenital abnormalities (dmdd.org.uk). I will review the results to date, focusing on abnormalities affecting the developing heart and what they might teach us about the origins of normal cardiac anatomy.
No ethical approval was required for this study.

S11 Deciphering Broken Hearts: a study in mice

D. J. Henderson

Cardiovascular Research Centre, Institute of Genetic Medicine, Newcastle University, Newcastle upon Tyne, UK

Our understanding of how the mammalian heart develops has changed fundamentally over the past 15 years. In the last century, it was believed that all of the cells that would contribute to the myocardium of the fully formed heart were present and already patterned in the cardiac crescent and primitive heart tube. However, in 2001, three seminal papers suggested that this premise was incorrect and that the cells that form the myocardium and the endocardium of the inlet and outlet parts of the heart (the outflow tract, right ventricle and much of the atria) actually arise from a second heart-forming region, now known as the second heart field (SHF), that adds onto the primitive heart tube after it has formed. This new model is now widely accepted. However, the heart is more than just a myocardial pump with an endocardial lining. We now know that many of the cells that form the other key cell types in the heart (epicardium, fibroblasts, smooth muscle cells, fibrous septal cells, valve interstitial cells, nerves) arise outside the heart-forming fields and migrate into the heart after it has formed. Understanding the interplay between these different lineages of cells has changed our understanding not only of how the heart forms, but also of how congenital heart defects arise.
In this talk I will focus on the development of the cardiac outflow tract and discuss how the interaction between different cell lineages determines the anatomy of the mature aorta and pulmonary trunk, and explains common congenital and adult pathologies that affect them.
The studies were approved by the Newcastle University Animal Welfare and Ethical Review Board and conformed to the Animals (Scientific) Act 1986 (UK) and Directive 2010/63/EU of the European Parliament.

S16 Exploring plasticity during mouse molar development

R. Lav, J. Fons Romero and A. S. Tucker

Department of Craniofacial Development and Stem Cell Biology, King's College, London, Guy's Hospital, London, UK

Odontogenesis involves a sequential progression through a series of morphodifferentiation stages. An initial glance at the histology of tooth development gives an impression of similarity between the buccal and lingual aspects of the developing tooth germ. However, this apparent histological symmetry belies

differences in the underlying gene expression. Sox2 is one such gene that is expressed primarily on the lingual aspect of the developing tooth germ and contributes asymmetrically to its development. Our investigation pertains to the difference in the developmental dynamics of the buccal and lingual aspects of the developing molar tooth germ. For this, we take advantage of murine molar explant cultures to perform tooth-splitting and fate-mapping experiments. Through our experiments, we explore the role of progenitor cells and the inherent plasticity of specified odontogenic cells, in tooth development. In addition, we have utilised Sox2-CreERT2;R26-TdTomato transgenic reporter mice for live imaging of cell movement and fate mapping of the Sox2-derived cells. Through these experiments, we hope to understand the inherent differences in the seemingly synchronised development of the molar tooth germ. All animal experiments were conducted in accordance with the guidelines set by the UK Home Office under a registered project licence.

S17 The effects of fetal immobility on joint shape morphogenesis in the mouse: a quantitative study using 3D image registration

P. Sotiriou,[1] P. Murphy,[2] R. A. Rolfe[1] and N. C. Nowlan[1]

[1]*Imperial College, London, UK and* [2]*Trinity College, Dublin, Ireland*

Arthrogryposis and developmental dysplasia of the hip, two conditions with abnormal neonatal joint shape, are associated with reduced or abnormal fetal movements. The severity and incidence of congenital joint shape abnormalities is not equal to all joints; the hip joint is the most commonly affected joint in neonates, and the joints of the arms are more frequently affected than those of the legs in infants with arthrogryposis. In animal models of abnormal fetal movements, different bones and joints are affected with varying degrees of severity. However, because of the complex nature of measuring changes in 3D shapes, changes in joint shape in these models have never been quantified.

In this project, image registration techniques were used to identify, and quantify, differences in joint shape between immobile mouse embryos (absent skeletal muscle due to lack of any functional copies of Myf5 and MyoD) and their healthy littermates. Hindlimbs and forelimbs from Theiler Stage (TS) 23 immobile and normal littermate mouse embryos (n = 5–7 for each) were stained for cartilage, and scanned in 3D using optical projection tomography. All experiments complied with European legislation. The scapula, humerus, radius, ulna, pelvis, femur, tibia and fibula were virtually segmented, and multiple rigid registrations performed to align bones of the same type precisely. Mutant and control atlases were created and overlaid to identify region-specific size and shape changes in the articular surfaces of each rudiment. Multiple joint shape parameters of each post-alignment scan were measured and statistical tests performed.

Statistical analysis of the measurements revealed that, of the four major joints examined, only the shoulder and elbow joint shapes were significantly affected in the immobile embryos. In contrast to the prevalence of hip abnormalities in human babies, the immobile embryos exhibited a normal hip joint shape at TS23. This study demonstrates that the forelimb joints of immobile mouse embryos are more severely affected than the hindlimb joints, as occurs in some forms of arthrogryposis. A greater understanding of joint shape abnormalities in immobile mouse embryos brings us closer to developing treatments compensating for reduced or abnormal fetal movement in humans. No ethical approval was required for this study.

S19 MRI and clinical patterns in adult humans with symptomatic meniscal tears necessitating knee surgery

S. A. Webb,[1] C. Brassett[2] and J. Chitnavis[3]

[1]*Selwyn College and School of Clinical Medicine, University of Cambridge, Cambridge, UK;* [2]*University Clinical Anatomist, Department of Physiology, Development and Neuroscience, Cambridge, UK and* [3]*Consultant Orthopaedic Surgeon, The Cambridge Knee Clinic, Cambridge, UK*

Despite its prevalence, the precise mechanism of meniscal tearing is unknown. In this study, the MRI appearances of the knees of 58 patients undergoing surgery for meniscal tears were analysed. Cases were selected retrospectively from one surgeon's database of 281 patients who underwent arthroscopic knee surgery consecutively between 2010 and 2014. Exclusions included 69 patients without meniscal tears, 32 with complex operations, and 122 whose pre-operative MRIs were unavailable. Clinical information was obtained from *pro formas* completed by the surgeon during clinic and upon operation. All 58 MRI scans were studied by one observer using a novel *pro forma* consisting of a series of closed questions to gather data systematically from MRI scans. When this *pro forma* was applied to 10 randomly selected scans, intra-observer repeatability was 82.4% and inter-observer repeatability 68.0%. The mean age of the 58 patients was 51.3 $\pm$ 12.7 years. Of these, 74.1% were male and 43.1% were right knee cases. Medial (MM) and lateral meniscal (LM) tears accounted for 86.2% and 37.9% of knees, respectively. MM tears most commonly involved both middle and posterior thirds (41.4% of knees), whereas LM tears most commonly involved all thirds (27.6%). Tears in both menisci were the most common in the horizontal plane, with 58.6% MM and 70.7% LM. Extrusion was 2.5 times more common in the MM. We propose that the coronary ligaments and ligaments connecting the medial collateral ligament (MCL) and MM might contribute to the pattern of meniscal tears in view of the following observations: (i) the prevalence of tears and extrusion in the MM over LM might result from forces transmitted in these ligaments, which are enriched by attachment to the MCL medially, but not the LCL laterally; (ii) the coronary ligaments are attached to both menisci in the horizontal plane: the plane in which most meniscal tears occur; and (iii) on MRI scans, the most common location of tears in the MM approximated the insertion into the MM of these MCL-MM ligaments. We therefore hypothesise a role for the coronary ligaments and these MCL-MM ligaments in force transmission during meniscal tearing. Ethical approval was granted by the Cambridge Human Biology Research Ethics Committee in June 2015.

S20 Role of hydrogen sulphide in mediating migration of human trophoblast cells

N. Sharma, S. K. Gupta, S. Saxena, S. Mochan, N. Sirohi, A. Sharma and R. Dhingra

Department of Anatomy, All India Institute of Medical Sciences, New Delhi, India

Introduction: Therapeutic effect of H2S on an animal model of pre-eclampsia showed reduction in blood pressure. Hydrogen sulphide (H2S) is a gasotransmitter reported to have a migration-promoting effect on endothelial cells. The aim of the current work is to study the effect of hydrogen sulphide on migration of trophoblast cells.

Material and methods: BeWo cells were exposed to H2S donor (NaHS) and hypoxia/reoxygenation (H/R) injury. The migration potential of cells was assessed with the help of wound-healing assay. Localisation and distribution of F-actin were seen with immunofluorescence.

Results: The differences in percentage wound-covered area and rate of migration of NaHS-treated vs. control groups and H/R-treated vs. H/R followed by NaHS-treated groups were found to be statistically significant ($P < 0.03$ and $P < 0.01$). NaHS-treated cells were characterised by the presence of numerous cytoplasm projections such as lamellipodia and filopodia. When administered exogenously upon the BeWo cells, H2S promotes cell migration by augmenting formation of lamellipodia and filopodia.

Conclusion: The results from the present study point towards the role of H2S in migration of trophoblast cells, thereby predicting a role in the pathogenesis of pre-eclampsia and other clinical outcomes associated with it such as IUGR. The new challenge of immediate interest is to find out the potential receptor utilised by H2S.

Ethical approval was granted by the ethics committee of the All India Institute of Medical Sciences, New Delhi on the 27 July 2015.

S21 Electromagnetically navigated foramen ovale injections for the treatment of trigeminal neuralgia – a human cadaveric study

M. Lonie,[1] I. Pimentil,[1] A. Wiggins,[2] P. Bodkin[2] and A. Venkatesh[1]

[1]*School of Medicine, Medical Sciences and Nutrition, University of Aberdeen, Aberdeen, UK and* [2]*Department of Neurosurgery, Aberdeen Royal Infirmary, Aberdeen, UK*

Trigeminal neuralgia is a well-known cause of severe paroxysmal episodic craniofacial pain occurring along the distribution of one or more trigeminal divisions. A well described minimally invasive treatment for this condition is the ablation of the trigeminal ganglion (TG) by percutaneously injecting glycerol into Meckel's cave (MC) via the foramen ovale (FO). This procedure is usually performed using surface landmarks and fluoroscopy to guide the needle into the cheek and then through the FO in the skull base to reach MC. Recently, electromagnetic (EM) neuronavigation has being used stereotactically to guide the needle into MC. Using seven embalmed cadaveric specimens aged 81–93 years (14 FO injections), we placed a spot of dye at the tip of a stereotactically placed needle and subsequently dissected the specimens to determine dye location within MC.

Specimens first underwent a volumetric CT scan and images were transferred to the neuronavigation system (StealthStation s7, Medtronic). The FO was identified on the navigation system and a trajectory was planned pre-injection. A standard cannulated needle was used together with the EM guidance stylet (AXiEM, Medtronic). The needle was advanced through the FO to a point 4 mm above the middle cranial fossa floor. 0.25 mL of 4 mg mL^{-1} indocyanine green (ICG) dye was injected. Specimens were dissected and the dye location was recorded on a proforma dividing the TG into seven anatomical regions (N, G1, G2, G3, V1, V2, V3).

Fourteen FO injections were performed. Subsequent dissection showed dye within MC in 11 instances (within the V3 segment in 9/11 and within the G3 segment in 6/11). In three instances no dye was found within MC. Dissection along the injection track revealed dye within the lateral pterygoid muscle in one case, and no dye in 2. These three instances of absent/ectopic dye may be explained by loss of the dye volume within the 'dead space' of the needle (~0.2 mL) or inaccuracy of volume measurement. No dye was found in the immediately surrounding structures (such as the internal carotid artery) on wider dissection.

Our pilot study demonstrates that percutaneous EM-navigated FO injection is an accurate method for targeting the trigeminal ganglion. No ethical approval was required for this study.

S22 Embryos, teratogens and birth defects

N. Vargesson

School of Medicine, Medical Sciences and Nutrition, Institute of Medical Sciences, University of Aberdeen, Aberdeen, UK

Our understanding of normal vertebrate embryonic development has been helped by studying processes that cause and result in malformation. Vertebrate embryos have long been used to understand how drugs with known negative actions upon development, known as teratogens, act and interfere with embryogenesis, the most notorious example being thalidomide. Such studies, in a range of species, have provided vital information about drug action and have also shed light on mechanisms vital for normal development, for example, the role of the forming vascular system in embryonic development and how abnormal changes to vascular patterning may result in a birth defect. Indeed, analyses of several human limb anomalies indicates that vascular pattern changes (or lack of) during embryogenesis could be responsible for the malformations. A discussion on the vascular system, its role in normal and abnormal embryonic development, and factors and causes that result in a failed vascular system will be reviewed. In addition, the use of vertebrate embryos to screen a variety of drugs and compounds to ascertain their safety and give an indication of their potential actions in humans will be outlined. Examples to be discussed include screening of analogues of thalidomide designed to retain clinical, positive benefits which lack the negative side-effects.

S24 Evolution of the animal face: from principles to mechanisms

A. Abzhanov

Department of life Sciences, Imperial College London, London, UK

Understanding the origins of animal diversity is one of the chief challenges to the modern biological sciences. We aim to reveal molecular mechanisms underlying evolutionary processes that generate morphological variation, and to show how particular changes in embryonic development can produce morphological alterations for natural selection to act upon. The principal focus for our studies is on the animal face and head. Cranial diversity in vertebrates is a particularly inviting research topic as animal heads and faces show many dramatic and unique adaptive features which reflect their natural history. Most of the facial diversity depends on the shapes and sizes of the bones and cartilages that make up the cranial skeleton. I will describe how our investigations of craniofacial skeletal development are helping us to uncover mechanisms that generated cranial diversity during evolution. We employ a synergistic combination of geometric morphometrics, comparative molecular embryology and functional experimentation methods to trace cranial evolution in reptiles, birds and mammals, some of the most charismatic animals on our planet.
No ethical approval was required for this study.

S25 Using clinical, mathematical and engineering techniques to further understand the influence of the anterior cranial fossa in craniofacial trauma: a human study

J. R. Stephens, K. Patel, M. Gaglione, S. LeComber, E. Barbieri, S. Border, B. T. Evans and S. Holmes

The Craniofacial Trauma Research Group, Queen Mary University of London, London, UK

Craniofacial trauma involving the anterior cranial fossa (ACF) produces a heterogeneous group of fracture patterns. Variance is influenced by the osseous anatomy, biomechanical properties of the craniofacial construct, and the magnitude and vector of the impacting energy. The anatomy of the central and lateral regions of the ACF varies significantly. Our aim was to study ACF fractures and evaluate how the anatomy of the ACF affects force propagation and clinical outcome in craniofacial trauma.
Eighty-one patients sustaining a frontobasal fracture were identified from two major UK trauma centres. Clinical outcome was correlated with the direction of impacting force to the frontal bone – either anteriorly or laterally. Linear metrics applied to individual fracture patterns allowed mathematical quantification of fracture length, nodes, termini and fractal dimension. A finite element analysis model was constructed further to evaluate how the anatomy of the skull base was disrupted from varying velocities and vectors.
Clinical analysis indicated that lateral impacts resulted in a significantly lower GCS and increased requirement for intubation ($P < 0.001$ and $P < 0.001$, respectively). Mathematical investigation revealed that anterior forces produced fractures in the immediate vicinity of impact, with lateral blows producing more widespread and distal damage (variance ratio test: $F_{48,31} = 0.092$, $P < 0.001$), more frequently extending into the middle and posterior fossae. As total fracture path length increased, the fractal dimension (FD) of the pattern increased more rapidly for anterior impacts than for lateral impacts (linear model: length $F_{1,77} = 21.25$, $P < 0.001$; FD: $F_{1,77} = 6.49$, $P = 0.013$). Modelling allowed imaging of energy transfer across the ACF, showing energy absorption by the paranasal sinuses and thin orbital roofs and energy transmission along the robust sphenoid wing.
Using three complementary techniques we have established that lateral blows produce long, relatively unbranched fracture patterns, whereas anterior blows create more complex and highly reticulated patterns. The ACF anatomy influences these findings with its delicate and lattice-like configuration within the central skull base serving as a 'crumple zone' and absorbing force; a feature absent in the lateral ACF, where a lack of collapsible interface exists. Full ethical approval was granted under REC13/WM/0194.

S26 Dissecting the causes of frontal cortex abnormalities in a mouse model for CHARGE syndrome

A. Donovan and A. A. Basson

Department of Craniofacial Development and Stem Cell Biology, King's College London, London, UK

Dominant mutations in the gene encoding the ATP-dependent chromatin remodelling factor CHD7 are the major cause of CHARGE syndrome (Coloboma, Heart defects, Atresia of the choanae, Retardation of growth, Genital anomalies and Ear defects). Many individuals with CHARGE syndrome have been reported to have deficits in executive functioning, often associated with a diagnosis of an autism spectrum disorder. These manifest as problems in behaviours such as maintaining attention, impulse control, and understanding the effects of their actions on others.
The neurodevelopmental and neuroanatomical underpinnings of these defects are not yet known. However, a number of functional studies in both human and mouse have identified the prefrontal cortex as the principal brain region governing executive functions. We will therefore use a $Chd7^{+/-}$ mouse model to investigate the impact of Chd7 heterozygosity on the development and function of the prefrontal cortex. I aim to characterize changes in anterior-posterior patterning of the neocortex in the $Chd7^{+/-}$ mice and investigate the interaction of Chd7 with the fibroblast growth factor (FGF) signalling pathway. I will discuss the first results of these studies by reporting subtle changes in the expression of Fgf8 in the anterior neural ridge (ANR), which direct anterior-posterior neocortical regionalisation.
To define specific changes in gene expression that may underlie neurodevelopmental abnormalities in $Chd7^{+/-}$ mice, genome-wide transcriptomic analysis at several stages in frontal cortex development will be carried out via RNAseq.
To further observe neuroanatomical changes resulting from Chd7 haploinsufficiency, MRIs of adult $Chd7^{+/-}$ brains will be carried out and will be compared with those of CHARGE syndrome patients.

Overall, these studies will provide insights into the structural, developmental, genetic and electrophysiological alterations that underlie prefrontal cortex abnormalities in $Chd7^{+/-}$ mice, and allow us to begin relating these to patients with CHARGE syndrome.
All animal work was approved and performed under HO PIL 70/6694.

S28 Temporal effects of movement on early joint shape development in the chick

D. Bridglal and N. C. Nowlan

Department of Bioengineering, Imperial College London, London, UK

The most common musculoskeletal condition post-natally is developmental dysplasia of the hip (DDH), a term describing the array of malformations concerning the hip joint. DDH can lead to compromised locomotion and premature degenerative disease. During development, a reduction in fetal movement is a common risk factor for DDH.
Despite the relevance to skeletal malformations such as DDH, we have almost no understanding of mechanisms underlying early joint morphogenesis. The immobilised chick embyro has been proposed as a model system for early-onset DDH, and recent work demonstrates that fetal movements are important for joint development. Hence, we aim to understand precisely when, and in what way, fetal movements impact upon early hip joint morphogenesis. Cartilage rudiments of the chick hindlimb are apparent from embryonic day (E) 5. Independent limb movements first occur at E6.5, with joint cavitation occurring at E8.5. Embryonic chicks were immobilised with decamethonium bromide to remove dynamic muscle forces for (i) cumulatively longer periods of time, and (ii) 2- and 3-day periods, with both periods falling within the window between E3 to E8. Chicks were harvested at E9. Three-dimensional reconstructions obtained from optical projection tomography were used to obtain morphological measurements of rudiments, and cavitation was assessed using histological sections stained for cartilage.
In all examined specimens immobilised from E3 to 8, E4 to 8, or E5 to 8 ($n = 3$ per group), hip joints were fused and without a joint cavity. However, immobilising for shorter periods of time from E6 to 8 or E7 to 8 led to partial cavitation in some hip joints, with other joints cavitating normally. Analyses of joint shapes demonstrated that the longer the chick was immobilised, the more abnormal the emergence of joint shape. The most significant reductions in acetabular height, width and depth, and femoral head epiphyseal width were seen in chicks immobilised from E3 to 8. However, immobilisation from E5 to 7 led to similar effects on shape as immobilisation from E3 to 8. Cavitation was also absent in hips immobilised from E5 to 7 ($n = 3$).
These results indicate that fetal movement between E5 and E7 are critical to cavitation and to joint shape, potentially related to the timing of the first limb movements at E6.5.

S29 The role of Gas1 in murine salivary gland development

D. Cuckovic,[1] M. Seppala,[1,2] M. T. Cobourne[1,2] and A. Tucker[1]

[1]*Department of Craniofacial Development and Stem Cell Biology, King's College London Dental Institute, London, UK and* [2]*Department of Orthodontics, King's College London Dental Institute, London, UK*

Parasympathetic innervation plays a fundamental role in salivary gland development maintaining epithelial progenitor cells. Reciprocally, the epithelial progenitors have been shown to mediate critical steps of gangliogenesis and influence neuronal survival. The ductal epithelium establishes neuronal-epithelial communication and the neurons coalesce around the epithelial bud stalk, resulting in the formation of the submandibular parasymphathetic ganglion. Axonal projections extend out from the ganglion towards the epithelial endbuds. Shh signalling is critical for SG development, and Gas1, a key component of the Shh pathway, has recently been shown to have a role in axonal guidance. Given this, we aimed to address the role of Gas1 in SG development using a mouse model. Interestingly, Gas1-positive cells were found in SG epithelium from early developmental stages in a dynamic pattern. By embryonic day 12, Gas1-positive cells were localised at the epithelial bud stalk, but at later stages they were widely distributed in the terminal endbuds. Interestingly, loss of *Gas1* led to hypoplastic SGs with reduced branching ability and premature depletion of early epithelial progenitors. The glands showed disrupted innervation with decreased axonal projections accompanied by ganglion hypoplasia and misplacement. Furthermore, suppression of Shh signalling using cyclopamine severely affected the branching ability of *Gas1* knockout salivary glands compared with the effect observed in wildtype embryos, suggesting that Gas1 positively regulates Shh signalling within this organ. These data indicate a critical role for Gas1 in epithelial patterning and SG innervation.
All work performed complies with UK Home Office regulations and all licences are in place.

S30 Altered cellular development in the embryonic rat spinal cord following maternal immune activation

R. Anderson,[1] T. Foley,[2] J. Radford,[2] S. O'Hallaron,[1] G. W. O'Keeffe[2] and K. W. McDermott[1]

[1]*Graduate Entry Medical School, University of Limerick, Limerick, Ireland and* [2]*Department of Anatomy and Neuroscience, University College Cork, Cork, Ireland*

Retrospective human studies and subsequent studies in animal models have suggested that maternal immune activation (MIA) may be a risk factor in the development of neurological disorders such as schizophrenia, autism, cerebral palsy and epilepsy in offspring. These studies indicate that the effect of MIA on offspring may differ depending on CNS region, gestational timing of insult and cell population examined. Much work remains to fully appreciate the importance of these spatial and temporal determinants in CNS response to MIA.

The effects of MIA on spinal cord (SC) development were investigated in immunohistochemical studies. All animal procedures were carried out in accordance with Republic of Ireland Department of Health and Children licences, compliant with the Institutional Animal Care and Use Committee, and complied with the European Council Directive (86/609/EEC). Time-mated dams received a single intraperitoneal injection of 50 μg kg^{-1} lipopolysaccharide (LPS), 100 μg kg^{-1} LPS or saline on embryonic day (E)12, 14 or 16. At 5 h post-injection, dams were anaesthetised with pentobarbital and decapitated. Embryos were harvested by laparotomy, fixed, frozen and cryosectioned at 15 μm. Rostral, intermediate and caudal slices were immunofluorescently stained with the antisera Olig2, Iba-1 and Reelin (a marker of the oligodendrocyte lineage, the microglial lineage, and a glycoprotein involved in cell migration during early development, respectively).

Olig2-positive cell number decreased in both the grey matter (GM) and white matter (WM) of the rostral SC of offspring 5 h post-maternal injection with 100 μg kg^{-1} LPS at E16. This decrease was not observed in intermediate or caudal regions. No changes were observed following 50 μg kg^{-1} LPS injection at either E14 or E16. The number of Iba-1-positive cells is also reduced in SC WM and GM at E16 5 h after maternal injection of 100 μg kg^{-1} LPS.

MIA appears to have an acute effect on microglial and oligodendroglial cell populations in the developing rat SC. Further analyses will be completed to fully elucidate the means by which MIA affects development of these key cell populations in the SC embryonically and, indeed, during postnatal development.

S31 Where does near-peer teaching belong in the curriculum? A study investigating student experience, knowledge gain and retention

W. J. C. Parton,[1] C. H. Harrison,[1] M. A. Myers,[1] E. G. Seaby,[2] A. Elmansouri,[1] S. R. Hall,[1] J. R. Stephens[1] and S. Border[1]

[1]*Centre for Learning Anatomical Sciences, University of Southampton, Faculty of Medicine Southampton General Hospital, Southampton, UK and* [2]*Department of Human Genetics and Genomic Medicine, Faculty of Medicine, University of Southampton, Southampton, UK*

Near-peer teaching (NPT) is a recognised method of instruction utilised by medical institutions worldwide. Near-peer tutors are often 1–2 years senior in their stage of training than the learners whom they teach. At the University of Southampton near-peer tutors have provided frontline and revision tutorials covering the cranial nerves. Both sessions were fully integrated into the clinical head, neck and neuroanatomy curriculum and formal teaching timetable. We define frontline teaching as the first time the taught material is communicated to the learner. NPT provides a multi-beneficial educational platform with benefits for learners, tutors and medical faculties. Cognitive and social congruencies between the student learner and student teacher have been postulated to enhance the dynamics of the learning environment. Although this is well established within the literature, to our knowledge no investigators have compared students' experience of frontline and revision NPT tutorials. This study aimed to: (i) compare second-year medical students' attitudes, perceptions and experiences of near-peer tutors between curriculum-based tutorials and revision tutorials, and (ii) measure the knowledge gain between frontline and revision small group tutorials. A Likert style survey instrument (Cronbach-α = 0.74) was distributed at the end of both tutorials. Participants completed paper-based, formative, baseline and post-exposure knowledge tests. A total of 185 and 89 second-year medical students attended the frontline and revision tutorials in week 3 and 8 of the module, respectively. Of these, 183 frontline and 89 revision participants completed the Likert survey. Overall, teaching and factors relating to social and cognitive congruence scored significantly higher for frontline than revision tutorials ($P < 0.05$). Comparison of baseline and post-session knowledge assessments in week 3 ($n = 185$) and week 8 ($n = 89$) of the module showed a significant gain in baseline knowledge ($P < 0.0001$) that was retained between the two sessions ($P < 0.05$). Our results demonstrate that students' experiences of NPT are different for curriculum and revision tutorials. Assessment scores demonstrate the efficacy of NPT. Future studies will focus on large and small group dynamics and objectively identify the optimum point in the curriculum at which NPT is most efficacious. This study was approved by the host institutions faculty-based ethics committee 23736.

A1 Anatomage: a worthwhile resource for self-directed learning in the medical school setting?

J. Allardyce, M. Goggin and K. McDermott

Graduate Entry Medical School, University of Limerick, Castletroy, Limerick, Ireland

Traditionally, anatomy has been taught predominantly with the use of cadavers. However, with the formation of new medical schools and the ever-increasing need for space in existing institutions, dissecting room facilities are often sacrificed, and other teaching modalities and technologies have been introduced in their place. The relatively recent development of interactive 3D learning technologies has been rapid, and current research in this area is very active. However, the literature has yet to progress to a point where there is definitive evidence weighing up the benefits and weakness of actual dissection vs. virtual dissection.

The Graduate Entry Medical School (GEMS), University of Limerick (UL) has purchased three Anatomage tables to facilitate Anatomy teaching for medical students in the university. Being an institution without access to a dissecting room, on site, Anatomage promises to provide access to 3D human anatomy to rival that of a cadaver. Anatomage claim 'The virtual cadaver is easy to interact with and manipulate via intuitive controls, which provides an advantage over other anatomy education solutions' (Anatomage website November 2016). Internationally, anecdotal evidence from users suggests that there is some merit to these claims.

Our aim, in the long run, is first to produce a comprehensive study of the benefits and weaknesses of Anatomage, compared with other teaching technologies in medical education, and cadaveric dissection. Secondly, we want to decipher the best way to incorporate Anatomage into the UL graduate entry medicine curriculum. Today, we present some preliminary data,

reflecting the opinions of first- and second-year graduate entry medical students about this new resource. Students were asked to fill out a short survey, following their weekly anatomy tutorial. The data collected from each cohort, first (Y1) and second (Y2) year were also compared, to look for differences between students who had access to Anatomage from the beginning of their studies (Y1) and those that did not (Y2).

A2 Audit of practical anatomy teaching methods to dental students

W. El Kininy and S. Davy

Trinity College Dublin, Dublin, Ireland

An audit of dental student practical anatomy teaching was performed at the anatomy department of Trinity College Dublin. The core curriculum for head & neck anatomy was used as the gold standard.

A group of 44 first-year Dental students were assessed after a routine bony osteology practical of the skull and mandible with a diagram-based picture assessment of the cranium and skull without any intervention during the practical – the practical was allowed to run as normal with tutorial-based learning without clear objectives laid out. The results were poor, with many student unable to identify many foramina or bones of the skull.

One week later, the same group of students was given clear, written objectives at the beginning of the same bony osteology practical. The practical was largely peer-to-peer teaching with tutorials or demonstration by the instructors provided as required, with the students following the objectives to completion. After the anatomy practical, the same diagram-based assessment was given to the students.

The results were significantly better, with an average 40% improvement in the knowledge of foramina of the skull in addition to the identification of the different bony landmarks.

The audit identified that student-led teaching with clear pre-marked written objectives before every practical with explanation and demonstration as required by the faculty produced more effective results in student comprehension of the anatomical topic at hand, in keeping with the objectives laid out in the core anatomy curriculum of Trinity College Dublin. This method has since been employed in every Dental student anatomy practical.

A3 Blended learning approaches for improving cell biology and embryology in year 1 medicine

H. Anscomb

College of Medicine and Dentistry, James Cook University, Townsville City Australia

Year 1, semester 1 of our MBBS program begins with a formal introduction in cell biology before this subject is expanded to look at the development and structure of the human body through systems. However, many students (an average 39% of students in 2013/2014) enter the program without having previously studied biological sciences in years 11 and 12. As such, students have reported that they struggle to keep pace in their study of this subject, and also find other aspects of anatomical study (embryology, gross anatomy and pathology) challenging later in the program.

Learning analytics data has indeed demonstrated that success in the initial cell biology module (exam score >74%/D) is linked to success in the gross anatomy module that follows ($P < 0.05$), irrespective of previous study. This strongly suggests that improved engagement and success within year 1 cell biology could lead to improved learning outcomes for students in other aspects of anatomical science, too.

Increased cohort size in the medicine program in the last 5 years has lead to decreased study time in laboratory classes for students, and in 2016 the cell biology teaching team developed a series of instructional cell biology videos to supplement students' private study. Preliminary data collected on the use of these resources have demonstrated a positive student response. Students reported that they found the ability to review materials outside of class highly beneficial and the end of semester exam results average was improved for this module (compared with 2014 results).

A project that aims to embed course-wide technology-enhanced learning approaches into the medicine program will begin in 2017 with the aim of improving student engagement and student performance. Based on our findings so far, an E-book for delivery of the cell biology module has been developed to allow students to progress their knowledge at their own pace, review materials, extend and better apply their knowledge.

Ethical approval was granted from James Cook University to study and present this data.

A4 Designing a clinical anatomy and pathology curriculum for 'transition' into clinical training, through the application of First Year Curriculum (FYC) principles

H. Anscomb

College of Medicine and Dentistry, James Cook University, Townsville City, Australia

Transition pedagogy utilises curriculum approaches that aid student engagement, success and retention. This is typically applied to First Year Higher Education (FYHE) students. However, the transition from preclinical to clinical studies within many medical programs represents a similar shift in the student learning experience. Here we describe the intentional curriculum design of clinical anatomy and pathology in year 4 of a 6-year MBBS program. The six First Year Curriculum (FYC) principles of transition, diversity, design, engagement, assessment and evaluation/monitoring have been embedded within the year 4 program.

Transitioning students from the learning of foundation science to the advanced application of scientific principles is organised through integrated teaching (and assessment) of clinical anatomy, pathology, clinical examination and evaluation. This learning is contextualised within various healthcare settings: primary health, hospital inpatient and outpatient and rural placement.

The integrated teaching design provides an explicit foundation for scaffolding the transition from the preclinical to the clinical context, while utilising curriculum design principles that promote active, collaborative learning and engagement that is consistent with the clinical workplace. Assessment is aligned to

this integrated approach through the use of case-based clinical practical exams, and theory papers.
Student diversity is primarily addressed through years 1–3 of the program, through the scaffolding of practical and generic skills that assist students with the demands of university study. This is supplemented in the clinical transition through a clinical mentoring program and a number of reflective activities that help students and staff to evaluate progress throughout the transition year and enable timely interventions (such as additional skills sessions) to be accessed.
Lastly, evaluation of the new curriculum is conducted regularly, especially during this implementation phase and is conducted through a combination of externally benchmarked, institution-wide, and college-specific feedback tools.
Having utilised FYC principles in the design of the year 4 program, further work is now needed to see how the curriculum impacts upon the transition to clinical study, the students' experiences, progression and the employer feedback. However, initial feedback has shown a positive improvement on students' experiences of the transition to clinical study.
No ethical approval was required.

A5 Blending near-peer teaching in anatomy education: a comparison of student attitudes towards integrated approaches

D. Sidebottom, A. Lowry, W. Parton, C. Harrison, M. Ahn, M. Myers, A. Elmansouri, S. Mcelligott, J. Stephens, S. Hall, E. Seaby, N. Carr and S. Border

Faculty of Medicine, University of Southampton, Southampton, UK

Near-peer teaching (NPT) is commonly used in anatomy education and serves to benefit the faculty, student teachers and learners. However, deciding how best to implement NPT inside the curriculum is still a matter of debate. At The University of Southampton we have experimented with an established NPT programme in neuroanatomy to ascertain whether NPT works best as face-to-face delivery or via online (virtual) delivery. Furthermore, we attempted to blend NPT contributions with established faculty-led approaches to create a hybrid NPT model and aimed to establish whether this approach to teaching was desirable.
A total of 168 BM5 second-year medical students studying a neuroanatomy module took part in the evaluation and were asked to rate a series of non-lecture-based teaching sessions using a Likert style survey instrument. The evaluation involved comparing anatomy sessions that were delivered either exclusively by NPTs, in collaboration with the faculty (hybrid model), or by NPTs online. Comparisons were also made with neuropathology sessions which utilised NPTs (hybrid model).
The results indicate that student-led face-to-face sessions were preferred to faculty-led practical anatomy sessions which contained some NPTs (the hybrid model) ($P = <0.05$) but not to either of the exclusively faculty-led anatomy workshops or lab-based demonstrations. There was no significant difference between face-to-face NPT and virtual online NPT delivery; both were rated very highly. Students favoured both of the exclusive NPT anatomy initiatives over the pathology hybrid sessions (NPT anatomy vs. pathology $P < 0.001$ and NPT online anatomy vs. pathology $P < 0.001$). There was no significant different between either . the anatomy or pathology hybrid sessions.
The results indicate that online NPT delivery is as effective as face-to-face NPT and that teaching collaborations between NPTs and faculty members in the same session are less desirable than either of the exclusively led faculty or NPT sessions. Reasons for this may include changes in the teaching dynamics which affect social and cognitive congruence, although further work is required to test this hypothesis. This work was approved by the Faculty Ethics committee.

A6 Building near-peer teaching into the anatomy curriculum – a national perspective from anatomy educators and students

J. Stephens,[1] S. Hall,[1] C. Harrison,[1] C. Smith,[2] W. Parton,[1] M. Myers[1] and S. Border[1]

[1]*Centre for Learning Anatomical Sciences, University of Southampton, Southampton, UK and* [2]*Department of Anatomy, Brighton and Sussex Medical School, Brighton, UK*

Near-peer teaching (NPT) is a highly valuable resource in medical education, with a long established history in anatomy. In the past its application has often been informal and unregulated; however, it is now becoming increasingly common for Faculties to work in partnership with students and deliver NPT as part of a formal curriculum.
Implementing an effective and sustainable NPT programme takes considerable planning and organisation, but if applied appropriately it can significantly enhance the student learning experience, improve knowledge and build a spirit of engagement among learners. Designing the 'most effective' NPT session is dependent on many factors and remains open to the individual educator's choice and experience.
As invited speakers at The Anatomical Society Summer Meeting, 2016, the authors delivered a workshop exploring how to imbed an effective and sustainable NPT programme into their anatomy teaching. Attendees had the opportunity to blueprint a working model of NPT into a hypothetical anatomy curriculum. Discussions focused on evaluating key criteria cited from the literature as a basis for determining best practice.
Seventy-eight participants took part in the workshop. Overall, the most popular design of the NPT programme was a combination of frontline and revision teaching (62%). Participants felt the teachers should attend formal training sessions (50%) with their experiences counting towards a fully accredited award/qualifications (38%).
The consideration that proved most controversial was the educational distance between the learner and the teacher. The most popular educational distance (41%) was +2 years, a finding consistent with recently published data. Applying NPT in musculoskeletal anatomy attracted the greatest majority of votes (66%), with participants reporting that the 'functional' nature of this body system lent itself well to this approach.
Benefits of NPT have been well documented within anatomical education. However, the exact format these sessions should take, remains open to individual judgment and experience. It is encouraging to see the majority of anatomy educators are open to the idea of incorporating NPT into curriculums, although reservations do persist in some areas, particularly with maintaining quality control. This was an educational audit from an AS workshop and therefore ethical approval was not sought.

A7 The benefits to student teachers involved in near-peer teaching

J. R. Stephens, S. Hall, M. G. Andrade, W. Parton, C. Harrison, E. Seaby, M. Ahn, S. McElligott, M. Myers, R. Parrot and S. Border

Centre for Learning Anatomical Sciences, University of Southampton, Southampton, UK

Near-peer teaching (NPT) is a well used pedagogical method in anatomy and is defined by the student being taught by other more advanced students. The benefits of NPT to the student learner have been widely investigated and include freer discourse and better resolution of weaknesses. The benefits to the student teachers have been theorised; however, currently there is no evidence to support them.

Student teachers were identified from the neuroanatomy near-peer teaching programme which has run for the last 6 years at the University of Southampton. A bespoke survey instrument which measured teachers' attitudes and academic rewards was distributed via email to the student teachers. The survey used 0–10 scores to measure the teacher attitudes, and free text to record the academic rewards.

Sixteen near-peer teachers completed the questionnaire (38% response rate) with six of them now being qualified doctors. The teachers rated NPT 8.2 out of 10 for helping them better understand the material, 8.8 out of 10 for aiding long term knowledge retention, and 7.3 out of 10 for how well NPT motivated them to continue studying the material after the teaching session was over. The teachers rated the enjoyment of NPT 9.1 out of 10 and how rewarding it was as 9.5 out of 10. Of the six qualified doctors, five of them gained points on the Foundation Programme Application System as a result of being involved in a NPT programme.

The results demonstrate that being a near-peer teacher is an enjoyable and rewarding experience for the teacher. It is also able to influence student attitudes to promote future engagement in teaching which will continue to benefit other learners even after the initial programme is finished. In addition to the changes in student teacher attitudes towards teaching, being involved in NPT has tangible rewards which improve future employability.

A8 Can near-peer tutors alter students' knowledge and perceptions of neuroanatomy? A pilot neurophobia intervention

W. J. C. Parton, A. Elmansouri, M. A. Myers, C. H. Harrison, E. G. Seaby, S. R. Hall, J. R. Stephens and S. Border

Centre for Learning Anatomical Sciences, University of Southampton, Faculty of Medicine Southampton General Hospital, Southampton, UK

Neurophobia is a worldwide phenomenon experienced by qualified physicians and medical students alike. Described by Josefowicz in 1994, neurophobia manifests as a lack of confidence in applying the fundamentals of neuroanatomy and neuroscience to clinical cases. Poor neuroanatomical knowledge and inadequate teaching have been highlighted as significant contributors to neurophobia. The challenge facing medical education practitioners is to establish efficacious and replicable techniques of targeting neurophobia at its source. Methods to combat neurophobia have included online educational resources, improved clinical teaching and national prizes. At the University of Southampton, neuroanatomy is taught during the nervous and locomotor 2 module (NLM2). Alongside faculty instruction, near-peer tutors (NP-t) have been integrated into the curriculum as small group facilitators, anatomy demonstrators and developers of online educational resources. NP-t are senior medical students 1 or 2 years advanced in their stage of training; the benefits of NP-t are well established in the literature. To our knowledge, no institution has evaluated the use of NP-t to combat neurophobia. Prior to the start of NLM2, 30 second-year medical students attended an introductory neuroanatomy teaching day, organised and delivered by fourth-year NP-t. Subjects included cortical topography, arterial supply, ventricles and basic neuroscience. Learners' attitudes and perceptions were measured using a Likert questionnaire (response rate = 100%; Cronbach-alpha = 0.65). Changes in knowledge were assessed using pre- and post-teaching pro-section-based assessments. Following teaching, only 40% of respondents felt that neuroanatomy was hard/very hard compared with other areas of anatomy. Additionally, mean learner assessment scores increased by 7.14 ($P < 0.001$; Cohens-$d = 2.38$) between the pre- and post-teaching assessments. Despite this, apprehension about NLM2 did not significantly change ($P = 0.329$), and in eight participants, apprehension increased. Results from this pilot neurophobia intervention show that, even though NP-t can positively influence students' perceptions of neuroanatomy, it has mixed effects on alleviating apprehension towards the NLM2 module. Participants in the study will be followed up over the course of the NLM2 module using formative and summative assessment scores to gauge the tangible effects of this study day on actual performance.

A9 Consolidating surface anatomy knowledge through anatomical body painting: a mixed methods evaluation

A. Venkatesh

Institute for Education in Medical and Dental Sciences, School of Medicine, Medical Sciences and Nutrition, University of Aberdeen, Aberdeen, UK

Teaching healthcare students surface anatomy (SA) is important because as practitioners, one of the ways in which they are likely to encounter anatomy is SA. Anatomical body painting (ABP) is an innovative SA teaching technique gaining popularity worldwide. Qualitative studies in literature describe its many benefits. We aimed to quantify this benefit during a Head and Neck SA session. We compared two groups of students; 19 postgraduate Dentistry, and 21 MBChB students (17 school leavers; four graduates) at similar stages of their Head and Neck Anatomy curriculum, taking an SA session with similar learning outcomes. MBChB students used plastic anatomical models and palpated for pulse points (control group). Dentistry students (ABP group) painted surface structures including pulse points using instruction sheets. A pre-test comprising seven multiple choice questions with a total of 15 correct choices was administered to both groups of students at the start of the session. The same test was administered at the end of the

session (immediate post-test). Completion was optional and consent implied. Net enhancement in SA knowledge was calculated by adding increase in correct options chosen to decrease in incorrect options chosen.
SA knowledge enhancement in the ABP group was 2.5 marks (out of 15), as opposed to our control group, who yielded a net decrease of 0.4 marks (increased incorrect choices probably through guessing). Focus group with ABP group students highlighted similar benefits as in the literature: ABP was fun-promoting active participation in a relaxed atmosphere. The visual and kinaesthetic component made it memorable. Anatomy staff members commented that ABP promoted appreciation of clinically relevant SA, active student participation and peer learning. A structured approach could help students overcome discomfort when palpating/painting on peers and the novelty aspect of ABP ensures participation.
For focus groups participants, consent to use of anonymised transcripts of recordings for dissemination was obtained. Based on this successful pilot, anatomical body painting was successfully introduced into the larger Year 2 MBChB class (class size ~170 students), receiving positive comments on course evaluation forms.
We conclude that ABP is an innovative and valuable teaching tool that promotes consolidation of clinically relevant surface anatomy.

A10 Enhancing student learning of human embryology with a prototype e-learning resource

I. Keenan,[1] Z. N. Solim,[1] S. Quigg,[2] J. Kerwin[3] and S. Lindsay[3]

[1]*School of Medical Education, Newcastle University, Newcastle upon Tyne, UK;* [2]*School of Biomedical Sciences, Newcastle University, Newcastle upon Tyne, UK and* [3]*Institute of Genetic Medicine, Newcastle University, Newcastle upon Tyne, UK*

Studying human development is important for student understanding of congenital malformations and an appreciation of the origins of adult anatomical structures, but the format of medical and biomedical curricula can limit the curricular time devoted to clinical embryology. Human development is a complex process that involves three-dimensional (3D) changes in morphology and gene expression over time. It is therefore necessary to develop embryology learning resources that are not only suitable for self-directed study activities but also provide students with an insight into the dynamic 3D nature of human development. Here we describe the next step in our development of a prototype embryology e-learning resource (PEER). This project has utilised material from the Human Developmental Biology Resource (HDBR), a collaboration between the Institute of Genetic Medicine, Newcastle University and the Institute of Child Health, University College London. Cross-sectional images were obtained from human embryos using optical projection tomography, thus avoiding the requirement to physically section each sample. Embryological structures were then highlighted using MAPAINT software. Having previously begun the process of identifying the developing gut tube in our previous work, we have now completed the painting of both the developing gastrointestinal and respiratory systems in embryos at Carnegie stages 13–18. Short video captures of rotating embryos with painted embryological features have now been created using AMIRA software by loading painted domains onto a virtual 3D embryonic model, which allows rotation in 360° and in 3D. These models have then been labelled using Adobe PHOTOSHOP to provide appropriate and specific course content for both lecture-based and computer practical teaching, in addition to self-directed learning.
We are currently developing our resource as an online interactive tutorial for students at Newcastle University that includes our 3D movies and labelled images. The tutorial will be evaluated through utilising pre-post testing and questionnaire methods in comparison with existing animated embryology resources. It is expected that the benefits to students of providing 3D and dynamic interactivity in a resource developed using real human embryos will serve to enhance learning of embryology compared with other tools.
Ethical approval has been granted for use of human embryos.

A11 Live tweeting at Anatomical Society Conferences: A short history of its rise and impact

I. Keenan,[1] S. Border[2] and C. Hennessey[3]

[1]*School of Medical Education, Newcastle University, Newcastle upon Tyne, UK;* [2]*Academic Unit of Medical Education, Faculty of Medicine, University of Southampton, Southampton, UK and* [3]*Division of Medical Education, Brighton and Sussex Medical School, Falmer, UK*

Social media (SoMe) are becoming fundamental components of networking and dissemination at academic conferences. We sought to investigate the role, usage and contributions of the social network Twitter at Anatomical Society (AS) conferences to identify the value of this approach to delegates, members and the Society.
Data show that Twitter usage (defined by the number of users and tweets using the conference hashtag) associated with AS conferences has increased since 2011. This reflects the overall increase in uptake of Twitter as an academic tool during this period. The first AS conference with associated Twitter activity was the Winter 2011 meeting, which included six tweets by three users. No further activity was identified at conferences until Winter 2013. This coincided with the appointment of an AS SoMe Editor, and achieved a total of 129 tweets by five users.
More recently, the increased availability and utility of analytics have allowed more detailed investigations of SoMe activity to be performed. Using data from the online Twitter event tool Eventhashtag.com, we describe detailed analytics of the AS Winter 2015 meeting (#anatsocwinter15, 135 tweets, 72 users) and the joint AS and British Association of Clinical Anatomists Summer 2016 meeting (#anatsocsummer16, 541 tweets, 198 users) and show a dramatic increase in activity between these events. The timing, types and impressions of tweets, and the number and activity of users have provided insights into usage and reasons for this growth. Replies to tweets did not show the same relative change, indicating that Twitter is used to network and share ideas rather than to initiate dialogue.
These findings show Twitter can be a valuable tool at conferences and demonstrate the importance of the role of official social media platforms for the modernisation of academic organisations. We conclude that Twitter can be beneficial for sharing current research and connecting with

peers and non-attendees as a basis for potential future collaborations, and can be effective for highlighting conferences and society memberships.
Ethical approval was not required as data were gathered from the public domain. The author (I.D.K.), as AS SoMe Editor, declares a potential conflict of interest.

A12 Teaching anatomists to draw: observational drawing as an educational approach

I. Keenan[1] and L. Shapiro[2]

[1]*Anatomy and Clinical Skills Centre, School of Medical Education, Newcastle University, Newcastle upon Tyne, UK and* [2]*Lateral Leap Drawing Education, Cape Town, South Africa*

Educational theory and research evidence have provided support for the notion that observational drawing can be valuable for anatomy learning. While many people claim they cannot draw, and believe that drawing is only for the talented or gifted, the act of drawing is fundamentally the making of marks on paper. As such, an individual can be trained to make marks that correspond to and describe the 3D form of an object.
Leonard Shapiro is a drawing teacher specialising in observation. We present a description of his drawing workshops designed specifically for academics and students of anatomy and held previously in South Africa at the Faculty of Health Sciences, University of Cape Town. The primary purpose of his workshop is the deep observation of anatomy, achieved through a specifically designed haptico-visual observational drawing (OD) technique. This novel method aims to increase the perceptual understanding and cognitive memorisation of the form of an object through multi-sensory observation including touch, manipulation and sight, and the simultaneous drawing of a 3D object onto a 2D surface. After studying an object using observational drawing, the drawer-observer should be able directly to retrieve the visuospatial appearance of the object from memory without actually viewing it. This approach is therefore ideal for learning and understanding the form of anatomical and biological structures.
We aim to provide OD workshops at Newcastle University in 2017 for UK-based anatomy instructors. Workshop participants will enhance their knowledge of anatomy through developing their observational skills, in addition to gaining an awareness of this approach for use in their own future teaching practice. We intend to perform research evaluations to identify the nature of learning through observational drawing and the educational value of our approach. In our recent publication, we describe the development and evaluation of a novel artistic learning process observe-reflect-draw-edit-repeat (ORDER). Furthermore, we anticipate combining the most powerful aspects of OD and ORDER to develop an improved observational drawing technique that can greatly enhance anatomy learning for both instructors and students alike.
We declare a conflict of interest with respect to Leonard Shapiro gaining financially from workshop facilitation.

A13 How well do clinical students know the dermatomes?

A. D. Walker, D. Challoumas, J. Lunn and C. Brassett

Human Anatomy Teaching Group, Department of Physiology, Development and Neuroscience, Downing Site, University of Cambridge, Cambridge, UK

Familiarity with the dermatomes is important in clinical practice, especially in determining the level of compression in radiculopathy or spinal cord injury. This study aims to assess the knowledge of dermatomes in 177 fifth-year medical students at the University of Cambridge. At an anatomy revision session, they first completed a questionnaire about relevant teaching and their perceived confidence in knowledge of the dermatomes, from 0 (not at all confident) to 8 (very confident), and were then asked to draw anterior and posterior dermatome maps on a diagram. No opportunity for revision was given and the task had to be completed in 5 min. Data were tabulated and analysed using Microsoft Excel 2013 and Stata/IC14. Results showed that anterior dermatomes were attempted by 168/178 (94%) students and posterior dermatomes by only 36/178 (20%). Of these 36, 17 labelled dermatomes above T4, but without areas supplied by C5–8. The following regional dermatomes were correctly drawn: upper limb (with the middle finger as C7 and others arranged accordingly) by 58%; trunk (with the nipple at T4 and umbilicus at T10) by 44%; lower limb (with L4 and L5 on medial and lateral calf, respectively, and S1 on plantar foot and/or posterior calf) by 25%; and perineum (concentric circles with S3, S4 or S5 represented) by 60% of students. The mean 'perceived confidence' score was 3.64/8 (SD 1.58). This was significantly higher in students who correctly traced upper limb dermatomes (3.88 vs. 3.32, $P = 0.019$). It was also higher for students who correctly traced lower limb, trunk and perineal dermatomes, but the differences were not statistically significant. Students who had revised dermatomes in the past 6 months and those who had been taught in the past 12 months were more likely accurately to label all regional dermatomes, although this did not reach statistical significance. Overall, these results would indicate that medical students lacked confidence in their knowledge of dermatomes. Possible reasons include the existence of conflicting dermatome maps, difficulty in appreciating the importance of dermatomes, and lack of use prior to clinical placements. Recommendations for ways to improve this knowledge in anatomy teaching will be discussed.

A14 Smartphone apps; will knowledge of anatomical variation be lost?

S. Davy, M. Coalter, D. Lennon, W. El Kininy and D. Barry

Trinity College, Dublin, Ireland

In the current technological age, increasing numbers of students are using modern learning resources such as smartphone apps to learn anatomy. However, those who use smartphone apps as their primary source of anatomical knowledge may miss out on extra details only covered in anatomical textbooks and scientific papers. The aim of this

study was to critically assess depth of coverage of anatomical variation offered by modern anatomy resources.
Upon discussion with senior anatomy lecturers at our institution, nine distinct clinically relevant anatomical variations were chosen as tools to assess various learning resources. Each resource was graded on a scale of 0–3 depending on its depth of coverage of variant anatomy. A total was then accumulated to give each resource an overall score.
Peer-reviewed scientific papers scored highest, followed closely by Gray's Anatomy. Smartphone apps which incurred a charge, scored moderately, whereas free apps performed poorly.
In conclusion, students are increasing their use of modern learning resources to study anatomy. However, we have shown that smartphone apps either omit or describe anatomical variation poorly, and perform poorly compared with textbooks and scientific papers. Therefore, students who use apps may lack knowledge of anatomical variation. We suggest that students should be advised to study anatomy from comprehensive learning resources.

A15 Spatial awareness training in first- and second-year medical students

R. Gonzales and C. F. Smith

Brighton & Sussex Medical School (BSMS), Medical Research Building, University of Sussex, Falmer, UK

The goal of this study is to understand the factors that affect learning of anatomy and surgical skills in medical students. In a longitudinal study, 29 medical students received training on spatial ability. Their performance was measured before and after the training intervention and compared with that of 79 of their classmates, who participated as the control group. In addition, 92 students participated in an initial spatial ability evaluation. The spatial ability test measured the capacity to understand and remember the three-dimensional relations of objects, a useful skill to navigate anatomical structures and the surgical field. The training workshops reviewed elemental concepts on spatial awareness with hands-on practice of spatial skills. Results showed that all participants on average tended to gain high spatial ability scores (mean = 65.5%, median = 70, SD 18.8). Female students scored significantly less than males ($P < 0.05$: Females median = 60, Males, median = 75). All of those who took the test twice, improved significantly, with higher scores on the second test (median = 75) than on the first (median = 70) ($T = 2703$, $P < 0.001$). Students who received training obtained higher scores in spatial ability (mean = 8.45, SE 1.98) compared with the control group (mean = 4.60, SE 1.64). This difference, 3.8, was not significant ($P = 0.19$); however, it does represent a small effect ($d = 0.27$). The effect was larger in males (intervention $r = 0.86$, control: $r = 0.36$) than in females (intervention $r = 0.47$, control $r = 0.25$). Further spatial awareness training might enhance the effect of non-tested factors that improve spatial skills in anatomy students.

A16 The Twitter Polling Tool: can it be used to increase anatomy exam scores?

C. Hennessy

Department of Anatomy, Brighton and Sussex Medical School, University of Sussex, Brighton UK

The number of anatomy educators using Facebook and Twitter as an educational adjunct is on the rise and there have been several reports on how these platforms have had positive outcomes: increasing student engagement with the subject of anatomy, facilitating communication with educators and reducing student anxiety levels in the lead up to assessments by offering learning support information. There have been fewer reports on whether these platforms impact on student's anatomy knowledge. One previous investigation demonstrated that the frequency of student engagement with a dedicated Twitter hashtag did not increase their exam scores; however, the methodology did not provide specific learning feedback opportunities to students. Since then, Twitter has introduced a polling tool. This project investigated whether the polling tool could effectively be used to deliver regular MCQ-style questions (followed by feedback) to students and whether students' use of this tool had an effect on exam scores.
Second-year medical students enrolled on the Musculoskeletal and Immunity Module 2015 ($n = 140$) at Brighton Sussex Medical School were offered the #m204anatomy Twitter hashtag as a learning support tool which included weekly Twitter polls questioning students' anatomy knowledge. The correct answer/voting option was subsequently tweeted by the investigator offering feedback to students. At the end of the module students were invited to complete a questionnaire asking whether they had answered Twitter polls throughout the module. Students were also asked for consent to use their exam scores to compare against their use of Twitter polls. Ethics approval to carry out this investigation was granted by the Research Governance and Ethics Committee Ref No: 16/001/HEN. Ninety-three students successfully completed the questionnaire and gave consent. The 62% of students who did not use the Twitter polls; their mean anatomy exam score was 59.7%. The 38% of students who did answer the Twitter polls had a mean exam score of 67.2%, significantly higher ($P = 0.012$) than that of those who did not use the Twitter polling tool. This indicates that the Twitter polling tool can be used as a formative assessment tool to prepare for examinations.

A17 Use of Complete Anatomy Lab as a supplementary learning resources: students' perspective

T. Kylkilahti, M. G. Sagoo and R. Wingate

King's College London, London, UK

Complete Anatomy Lab is an online anatomy software currently available on iPads. It combines an interactive three-dimensional human anatomy model with an option to create content ranging from customised models to recordings, teaching material and quizzes.
The purpose of this study is to test the efficiency of Complete Anatomy as a supplementary study resource. All the existing

functionality of the software was analysed in detail. To obtain students' views, nine students from biomedical and medical cohorts at King's College London completed a sample tutorial created using the software, and answered a survey regarding its use and effectiveness as an anatomy learning resource.
The majority of the students gave positive feedback on the functionality of the software, particularly its customisability and muscle motion animations, which is a major technology uplift of this software compared with the other anatomy online tools available on the market.
Although the response was positive, the main issues of the software are not in its content or features but rather its accessibility and transferability of the material produced. The software can only be accessed on iPads and the content created within the software cannot be shared outside the application without losing its interactivity. This limits its usefulness in a multi-platform learning environment.

A18 Using classic and technology-based methods to enhance the student learning experience in histology

S. Morton,[1,2] T. S. Cecot,[1,2] Z. Bayram-Weston,[1,2] S. Border,[1,2] S. I. Paterson,[1,2] J. R. Skidmore[1,2] and A. S. O'Malley[1,2]

[1]*Centre for Learning Anatomical Sciences, Faculty of Medicine, University of Southampton, Southampton, UK and* [2]*Centre for Comparative and Clinical Anatomy, University of Bristol, Bristol, UK*

Histology is taught across the first 2 years of the BM5 medical curriculum at the University of Southampton; however, poor student feedback encouraged redevelopment. Histology was changed from a stand-alone subject to one that is fully integrated with gross anatomy. ePracticals were introduced, for completion before anatomy practicals. Two-hour microscope-focused histology practicals were replaced with 1-h interactive workshops, allowing students to consolidate their knowledge through team-based, clinically oriented tasks, and iPads were used to provide instant feedback on students' progress.
The aims of the study were to quantify student attitudes towards the new format of histology teaching, and to investigate which of the elements that were changed, may have affected their learning experience. Evaluation data from two Year 1 (old format) and two Year 2 (new format) modules were collected after each module using a five-point Likert scale survey. At the end of Year 2, students ($n = 203$) were asked to compare their histology learning experiences in Year 1 with Year 2 using a 10-point Likert scale survey, which covered their attitudes towards histology as a subject and the main elements of teaching that were changed: integration with anatomy, its relevance to medicine, team-based learning approaches, and feedback. Kruskal–Wallis ANOVA on Ranks and Dunn's pairwise comparison were used to test for differences in perception between the Year 1 and Year 2 formats. Spearman tests were used to identify correlations between students' attitudes and the changed elements. In the module evaluations, significantly more students ($P < 0.05$) rated Year 2 histology positively (68.6 and 71.0%) compared with Year 1 (31.8 and 46.3%). Student attitudes towards histology were significantly more positive after Year 2 than after Year 1 (median 6/10 vs. 3/10; $P < 0.05$) as was their opinion of the teaching (median 7/10 vs. 4/10; $P < 0.05$). There were correlations between attitude after Year 2 and opinions of teaching ($R = 0.610$; $P = 2 \times 10^{-7}$), integration ($R = 0.658$; $P = 2 \times 10^{-7}$) and perception of feedback ($R = 0.642$; $P = 2 \times 10^{-7}$). The opinion of the teaching also correlated with integration ($R = 0.730$; $P = 2 \times 10^{-7}$).
With emerging technologies and an expanding curriculum, there is more reason to modernise histology teaching through a blended approach. Through integration with anatomy and increased feedback, student attitudes towards histology have been significantly improved.

B1 'Bend it like Ronaldo'. Deriving a practical anatomically correct standing posture from sports, physical fitness disciplines and performance arts

M. H. Bob and D. Pepine

'Iuliu Hatieganu' University of Medicine and Pharmacy, Cluj Napoca, Romania

The purpose was to elaborate a set of practical steps that allows assuming and maintaining an anatomically correct standing posture for subjects with an orthopaedically normal locomotor system, requiring no particular exercise equipment, environment or external devices. Balanced physical training brings standing posture closer to the anatomical ideal, as seen in professional athletes. Postural exercises available to the general population involve fairly complex routines which ignore the capacity of the subject to assume the correct standing posture.
Inclusion and exclusion criteria for physical posture approaches were established according to the scope of the study.
Basic postures in various sports (football, basketball, body building and fitness, boxing, fencing, swimming), physical fitness disciplines (medical gymnastics, Pilates, Ashtanga Yoga) and performance arts (classic ballet) were documented in theory and practice under professional guidance.
One specific positioning for each weight-bearing segment was selected, respecting the inclusion and exclusion criteria.
The anatomically correct posture identified involves the performing of, in order: Feet together or shoulder-wide, knees hyperextended, pelvis in neutral (ASIS and pubic symphysis in the same frontal plane), lowered scapulae ('led scapulae'), and head in neutral (Frankfort line kept horizontal). Next level of proficiency includes adduction of costal arches (the 'subxyphoid compass pinch'), making respiration possible only in the abdominal mode.
Professional sports, physical fitness disciplines and performance arts studies can yield information to formulate an anatomically correct standing posture with practical application. The necessary steps require knowledge of several superficial anatomical features and everyday practice. Future directions of study include measuring the height difference in relaxed vs. anatomically correct posture, and its correlation with vertebral column curvature modifications through photogrammetry.

B2 A cadaveric study of morphological relationship between superficial cortical and deep grey matter structures in adult human brains

O. O. Azu, E. C. S. Naidu and E. Y. Haghegh

Discipline of Clinical Anatomy, School of Laboratory Medicine and Medical Sciences, Nelson R Mandela School of Medicine, University of KwaZulu-Natal, South Africa

While various neurodegenerative diseases affect the cortical mass (and deep grey matter) differently, finding an optimal and accurate method for measuring thickness and surface area of the cerebral cortex remains a challenging problem due to the highly convoluted surface of the cortex. We investigated the superficial and deep grey matter thickness and surface area in a sample of cadaveric specimens at the Discipline of Clinical Anatomy, Nelson R Mandela School of Medicine, University of KwaZulu-Natal, South Africa, to provide some clue as to possible variations in these parameters.

Sixty brain samples (Ethics: BE 134/14) were uniformly sectioned at 5-mm thickness, and eight slices containing the deep nuclei were taken from each brain and stained by Mulligan's technique. Thickness was measured at selected angles of 0°, 45°, 90°, 135° and 180° for both right and left cerebral hemispheres. The cortical thickness and surface area of selected slices for both the superficial cortex and the corresponding deep nuclei were measured.

Mulligan's stain produced good grey matter differentiation and clear images that enabled manual delineation of structures. There was rightward asymmetry of cortical thickness of the selected slices at the suggested angles which corresponded to structurally and functionally important brain regions. There was a positive correlation between the mean surface area of superficial cortex and deep nuclei across the regions of interest (ROI).

Baseline data from 55 brain samples provided a range of means and 95% confidence intervals for the three parameters of cortical thickness, cortical surface area and surface area of deep nuclei to be made for a reference table comprising eight coronal slices taken at five angles. This allows an objective assessment of thinning of the cortex or loss of deep grey matter to be made from measurements of the same parameters for the equivalent slices from a postmortem brain slice or an appropriate radiographic image.

B3 Pancreatic histomorphology in rat model of diabetic antiretroviral therapy: the role of *Hypoxis hemerocallidea* adjuvant

O. O. Azu, S. Mdlalose, I. O. Onanuga, A. I. Jegede, U. Offor and E. C. S. Naidu

Discipline of Clinical Anatomy, School of Laboratory Medicine and Medical Sciences, Nelson R Mandela School of Medicine, University of KwaZulu-Natal, South Africa

Metabolic perturbations including derangement in glucose levels are rife following highly active antiretroviral therapy (HAART). Finding alternative adjuvants for mitigating this occurrence has remained the drive of researchers in HIV medicine.

We therefore tested the hypothesis that *Hypoxis hemerocallidea* (Hyp) mitigates pancreatic distortions in HAART.

Following ethical clearance (056/15/Animals) 80 adult male Sprague-Dawley rats, aged 9–10 weeks (188.98 $\pm$ 4.5 g) were grouped into experimental and control (groups A–K) and treated according to approved protocols for 2 months. Tissues and samples were collected at end of treatment and results analysed.

Derangement in glucose levels was maximal in diabetic controls with aggravation in HAART and Hyp groups. This tallied with bodyweight changes. White blood cell (WBC) counts were significantly reduced ($P < 0.05$; 0.001) in diabetic groups treated with Hyp, HAART and melatonin but RBC count was elevated ($P < 0.05$) in diabetic groups treated with melatonin and Hyp low dose. Hb levels, haematocrit and platelet counts were not significantly different from controls ($P > 0.05$). Pancreatic histology showed depletion in the number of islets of Langerhans in HAART group, diabetic+HAART, melatonin and Hyp, respectively.

We conclude that hypoglycaemia following HAART and Hyp adjuvant therapy warrants further investigation.

B4 Renal histological changes in spontaneously hypertensive rats following adjuvant use of *Hypoxis Hemerocallidea* in highly active antiretroviral therapy

O. O. Azu,[1] T. M. Nkwagatse,[1] A. I. Jegede,[1,2] U. Offor,[1] I. A. Peter,[1,3] E. N. Akang[1,4] and E. C. S. Naidu[1]

[1]*Discipline of Clinical Anatomy, School of Laboratory Medicine and Medical Sciences, Nelson R Mandela School of Medicine, University of KwaZulu-Natal, South Africa;* [2]*Department of Anatomy, Ladoke Akintola University of Technology, Ogbomoso, Nigeria;* [3]*Department of Anatomy, Faculty of basic Medical Sciences, University of Uyo, Uyo, Nigeria and* [4]*Department of Anatomy, College of Medicine, University of Lagos, Lagos, Nigeria*

Although morbidity and mortality have been significantly reduced following the introduction of highly active antiretroviral therapy (HAART), there still remains the unexplained issue of renal toxicities, especially with co-existing hypertensive changes in HAART.

We hypothesise that hypertension does not affect renal architecture following HAART.

Thirty male, spontaneously hypertensive rats (SHRs) and five Sprague-Dawley controls, 16 weeks old weighing 242.6–286 g were used. The protocol was approved (008/15/Animal) and animals divided into seven groups (A–G) of five rats each; treatment was according to protocols. At the end of experiment, samples were collected and kidney tissue analysed.

Differences in values for Serum Cr (reduced) and BUN (increased) in hypertensive controls were non-significant ($P > 0.05$) compared with normal controls. BUN was significantly reduced ($P < 0.05$) following Hyp_{200} treatment. Renal histological changes showed architectural denudation in glomerular capsule with loss of integrity in Bowman's space in HAART group. Capsular thickening was exaggerated in HAART+Hyp_{200}. Hyp_{200} groups also showed complete loss of glomerular apparatus with infiltrations on interstitial mass.

We conclude that HAART under hypertensive conditions is deleterious to renal morphology and Hyp does not mitigate these changes.

B6 Cadaveric study of the anatomical relationship between femoral artery and vein with implications for catheter placement

N. Slim, A. D. Walker, A. Pilarski, C. T. West, M. Singh, J. E. Lawrence, C. Brassett and M. E. Gaunt

Human Anatomy Teaching Group, Department of Physiology, Development and Neuroscience, Downing Site, University of Cambridge, Cambridge, UK

Knowledge of the anatomical relationship between femoral artery (FA) and vein (FV) is essential to minimise complications in procedures involving catheterisation of either vessel. Inadvertent puncture of the FA while cannulating the FV for catheter ablation in the treatment of atrial fibrillation may lead to pseudoaneurysm formation with life-threatening rupture. Typically, the FV is described as entering the apex of the femoral triangle (FT) posterior to the FA, and ascending to lie medial to the FA at the inferior border of the femoral sheath. However, *in vivo* ultrasound studies and reviews of computed tomographic scans suggest that this arrangement is inconstant. This study aims to investigate the relationship of the femoral vessels in 40 cadavers. The femoral sheath was opened to expose the femoral vessels, and the origin of the profunda femoris artery (PFA) was also identified. The following measurements were taken using callipers: from the inguinal ligament (IL) to the distal point of the PFA origin; and from this point to where the FV lies posteriorly to and is entirely overlapped by the FA. Results showed that in all cases, the FV lay medial to the FA at the IL. At a mean distance of 107 mm (SD 20 mm) from the IL, the FV lay deep to the FA, although in 4/40 (10%) cases the vessels ran parallel to each other throughout the femoral triangle. The PFA branched laterally from the FA at a mean distance of 58 mm (SD 16 mm) from its origin, although in one case, it arose directly from the external iliac artery and ran parallel to the FA below the IL. In conclusion, this cadaveric study confirms the typical arrangement of the femoral vessels, with a small number of cases where the FV did not lie posteriorly to the FA at the apex of the femoral triangle. We suggest an optimal vascular access window within 40 mm of the IL, both to avoid the area of overlap between FA and FV, and to minimise confusion between FA and PFA. It is recommended that ultrasound visualisation should be performed prior to attempting catheter placement into the femoral vessels. All donors gave their consent under the Human Tissues Act 2004.

B8 Effect of ageing on large and medium sized vessels of choroid: a human study

C. Kumari and T. C. Nag

Department of Anatomy, AIIMS, New Delhi, India

The choroid of the eye consists a capillary bed, called the choriocapillaris, medium-sized vessels (arterioles and venules) of Sattler's layer, and large vessels (arteries and veins) of Hellar's layer. The choriocapillaris has been extensively studied due to its close proximity to retinal photoreceptors that receive nourishment from this layer and undergo degeneration in ageing and diseases. However, there are limited studies on the vessels of Sattler's and Heller's layer. Here, we examined the morphology of these vessels in normal ageing, with the aim to analyse medium and large vessel changes in healthy human choroid with age.

Donated eyes ($n = 15$, age of donors: 35–83 years) were collected from the National Eye Bank, AIIMS (All India Institute of Medical Sciences) and preserved in Karnovsky's fixative. Choroidal tissue from the submacular zone and mid-peripheral zone (3–5 mm from macular border) was separated and processed for transmission electron microscopic study.

There were changes in the components of the smooth muscle cells (SMC) of arterioles and arteries. The SMC changes (degeneration/loss of mitochondria and filaments) were associated with progression of age (>80 years of age). The loss of SMC thin filaments resulted in a decreased density of sub-plasmalemmal plaques from 4 to 1 μm^2 in 24-year-old vs. 83-year-old donors (25% decrease). The thickness of the SMC basal lamina of aged arterioles and arteries was increased two-fold from 274 nm in 24-year-old to 550 nm in 83-year-old donor choroids. In contrast to the SMC basal lamina, the basal lamina of the endothelial cells of the vessels showed a decrease in thickness beyond 80 years of age, from 275.17 nm in 24-year-old to 173 nm in 83-year-old donor choroids.

Changes noted in the vessels should affect the blood flow and vascular tone in the choriocapillaris, which might alter their physiological functioning and ultimate death.

Note: All tissue materials were handled following the tenets of the Helsinki Declaration, and after receiving approval from the institute human ethics committee (IEC/NP-318/2012/RP-25/2012).

B10 Macroscopic and microscopic analysis of the posterior cruciate ligament tibial extension on human Thiel-embalmed knees

M. Alobaidy,[1,2,3] D. Nicoll[1] and T. Wilkinson[2]

[1]*Department of Orthopaedic and Trauma Surgery, School of Medicine, University of Dundee, Dundee, UK;* [2]*Centre for Anatomy and Human Identification, University of Dundee, Dundee, UK and* [3]*Department of Anatomy, Faculty of Applied Medical Science, Umm AlQura University, Makkah, Saudi Arabia*

The posterior cruciate ligament (PCL) is variable in terms of its origin and termination. Understanding the morphology of the PCL and its tibial attachment is important in PCL reconstruction surgery. Sixty-six Thiel embalmed knees were dissected from 17 male and 16 female cadavers, average age 78 years (range 47–99). The PCL tibial attachment extensions inferior to the tibial plateau were measured using high-accuracy calibrated digital Vernier callipers at 0.01 mm. Cronbach alpha coefficient reliability tests showed no difference (0.91) in intra- or inter-observer measurements. A histological analysis was carried out to confirm the inferior tibial attachment of the PCL. The attachment was macroscopically and microscopically examined, and found to terminate in a fan shape, with some fibres blending with the periosteum of the posterior tibial surface. The mean tibial extension below the articular cartilage was

29.5 ± 8.9 mm (range 10.0–47.0), whereas tibial extension below the PCL facet was 15.5 ± 6 mm (range 3.0–29.6). There was no significant difference between the measurements for female and male specimens, or between right and left knees. Histological examination confirmed that the fibres extending inferior to the usual insertion level were posteromedial bundle fibres. These fibres may play an additional role in supporting and stabilising the knee posteriorly, and are anatomically important to consider during ligament reconstruction. Relevant consent was obtained at the time of body donation in accordance with the Human Anatomy (Scotland) Act 2006.

B11 Qualitative analysis of the developing human lumbar vertebral column: radiographic analysis of a fetal and perinatal sample

S. Goodchild, C. Cunningham and S. Black

Centre for Anatomy and Human Identification, University of Dundee, Dundee, UK

The human adult lumbar vertebral column plays an important role in weight-bearing and weight transmission between the trunk and the lower limb. Although there is a plethora of research focusing on the trabecular architecture within the adult lumbar vertebral column, the development of this trabecular architecture is less well established. This study is a preliminary investigation focusing on the qualitative analysis of the developing trabecular and cortical architecture of the human lumbar vertebral column. A total of 14 specimens, equating to 67 human fetal and perinatal lumbar vertebrae, were utilised in this study. Elements were radiographed and colour gradient-mapped to analyse the intensity pattern within the developing lumbar centra and hemiarches. In both age cohorts, centrum shape was found to be most variable at the posterior border, which was either billowed or flattened. An area of high radiographic intensity was observed within the centra that became increasingly organised with age. By the perinatal period, this area of radiographic intensity had developed into an organised, symmetrical pattern. Single, or multiple, anterior and posterior projections were present, along with paired anterolateral and posterolateral projections. In the neural hemiarches, radiographic intensity decreased with age, with fetal specimens exhibiting radiographic intensity within the future laminae and pedicles. In perinatal specimens, radiographic intensity was mostly found occurring in the future laminae. The variable shape of the posterior margin and associated variation of the anterior and posterior projections are most likely related to the developing vasculature, with ossification extending into areas where no vascular structures are present. The paired anterolateral and posterolateral projections are potentially caused by the proximity of the developing neural hemiarches and associated developing musculature and ligamentous structures. This forms part of a larger study that aims to document and understand the developing trabecular architecture in human lumbar vertebrae using μCT. No ethical approval was required.

B12 Relationship between the great auricular nerve and the modified Blair's incision: a cadaveric study

S. Theivendran, A. Alade and M. G. Sagoo

King's College London, London, UK

The use of the modified Blair's incision in parotid surgeries is common, running between the preauricular and cervical creases. This approach provides great visibility during surgery but can disturb the function of the great auricular nerve (GAN), giving rise to numbness around the lobule of the ear and the skin overlaying the parotid. The aim of the study was to record the relations of the GAN to bony landmarks and the incision line, enabling us to predict a revised incision path.

Both right and left sides of five formalin-fixed cadavers were used to obtain 10 sets of measurements, recording the distribution GAN in relation to two bony landmarks: the angle of the mandible and mastoid process. Measurements obtained illustrate the distance of both the stump GAN and its branches in relation to the bony landmarks. A modified Blair's line on clear film was overlaid onto the dissected area to observe its relation to the nerves.

The emergence of GAN occurs two-thirds of the way up SCM, at an average of 53.2 mm (± 6.8) from the angle of mandible and 69.2 mm (± 9.0) from the mastoid process. Regardless of the angle of the head, the mean distance of the mastoid process to the angle of the mandible is 55.6 mm (± 3.9). The branching of the GAN occurs almost equidistant from both the mastoid and the angle of the mandible, at 33.8 and 30.00 mm, respectively. The distal part of the incision crossed the GAN in 9/10 of all specimens. Although we are limited by a small study size, the measurements obtained fall within a relatively narrow range, all under 1 cm. All cadaveric studies conducted comply with the Human Tissue Act 2004.

The branching of the nerve occurs about 3 cm from both the mastoid process and angle of mandible; this may indicate the critical point to avoid during surgery. The frequent crossing of the distal part of the modified Blair's occurs because it runs parallel to the clavicle from the mastoid. Instead, following a perpendicular path from the mastoid process will avoid the vicinity of GAN, minimising any damage.

B14 Spacial variations in the arrangement of the facial nerve within the parotid gland and its relationship to the retromandibular vein, and the analysis of its anatomic significance to the extra-oral surgical approaches to the mandibular condyle

W. El Kininy, S. Davy, D. Barry and L. F. A. Stassen

Trinity College Dublin Anatomy Department, Ireland

The mandibular condyle presents a surgical region fraught with risks to the facial nerve. In traumatic settings, the mandibular condyle fracture is a controversial area of discussion in terms of the most appropriate management – either open or closed reduction. Historically, the majority of these fractures were managed with closed reduction; however, there is a growing body of evidence to support open treatment.

A gap exists in the literature as to the impact of facial nerve variations or variations of the facial nerve to the retromandibular vein on the various approaches to the mandibular condyle. The current extra-oral surgical approaches grossly rely on the normal position and divisions of the facial nerve and the retromandibular vein as a landmark and guide.
As part of an MD on the impact of facial nerve variation in mandibular condyle surgery, using 13 donors (eight female, five male) donated as part of the anatomy teaching programme in the Department of Anatomy, TCD – we are in the process of dissecting 26 parotid glands to elucidate any variations within this Irish population of donors. Fourteen parotid glands dissections have already been performed.
Provisional results have shown significant variations: 21% have shown variation in the relationship of the facial nerve to the intra-parotid veinous structures. In one case, the facial nerve main trunk was found deep to the superficial temporal vein (STV), exiting between it and the maxillary vein (MV), continuing superficial to the MV. This particular variation has not been reported previously in the literature.
We report our provisional results and discuss the current literature on facial nerve variation.
Consent for dissection of the human donors with imagery of their facial nerves was obtained for each individual donor under the voluntary consent process provided by each donor when signing onto the Trinity College Anatomy Department donor programme prior to their death. The consent form explicitly states that dissections for teaching and research along with appropriate imagery are allowed.

B15 The sternalis – more common than we believe? A cadaveric study

J. V. Krishnan,[1] J. Brittain,[1] J. Gabriel,[1] T. Murphy,[1] M. D. Reid,[1] V. Shaw[2] and C. F. Smith[2]

[1]*Nuffield Health Brighton, Brighton, UK and* [2]*Medical Education, Department of Anatomical Sciences, Brighton & Sussex Medical School, Brighton, UK*

Introduction: The sternalis is a normal anatomical variant that is typically poorly understood and not taught at undergraduate level. It is a thin, para-sternal strap muscle that originates from the upper sternum and infra-clavicular region with variable insertion points from the pectoral fascia to the rectus sheath. Its innervation and function are also relatively uncertain. The muscle can be found unilaterally (4.5% of cases) or bilaterally (1.7% of cases). Great variation exists in the reported incidence rates between the sexes and also among the different ethnic groups.
Methods: During the preparation for undergraduate anatomy teaching, 18 cadavers were prepared for dissection with removal of the skin and superficial tissue of the chest wall. A review of the existing literature was then carried out to understand the incidence of the sternalis.
Results: Of the 18 cadavers, three were found to have this anatomical variation of the anterior chest wall, giving an incidence rate of 16.7%. The small sample size of this study is a noted limitation but the findings do suggest further evaluation would be provident.
Conclusion: The incidence of 16% in our small cohort suggests that sternalis may be more common than previously thought and should be included in more regular undergraduate teaching. The sternalis has significant clinical applications in both radiology and reconstructive surgery – its presence is potentially misdiagnosed as benign or malignant breast lesions during screening. Additionally, the muscle can potentially be used in head and neck, chest wall or breast reconstructive surgeries.
The findings from this cadaveric study also suggest that compiling a database to record the occurrence of sternalis muscles would be beneficial in producing more accurate epidemiological data.
No ethical approval was required for this study.

B16 The accessory obturator nerve: an cadaveric anatomical study with relevance to anterior and lateral approaches to the spine

M. Protas,[1,2] B. Gardner,[1,2] M. du Pless,[1,2] M. Loukas,[1,2] R. J. Oskouian[1,2] and R. S. Tubbs[1,2]

[1]*Seattle Science Foundation Saint Georges University: School of Medicine, St George's University, True Blue, St George's, Grenada, W.I. and* [2]*Seattle Science Foundation: Swedish Hospital, Seattle, WA, USA*

The accessory obturator nerve (AON) is a variant nerve that passes alongside the obturator nerve medial to the psoas major muscle. It travels over the pubis and then descends dorsally to the pectineus muscle, where it might innervate the hip joint, pectineus, or anastomose with the obturator nerve. During lateral or anterior surgical approaches to the lumbosacral spine, this nerve should be considered. To aid in the decision making, a review of past literature on the AON and an anatomical study were conducted. Thirteen studies analysing a total of 2102 lumbar plexuses were considered. Twenty (40 plexuses) fresh-frozen adult cadavers (12F/8M) were dissected by an anterior approach. The AON was found to arise most commonly from L2-L3 (4 plexuses), with a prevalence of 30% (8M/4F) on the left side (7L/5R). The average length from origin to superior pubic ramus was 14.5 cm with a diameter of 1.2 mm (range 0.8–1.5), with all nerves found to travel medially to the psoas major muscle, medial to the femoral nerve and lateral to the obturator nerve. Two terminal branches anastomosed with the anterior division of the obturator nerve, and eight plexuses terminated deep (n = 2) or superficial (n = 66) to the pectineus origin. Past literature shows that the average prevalence is 13%, with origin from the L3-L4 plexus (63.60%) being the most common and from L2-L4 (10.60%) the second most common. A higher prevalence was reported in females and on the left side. Terminal branches typically joined the anterior (14.30%) or posterior branch (4.65%) of the obturator nerve. Surgical approaches to the lumbosacral spine, whether anterior or posterior, should consider the presence of the AON so as to minimise injury to it.
No ethical approval was required to conduct the anatomical study.

B17 The anatomy of the lateral femoral cutaneous nerve of thigh and its implications in total hip arthroplasty: a cadaveric study

J. Bartlett and J. Lawrence

Department of Human Anatomy, University of Cambridge, Cambridge, UK

The lateral femoral cutaneous nerve (LFCN) is the main neurovascular structure at risk during the minimally invasive anterior approach (MIAA) for total hip arthroplasty (THA). In this cadaveric study, we aimed to quantify this risk by examining the course of the nerve and its branches in the lower limb.

Forty-five hemipelves from 39 cadavers were dissected. The LFCN was identified proximal to the inguinal ligament (IL) and its course in the thigh identified. The positions of the LFCN branches in relation to the MIAA incision were measured using Vernier callipers and branching variants categorised.

In all, 44% of nerves crossed the incision of the MIAA, 47 ± 28 mm from the proximal end of the incision. Of those that did not cross the incision, the average minimum distance between the nerve and incision was 14.4 ± 7.4 mm, occurring 74.0 ± 37.3 mm from the proximal end of the incision. The anatomy of the LFCN showed considerable variability, with only 29 nerves displaying classical branching, giving rise to femoral and gluteal branches around the level of the IL. In the remaining cadavers, three novel branching patterns were observed: late, primary femoral and trifurcate. In the eight 'late branching' nerves, the nerve bifurcated after passing beyond the greater trochanter. Six nerves offered no gluteal branch and instead continued down the anterior thigh. This was described as 'primary femoral branching'. In two instances, the LFCN gave rise to three equal trunks that continued distally – 'trifurcate branching'.

The LFCN is at high risk during THA using the MIAA and our study emphasises the need for meticulous dissection during this procedure. Additionally, we identified two anatomical variants of the LFCN at greater risk during the MIAA – the late branching and trifurcate variants. As late branching variants give rise to the gluteal branch more distally, they are more likely to be at risk of injury, as the gluteal branch traverses the thigh perpendicular to the incision line. Trifurcate branch variants also carry a higher risk of injury owing to the proximity of their 'middle' branch to the incision.

Ethical approval was obtained from the Human Tissue Authority.

B18 Biomechanics of osteoderms in a lizard skull – a preliminary finite element study

J. Xue,[1] A. Marghoub,[1] S. Bertazzo,[2] S. E. Evans[3] and M. Moazen[1]

[1]*Department of Mechanical Engineering, University College London, London, UK;* [2]*Department of Medical Physics & Biomedical Engineering, University College London, London, UK and* [3]*Department of Cell and Developmental Biology, University College London, London, UK*

Osteoderms (ODs) are bone-like-rich organs found in the skin of many reptiles. The underlying mechanisms of their formation are not well-known. However, it is likely that biomechanical forces may contribute to the formation of osteoderms.

The aim of this study was to develop a finite element model of a reptile skull (with osteoderms) and to investigate: (i) the pattern of stress distribution across the skull with and without the osteoderms; (ii) the effect of bone–osteoderm interface properties and (iii) the effect of changes in the mechanical properties of osteoderms.

A series of finite element models of an Ocellated lizard (*Timon lepidus*) skull were developed. Bone and osteoderms were modelled with isotropic material properties. The skull was loaded in a simplified loading condition, i.e. it was constrained at the occipital condyle and loaded in three biting positions. As quadrate movement (streptostyly) during biting in *Timon* is still a matter of debate, simulations were performed with a mobile and a fixed quadrate.

Removing the ODs led to an increase in the level of stress across the skull roof. Further models with a movable quadrate showed a higher level of stress in the nasal region and a lower level of stress in the parietal region (parietal region). Modelling the gradual fusion of the osteoderms to the bone revealed a gradual reduction of the stress across the skull roof and a gradual increase of the stress within the osteoderms. A similar result was obtained when a fused interface between the bone-osteoderms was assumed and the mechanical properties of osteoderms were gradually increased (i.e. they became more bone-like).

These preliminary results suggest that the formation of osteoderms and their gradual fusion to the underlying bone might be the result of a local adaptation to relieve mechanical stress on the underlying bone. No ethical approval required.

C1 Alteration of haemodynamics causes differential expression in coronary vasculature-related genes of avian embryos

C. Perdios, K. L. Pang, M. Parnall and S. Loughna

School of Life Sciences, University of Nottingham, Nottingham, UK

In embryos the epicardium is an essential tissue for a functional heart. Epicardial-derived cells differentiate into cells that contribute to the heart as it develops, including fibroblasts and vascular smooth muscle cells. Previous studies have shown that a

heartbeat is required for epicardium formation. Further, preliminary studies from our laboratory have shown that the development of the epicardium is aberrant when the haemodynamics are altered. In our laboratory, the outflow tract (OFT) of HH21 chick embryos was banded with a double overhang knot, to change the heart's haemodynamics, and then harvested at HH29 and HH35. This study aims to investigate how the epicardium and some of its derived cell lines respond to altered haemodynamics in the developing embryo. As the aetiology of many congenital heart defects is unknown, we suggest that an alteration in the heart haemodynamics, leading to an irregular heartbeat, might provide an explanatory basis for some of these defects. Proliferation and apoptosis studies have been performed in treated and control embryos. Further, a number of genes that have a role in coronary vasculature development were analysed by qPCR in treated and control hearts, with differential expression found. Future studies will involve the morphological characterisation of the coronary vasculature and characterisation of functionally important genes, among others. All works in this study are Schedule 1 procedures and have been ethically reviewed at the University of Nottingham and all procedures and facilities are compliant with local and institutional guidelines.

C2 Anatomy and development of the mouse spinal cord stem cell niche

M. Canizares,[1] P. Felts[2] and K. G. Storey[1]

[1]*Division of Cell and Developmental Biology, University of Dundee, Dundee, UK and* [2]*Centre for Anatomy and Human Identification, School of Life Sciences, University of Dundee, Dundee, UK*

The adult mouse spinal cord central canal constitutes a stem cell niche that is activated after injury. Both the ependymal cells lining the central canal and dorsal radial glia-like cells have been proposed to have neural stem cell potential. The overall aim of this project is to characterise this endogenous neural stem cell niche in order to understand how it is generated and regulated.

The central canal derives from the proliferative ventricular layer of the embryonic neural tube. Using immunofluorescence to detect expression of molecular markers of cell behaviour, we have characterised the changing dimensions and organisation of the thoracic spinal cord ventricular layer during development, including the described process of dorsal collapse on embryonic day (E) 14.5. Our work suggests that a decrease in cell proliferation but not increased programmed cell death contributes to this collapse. In addition, we uncovered a novel sub-division of the ventral-most floor plate cell population from E15.5. In parallel with these observations, immunofluorescence and *in situ* hybridization were used to monitor gene expression changes in specific central canal cell populations, including components and targets of Notch, BMP and Shh signalling pathways.

These detailed anatomical studies laid the foundation for development and validation of an *ex vivo* slice culture assay to test gene function within this developing stem cell niche and track movements of specific cell populations. By capturing the ventricular zone transcriptome at key times leading to central canal formation we also adopt an unbiased approach to identification of genes regulating this process.

Animal work was performed under a licence issued by the UK Government Home Office and in accordance with European Community Guidelines (directive 86/609/EEC).

C5 Revitalisation of the mouse molar dental lamina

E. M. Popa,[1] M. Buchtova,[2,3] C. Andoniadou[1] and A. S. Tucker[1]

[1]*Department of Craniofacial Development and Stem Cell Biology, King's College London, London, UK;* [2]*Institute of Animal Physiology and Genetics, v.v.i., Academy of Sciences of the Czech RepublicBrno, Czech Republic and* [3]*Department of Animal Physiology and Immunology, Institute of Experimental Biology, Masaryk University Brno, Czech Republic*

Most mammals have two sets of teeth, a deciduous dentition replaced by a permanent dentition. However, the mouse possesses only one tooth generation.

Tooth development arises with the formation of the dental lamina, an epithelial U-shaped band in the oral epithelium that gives rise to and connects the tooth germs. In the mouse the molars form from a single placode in an anterior-to-posterior direction, with $Sox2^+$ cells in the posterior tail of the first molar giving rise to the second and third molars. Sox2 also localises to the dental lamina associated with two or multiple generations of teeth in other animals. The mouse has a transient epithelial rudiment protruding from the lingual side of the molar tooth germs, in a similar region to the lamina that forms successional teeth in diphyodont mammals. This structure houses Sox2-positive cells but later regresses. Our experiments show that stabilising Wnt/B-catenin signalling in these cells using a tamoxifen-inducible Cre-Lox system leads to the formation of tooth germs. Their dental identity was confirmed by expression analysis of enamel knot markers and other genes involved in tooth development. Interestingly, the phenotype severity reduces with later induction timepoints, suggesting a time window during which the targeted epithelium retains tooth-forming capacity. Our further experiments will aim to validate the successional nature of these tooth germs and to assess their mineralisation capacity during postnatal development. This data could lead to a better understanding of the molecular mechanisms required for tooth regeneration, with important practical implications for the field.

All work performed complies with UK Home Office regulations and all licences are in place.

C8 Effects of genetically reducing neuronal activity on myelination, axon targeting and maintenance of specified cortical neuron populations in mice

K. V. Korrell, H. Fodder, A. Hoerder-Suabedissen and Z. Molnar

Department of Physiology, Anatomy and Genetics, Oxford University, Oxford, UK

Myelin in the CNS is produced by oligodendrocytes to ensure fast saltatory conduction of action potentials at the Nodes of Ranvier. Myelination is dependent on neuronal activity but the underlying mechanism needs to be further investigated. To do

so, neuronal activity will be suppressed in selected populations of murine cortical layer V and VI projection neurons to determine how the missing signals will influence myelination. Therefore, two conditional SNAP25 knockout (Snap25 cKO) mouse lines will be used to prevent neurotransmitter release. Conditional Snap25 KO mice are bred on a floxed-stop-tdTomato background such that all silenced cells are tdTomato-labelled. Additionally, *in utero* electroporation will be conducted to express an additional potassium channel at the Nodes of Ranvier (Kir2.1) to prevent action potential propagation.

Analysis of fibres of Snap25 cKO mice that cannot release neurotransmitters reveals normal targeting and myelination during development but shows unexpected axonal degeneration that also alters myelination at later time points. The time course of degeneration occurs with a distinct progression depending on the silenced cell population. Analysis of oligodendrocyte precursors cells (OPCs) in the striatum of Snap25 cKO LVI revealed no difference to control animals.

In the next step, the proper formation of Nodes of Ranvier will be analysed as well as compaction of myelin membranes by transmission electron microscopy.

In summary, preventing neurotransmitter release in SNAP25 cKO mice does not affect the onset of myelination, oligodendrocyte precursors cells or fibre targeting, but does affect fibre and myelin maintenance.

All animal experiments were carried out in accordance with A (SP)A under a valid Home Office project licence following approval by a local ethical review committee.

C10 Exploring the link between human fetal biomechanics and joint shape

S. Verbruggen,[1] B. Kainz,[2] O. J. Arthurs,[3] J. V. Hajnal,[4] M. A. Rutherford,[4] A. T. M. Phillips[5] and N. C. Nowlan[1]

[1]*Department of Bioengineering, Imperial College London, London, UK;* [2]*Department of Computing, Imperial College London, London, UK;* [3]*UCL Great Ormond Street Institute of Child Health, London, UK;* [4]*Division of Imaging Sciences, Kings College London, London, UK and* [5]*Structural Biomechanics, Department of Civil and Environmental Engineering, Imperial College London, London, UK*

Mechanical stimulation generated by fetal kicking and movements is known to be important for prenatal musculoskeletal development, particularly hip joint shape. It is thought that changes in these dynamic movements give rise to altered joint shape at birth, leading to developmental dysplasia of the hip (DDH) or arthrogryposis (multiple joint contractures). In this study we investigate the biomechanics of fetal movements over gestational time, by combining advanced fetal imaging with computational modelling techniques.

We have developed a custom-designed tracking software, allowing us to characterise the joint displacements during fetal kicking from cine-MRI scans. These movements were then replicated in finite element and musculoskeletal models, allowing us to quantify the joint reaction forces and intramuscular forces in a fetal kick for the first time. These data were generated from observed kicks of fetuses at around 20, 25 and 30 gestational weeks ($n = 6$ or 7 per group). Finally, these forces were applied to geometries of fetal bones generated from postmortem MRI and CT scans, allowing characterisation of the mechanical stimulation experienced by fetal limb tissues during kicking, at each gestational age. It was found that the maximum stress and strain stimulation both increased with gestational age, increasing from 0.8 kPa and 0.06% at 20 weeks to 6.1 kPa and 0.3% at 30 weeks.

This study provides a new insight into the biomechanical environment *in utero*, through the use of cine-MRI data of fetal movements. This is the first quantification of changes in biomechanical stimulation in the fetal skeleton with gestational age due to movements *in utero*, elucidating an upward trend in this mechanical stimulation. This increasing trend with gestational age is important, as restricted leg movements late in gestation (such as occur with fetal breech position) increase the risk of DDH. Finally, this novel computational pipeline enables us to characterise patterns of stimulation *in utero*, which cannot be achieved experimentally. Further analysis of the observed trends in developmental biomechanics may shed new light on the link between fetal biomechanics and anatomy, and thus inform future preventative measures for neonatal joint conditions.

All imaging was acquired during ethically approved research.

C11 Getting under the skin of fetal alcohol syndrome: an investigation of ethanol-induced skeletal defects in a chick embryo model system

D. Brennan[1] and D. Clissmann[2]

[1]*Anatomy, School of Medicine, College of Health and Agricultural Sciences, University College Dublin, Dublin, Ireland and* [2]*Centre for Anatomy and Human Identification, University of Dundee, Dundee, UK*

Fetal alcohol syndrome (FAS) constitutes a range of developmental defects resulting from maternal ethanol (EtOH) consumption during pregnancy. Acute embryonic EtOH exposure can lead to delayed skeletal development, resulting in a range of bone and joint anomalies including radio-ulnar synostosis and altered long bone development. Surprisingly, relatively few in-depth studies have been carried out to investigate EtOH-induced skeletal malformations. The aim of this study was to explore the teratogenic consequences of EtOH on the developing skeletal system using a chick embryo model. Fertile chicken eggs (*Gallus gallus domesticus*) were incubated at 37 °C and 70% humidity for 24 h. A puncture hole was made through the eggshell and 10 μL of 0%, 1%, 5%, 10% or 20% ethanol solution in chick saline (0.73% NaCl) was injected directly into the yolk sac ($n = 5$/group). Puncture holes were sealed with tape and eggs returned to the incubator for an additional 9 days of development (E1 + 9). Embryos were subsequently dissected from surrounding vitelline membranes, stained using a standard Alcian Blue/Alizarin Red protocol and the radius removed for measurement purposes. Total radial length, percentage ossification and growth plate length were analysed ($n = 10$ radii/group). Overall, ethanol-exposed embryos showed signs of skeletal deformation. Defects were evident in (i) the cranial region with presentation of microcephaly in the 10% EtOH group; (ii) the clavicle, which showed signs of impaired osteogenesis with increasing dose; and (iii) a concentration-dependent decrease in ossification of the metacarpals. For radial measurements, a significant reduction in

the total radial length was found in both the 1% and 5% EtOH treatment regimes when compared with the saline control group. A statistically significant reduction in the percentage ossification of the radius was also noted in the 5% EtOH-treated group when compared with control counterparts. In conclusion, qualitative analysis has demonstrated that perturbation of skeletal development is induced by EtOH exposure, affecting the bones of the pectoral girdle and upper limb in particular. Quantitative radial measurement further confirmed an EtOH-induced effect on ossification. Further work exploring the best skeletal parameters to use in identifying FAS-related effects may contribute to future improvements in FAS diagnostic criteria. No ethical approval was required for this study.

C14 Predicting calvarial growth in normal and craniosynostosic mice using finite element analysis

A. Marghoub,[1] J. Libby,[2] C. Babbs,[3] A. O. M. Wilkie,[3] M. J. Fagan[2] and M. Moazen[1]

[1]*Department of Mechanical Engineering, University College London, London, UK;* [2]*Medical and Biological Engineering, School of Engineering, University of Hull, Hull, UK and* [3]*Weatherall Institute of Molecular Medicine, University of Oxford, Oxford, UK*

The cranium consists of several bones that are joined together at their edges by soft tissue called sutures. Early fusion of sutures is a medical condition known as craniosynostosis. Cruzon mouse is a well-established animal model displaying bicorporal suture fusion and is an invaluable model to understand the biomechanics of the skull growth. The aim of this study was to predict calvarial growth in wild type (WT) and mutant type (MT, Cruzon) ($Fgfr^{2C342Y+}$) mice.

Two ontogenetic series of WT and MT mice were scanned using micro-computed tomography. A 3D finite element model of a WT mouse skull at day 3 postnatal development age (P3) was created, including the bones, sutures and brain. The model was used to predict the WT and MT calvarial growth at P7, P10 and P20 where intracranial volume reaches a plateau in mouse. Input parameters to the model were estimated based on a series of parallel experimental studies. Nevertheless, several sensitivity analyses to the input parameters were performed and outputs were compared to *ex vivo* specimens in terms of the overall calvarial shape and bone formation at the sutures.

Sensitivity analyses showed that model predictions were sensitive to the input parameters. However, using the experimental data, the model could predict the radial expansion of the calvarial bones and bone formation at the sutures at P7 and P10 in WT mouse. For example, the difference in calvarial length, width and height between the *ex vivo* and FE predictions were 5%, 13% and 12%, respectively. Further, the model predicted the overall shape of the MT skull at P10, which has a slightly taller, wider and shorter profile compared with the equivalent WT skull at P10.

The models developed in this study are the first validated models of mouse calvarial growth. The close match between the predicted shape of the models and *ex vivo* data build confidence in the modelling approach. However, further studies are required to refine the models. Such models can be used in the long-term for patient-specific modelling of craniosynostosis. No ethical approval was required for this study.

C15 The origins of interneurons in the human cerebral cortex

J. McIntyre,[1,2] A. Alzu'bi,[1,2] S. Lindsay[2] and G. Clowry[1]

[1]*Institute of Neuroscience, Newcastle University, Newcastle upon Tyne, UK and* [2]*Institute of Genetic Medicine, Newcastle University, Newcastle upon Tyne, UK*

In rodents, inhibitory GABAergic interneurons originate in the ganglionic eminences and migrate to the cortex. However, it has been controversially suggested that in humans, a proportion of interneurons are cortically derived. Understanding differences in the developmental origins of human and rodent cortical interneurons may shed light on the evolution of cognitive function and the origins of neurodevelopmental diseases. We prepared dissociated cell cultures from the ganglionic eminences (GE) anterior cortex (ACx) and posterior cortex (PCx) of human fetal brain at 9–10 post-conceptional weeks (provided with the appropriate ethical permission, www.hdbr.org). Neural progenitor cells were isolated and expanded as neurospheres and then plated in standard differentiation media for 8 days to produce immature neurons. Cultures were fixed in paraformaldehyde and double-labelled by immunofluorescence using standard methods for β-tubulin (post-mitotic neuron marker) and GABA (interneuron neurotransmitter) calretinin (interneuron marker) or COUP-TFII (transcription factor characteristic of caudal ganglionic eminence-derived interneurons). Cells were cultured from three different fetal brains, each staining combination made in duplicate wells. Cell counts were made from three randomly selected fields of view from each well. The number of double-labelled cells was expressed as a proportion of β-tubulin-positive cells. The highest proportion of $GABA^+$ neurons was found in GE-derived cultures (53%), demonstrating that progenitor cells retain their regional identity during expansion and differentiation. However, $23 \pm 4\%$ of neurons also expressed GABA in ACx cultures, significantly more than in PCx cultures ($13 \pm 2\%$). In addition, $37 \pm 4\%$ and $28 \pm 2\%$ of neurons expressed calretinin in ACx and PCx cultures, respectively, with the majority of $calretinin^+$ cells also co-expressing GABA. COUP-TFII was expressed in $25 \pm 2\%$ of ACx and $16 \pm 2\%$ of PCx neurons. Thus some cortical progenitor cells possess an intrinsic programme for generating interneurons. Also, the anterior cortex may be a favoured region for cortical interneurogenesis.

C17 Zic4 lineage expression in developing choroid plexus: a murine study

I. M. Amado, T. Pratt and J. Clegg

Centre for Integrative Psychology, University of Edinburgh, Edinburgh, UK

The choroid plexus (CP) is a structured plexus of cells that develops during neuronal development. A combination of neuroepithelium and ependymal cells that line the ventricles are the main constituents of the CP. Its function includes secretion of cerebrospinal fluid (CSF), growth factors and transthyretin

(TTR). Secreted sugar polymer heparan sulphate (HS) has been hypothesised to regulate cell signalling. Previous work has shown that a mutant mouse embryo which had no HS in the Zic4 lineage had a hydrocephalus phenotype. Therefore, we identified a need to study the localisation of Zic4 lineage cells in the CP and to determine how depletion of HS affects the CP. We used immunohistochemistry to visualise the interface between HS and the Zic4 cell lineage. HS is synthesised by two proteins – Ext1 and Ext2. The gene for Ext1 was removed using the Cre/LoxP method under the control of the Zic4 driver, and a green fluorescent protein reporter transgene (switched on by Cre) was used to mark the Zic4 lineage and to check whether this lineage is present in the CP; CP was traced using TTR.
Data show that the CP is not entirely made of Zic4 lineage cells and Ext1 cKO, implying that HS does not affect the presence of the Zic4 lineage. The results indicate that Zic4 lineage is expressed in the CP, but does not provide us with an answer to the enlargement of the lateral ventricles, which may be associated with the secretion of factors from the CP.
All animal experiments were conducted in accordance with the guidelines set by the UK Home Office under a registered project licence.

C18 Modelling the development of human tissues *in vitro* using a combination of advanced cell technologies

L. Smith,[1] D. Owens,[1] M. F. Roger[1] and S. A. Przyborski[1,2]

[1]*Department of Biosciences, Durham University, Durham, UK and* [2]*ReproCELL Reinnervate Ltd, NETPark Incubator, Durham, UK*

The development of mammalian tissues is a critical process that is poorly understood. Current models involve using pluripotent stem cells in various methodologies, including the formation of embryoid bodies. These multicellular structures have been shown successfully to recapitulate events of early mammalian embryogenesis. However, as the size of cell aggregates increases, a larger diffusion distance is created, leading to a significant loss of viability and necrosis in the core of the structure. This is a result of the inability of nutrients and oxygen to diffuse into central regions. As a consequence, this limits the ability to conduct long-term studies of tissue development. As differentiation processes are time-dependent, this will impact greatly on the maturity and complexity achieved by pluripotent cell derived tissues using *in vitro* methods. Yet, it is well known that pluripotent cells can form highly differentiated tissues achieving high levels of complexity and maturity, as is seen in a teratoma assay *in vivo*. Through a combination of embryoid body culture and application of an inert three-dimensional porous polystyrene scaffold, we have improved the viability of embryoid bodies by enabling them to form flattened tissue discs, allowing reduced diffusion distances. This improves cell viability, extending their time in culture, resulting in increased cell differentiation. We have increased the complexity of the model by adding exogenous morphogens such as retinoic acid, Activin A and BMP4, each allowing for directed differentiation towards particular primary germ layers and enabling the detailed study of these developmental pathways. Histological staining and immunofluorescence for specific markers have revealed the identity, complexity and maturity of the tissues. Initial experiments used an embryonal carcinoma cell line to optimise methodology, allowing for proof of concept experiments. Further studies involving mouse embryonic stem cells have revealed the ability to maintain tissue disc structures in the *in vitro* environment for a significant amount of time without a reduction in viability, while forming complex differentiated structures. It is anticipated that this model will provide a novel *in vitro* alternative to the current pluripotency assessment method, the xenograft-based teratoma assay, as well as an approach to investigate the early stages of tissue development. No ethical approval was required for this study.

C20 The natriuretic system in human salivary gland health and disease: potential use for diagnosing early stages of malignancy in salivary glands

I. Miletich, A. Leone, A. Gulino, B. Belmonte, W. Arancio and C. Tripodo

Craniofacial Development and Stem Cell Biology, Dental Institute, King's College London, London, UK

The natriuretic peptide system comprises three ligands, atrial natriuretic peptide (ANP), brain natriuretic peptide (BNP) and C-type natriuretic peptide (CNP), and three receptors, NPR-A, NPR-B and NPR-C. Although the heart is the major source of ANP and BNP, production of ANP has been reported in a number of extra-cardiac sites, including the salivary glands (SGs). Our immunohistochemical studies show that all members of the atrial natriuretic system are present in human adult sub-mandibular SGs in three anatomical locations: the excretory ducts, the blood vessel walls and the peripheral nervous system. This is the first time the natriuretic peptide system has been shown to be expressed in the peripheral nervous system. We show that this pattern of expression is conserved between mice and humans, which suggests an important role for this system in SG function, possibly as a neurotransmitter or neuromodulator during SG homeostasis. We further identified that NPRA expression was elevated in the salivary gland stroma of a number of patients with oral squamous cell carcinoma (OSCC), whereas NPRA expression was downregulated in SG advanced primary OSCC, suggesting high NPRA levels could be used to diagnose early stages of malignancies in SGs. Further work currently characterises NPRA expression in the SGs of patients with OSCC.
All animal experiments were conducted in accordance with Home Office regulations. Human tissue samples were obtained from the archives of the Human Pathology section of the University of Palermo (Italy) with all procedures carried out in accordance with the Helsinki Declaration.