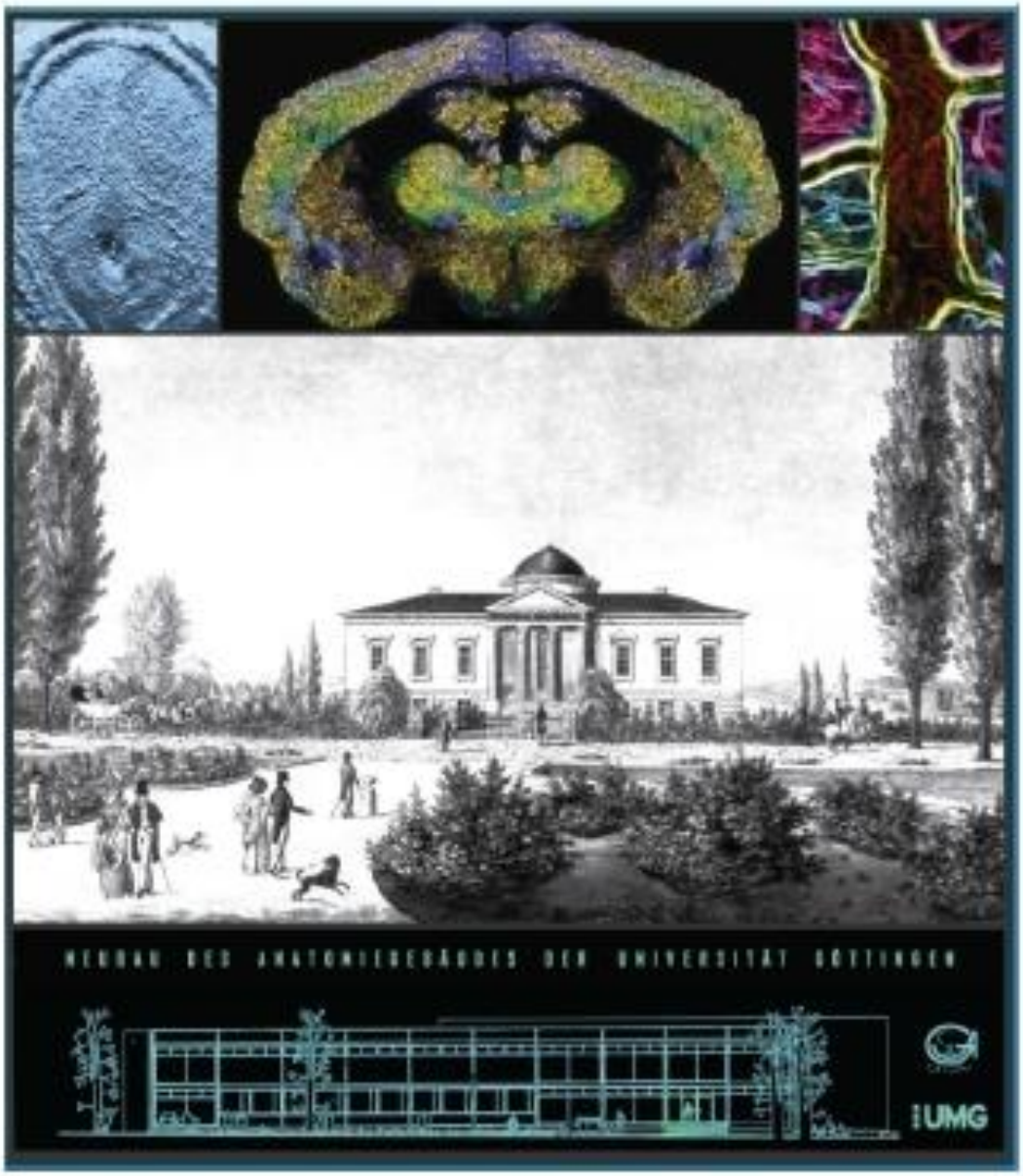
ANATOMISCHE GESELLSCHAFT



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To find your abstract or
an abstract of interest
please use the alphabetical list of
first authors of lectures and posters
starting on next page.

Gensch, R.	P46	Hütz, K.	L71
Geyer, S.	L2	Imig, C.	L23
Ghelani, T.	L46	Ingenwerth, M.	P98
Giesecke, T.	L51	Jabari, S.	P19; P50
Gilloteaux, J.	L15	Jamann, N.	L24
Grabiec, U.	P79	Jászai, J.	L30
Günther, J.	P6	Jedlicka, P.	L60
Guy, J.	L59	Jirkovská, M.	P132
Habicht, J.	P17	Johann, S.	L17
Häfelein, K.	P96	Jungenitz, T.	P51
Haider, G.	L34	Kachlik, D.	PP11
Hainz, N.	P80	Kandel, C.	P20
Hammer, C.	PP15	Karaer, E.	P7
Hamscha, U.	L52	Karnati, S.	L11
Harmoush, B.	P109	Kern, K.	P81
Harrach, D.	L16	Keshavarz, M.	P99
Hartung, K.	P110	Khayrullin, R.	P22
Hassan, W.	P111	Kirschneck, C.	P121; P122
Hattermann, K.	P97	Klimaschewski, L.	P53
Hellfritsch, A.	P47	Kling, K.	P123
Hermanowicz-Sobieraj, B.	P48	Klingenstein, M.	L18
Hirt, B.	L74	Klochkova, S.V.	P31
Hochstrasser, T.	L22	Klopries, K.	P23
Hoeflein, T.	P49	Kluth, D.	PP13
Höfflin, F.	L47	Knabbe, J.	P52
Hohmann, T.	L10	Knudsen, L.	L53
Hörmann, R.	P18	Kohrmann, A.	P8
Horstmann, H.	P138	Kokozidou, M.	PP17

Authors (alphabetical order)

Poster-Preview 0:

Titel: Lpa1, lpa2, lpa4, and lpa6 receptor expression during mouse brain development

Autoren/Adressen: Anja U. Bräuer (Universitätsmedizin Rostock), Olga Kieselmann (Charité – Universitätsmedizin Berlin), Sandra Schrötter (Charité – Universitätsmedizin Berlin), Jerold Chun (The Scripps Research Institute), Junken Aoki (University of Tokyo); anja.braeuer@med.uni-rostock.de

Abstract:
Lysophosphatidic acid (LPA) is a small bioactive phospholipid that acts as an extracellular signalling molecule and is involved in numerous cellular processes, including cell proliferation, migration, differentiation and motility. LPA acts by binding and activating at least six known G-protein-coupled receptors: LPA1-6. In recent years, LPA has been suggested to be an important player in both normal neuronal development and under pathological conditions in the nervous system.

We show the expression pattern of LPA receptors during mouse brain development, which we determined using quantitative real-time PCR (qRT-PCR) and immunocytochemistry.

Only LPA1, LPA2. LPA4, and LPA6 mRNA-transcripts were detected throughout developmental stages from embryonic day 16 (E16) until postnatal day 30 (P30) of hippocampus, neocortex, cerebellum and bulbus olfactorius in our experiments, while the expression of LPA3 and LPA5 genes was below detection level. In addition to our qRT-PCR results, we also analyzed the cellular protein expression of endogenous LPA receptors, with focus on LPA1 and LPA2 within postnatal brain slices and primary hippocampal neuron differentiation with and without cytoskeleton stabilisation and destabilisation.

We show development-dependent expression of LPA receptors in mouse brain and in cultured hippocampal primary neurons. Interestingly, we found that commercially available antibodies for LPA receptors are largely unspecific.

Poster-Preview 1:

Titel: Combined anatomy and surgery training for medical students — a spiral concept addressing future needs in healthcare

Autoren/Adressen: Stefan Klima (University of Leipzig), Sabine Löffler (University of Leipzig), Pierre Hepp (University of Leipzig), Hanno Steinke (University of Leipzig), Niels Hammer (University of Otago); nlshammer@googlemail.com

Abstract:
In recent years there has been a significant decline in trainee enrollments into orthopaedic and trauma surgery in Germany. This is further intensified by an increasing demand of orthopaedic trainees due to the ageing population and flexible part time work models. To address this issue, a program was established at the University of Leipzig aiming to increase student interest in surgery while enrolled in their five-year medical education and to learn surgically relevant knowledge

Five workshops are jointly offered by clinical anatomists and trauma and orthopaedic surgeons, including “Advanced dissection training”, “Biomechanics in healthy and pathological conditions”, “Surgical approaches”, “Principles of bone and joint surgery” and “Sports medicine”. The workshops have a duration of one week, are modular and sequential, mostly in a hands-on environment and with a student-teacher ratio of 2:20 or higher. A survey was taken to determine the desire and doubts about a future employment in surgery.

The workshops are considered being highly attractive for the students and usually prematurely booked fully. The interest in surgery increased in the in the further course of study, whereas fear of physical stress deceased. No differences were found for the genders.

Long-term evaluation has proven this concept to be highly efficient to improve the interest of medical students in surgery and to provide an optimal preparation for their future specialist training. Furthermore, the concept provides an effective and targeted measure for student selection into future training programs. However, the high financial and time efforts have to be considered.

Poster-Preview 2:

Titel: The rat vomeronasal organ is a vitamin d target

Autoren/Adressen: Andrea Rodewald (Klinikum der FSU Jena), Veronika Gebhart (Klinikum der FSU Jena), Gustav Jirikowski (Klinikukm der FSU Jena); gustav.jirikowski@med.uni-jena.de

Abstract:
Steroid hormones play a significant role as pheromones to control mood and behavior. The vomeronasal ogran VNO is known to be a target of olfactory steroids. Here we wanted to determine whether binding proteins for the steroid hormone Vitamin D 3 occur in the rat VNO

We employed immunohistochemistry, in situ hybridization and RT-PCR to determine expression of Vitamin D receptor VDR and Vitamin D binding protein DBP in the rat VNO.

A portion of the sensory cells in the VNO contained nuclear and cytoplasmic immunoreactivity for VDR. DBP was observed in the cytoplasm of sensory neurons, mostly confined to apical dendrites and to the basis of olfactory cilia. Both proteins were in part colocalized. VDR and DBP were also observed in cytoplasm of single ciliated cells within the non-sensory portion of the VNO. DBP was found in some of the vomeronasal glands (VNG), indicating secretion of the binding globulin. In situ hybridization with synthetic oligonucleotide probes complementary to DBP enconding transctripts revealed specific hybridization signal in some of the sensory cells, in single cells of the non- sensory epithelium and in VNG. With RT-PCR of RNA extracts from rat VNO we were able to amplify measurable amounts of both VDR and DBP encoding transcripts

It is likely that both vitamin D binding proteins are expressed in the rat VNO suggesting that this organ is a vitamin D target. Vitamin D3 and related metabolites may have pheromone like properties in rat.

Poster-Preview 3:

Titel: Dissection course in german speaking anatomy institutes – figures and perspectives

Autoren/Adressen: Anja Böckers (Ulm University), Friederike Hasselblatt (Ulm University), Tobias Maria Böckers (Ulm University); anja.boeckers@uni-ulm.de

Abstract:
While there´s a change in pardigm concerning the body donor`s anonymity at Angloamerican universities, it is yet unknown whether these innovations are transferable into German structures. A personification could strenghthen the perception of the donor as „first patient“ and students´ empathy. Therefore, it was the aim of this study to gather data about 1) the current status at German speaking anatomical institutes, 2) the opinion of anatomists, students and donors concerning personification of donors.

Anatomical institutes were invited to participate in an online-questionnaire. A random tenth of the registered donors at Ulm University received a questionnaire postally. Students were surveyed paper-based at the end of the dissection course.

Response rates run up to 73% (institutes, n=32/45), 76% (students) and 51% (donors). The majority of students desired additional information about their donor (78.1%) and agreed with the donor´s anonymity (79.3%). Donors supported the anonymous disclosure of information about their medical record (92.5%). In contrast, this information is only available in about 28% of the institutes. Overall, 78% of anatomists think that personification of the donor is not a reasonable teaching innovation.

Our data outline the present status of dissection courses and anatomists opinion on the personified use of body donors. Results describe a sore traditional picture of anatomists´ attitudes and activities regarding course organization or body donation. However, as donors are willing to confide additional information anonymously about themselves - information the students ask for in the course – a framework for a limited, anonymous personification should be developed.

Poster-Preview 4:

Titel: Surface marker-based isolation of human dopaminergic neurons for pharmacological studies and cell therapeutic applications

Autoren/Adressen: Vishal Menon (University of Freiburg), Julia Oechsler (University of Freiburg), Jan Pruszak (University of Freiburg); vishal.menon@sgbm.uni-freiburg.de

Abstract:
The loss of nigro-striatal dopaminergic (DA) projection neurons represents a hallmark of Parkinson's disease (PD), the second most common degenerative disorder of the central nervous system. Patient-derived pluripotent stem cells provide novel sources for generating DA neurons for autologous cell therapeutic approaches (stereotactic neural transplantation) as well as for pathophysiological study and pharmacological in vitro screens. For either of those paradigms, selecting DA neurons at a better-defined degree of purity has proven to be critical.

Using immortalized midbrain DA progenitors (LUHMES) and pluripotent stem cell-derived neural cultures, we employed the most comprehensive flow cytometric characterization of cluster of differentiation (CD) antigens expressed in human DA neuronal differentiation to date.

Determining the expression profiles of >250 CD antigens, we defined a panel of 20 markers for combinatorial, multicolor flow cytometric analysis of DA neural proliferation versus differentiation. The activated leukocyte-cell adhesion molecule (ALCAM; CD166) as well as tetraspanin (e.g., CD63, CD151) and integrin family members (e.g., alpha-2-integrin; CD49b) were identified as viable markers for vulnerable DA subsets in PD-related toxicity assays (MPP+, ferrous chloride; calcium ionophore A23187). Corresponding subpopulations could be enriched by cell sorting for DA phenotypic analysis and functional study.

Our approach identifies novel surface marker codes for DA neuronal cell isolation with utility for in vitro assays and future neural DA circuitry repair.

Poster-Preview 5:

Titel: Affection of the cerebellum in parkinson’s disease and dementia with lewy bodies

Autoren/Adressen: Kay Seidel (Universitätsklinikum Frankfurt), Mohamed Bouzrou (Universitätsklinikum Frankfurt), Nina Heidemann (Universitätsklinikum Frankfurt), Rejko Krüger (University of Luxembourg), Ludger Schöls (Hertie Institute for Clinical Brain Research), Wilfred den Dunnen (University Clinic of Groningen), Horst-Werner Korf (Universitätsklinikum Frankfurt), Udo Rüb (Universitätsklinikum Frankfurt); Kay_Seidel@gmx.de

Abstract:
The alpha-synucleinopathies Parkinson’s disease (PD) and dementia with Lewy bodies (DLB) exhibit hypokinetic motor symptoms and resting tremor. While the former correlates well to the degeneration of basal brain structures, the latter implies the involvement of cerebellar structures and/or associated networks and nuclei. Recent discoveries of the presence of alpha-synuclein pathology in precerebellar structures and cerebellar pathology in a familial PD case further implicate involvement of the cerebellum in idiopathic PD and DLB.

We investigated immunohistochemically stained sections from the cerebella of 6 patients with idiopathic PD, 1 patient with familial PD and 5 patients with DLB for the presence of alpha-synuclein positive aggregates and signs of neurodegeneration.

Our investigation revealed marked alpha-synuclein pathology in the cerebella of both PD and DLB patients that was mostly situated in the deep cerebellar white matter and nuclei, primarily in the form of Lewy neurites. Aggregation in the cerebellar lobules was present but mild. Additionally to neuronal aggregates, oligodendroglial alpha-synuclein aggregates could be found. Overt neurodegeneration was not observable.

The consistent affection of the cerebellum in PD and DLB may contribute to the tremor that is present in patients with either disease. Since aggregation pathology in precerebellar structures was established beforehand, the affection of the cerebellum supports the notion that the alpha-synuclein pathology may spread along anatomical interconnectivities. Additionally, the aggregation pathology observed in both disease groups showed considerable overlap, implying that PD and DLB are closely related and may even represent different points on the same clinical spectrum.

Poster-Preview 6:

Titel: Effect of isoprostanes on cardiac blood vessel growth and the activation of vessel wall-resident stem and progenitor cells

Autoren/Adressen: Jochen Bauer (Julius-Maximilians Universität Würzburg), Philipp Wörsdörfer (Julius-Maximilians Universität Würzburg), Ralf Benndorf (Martin-Luther Universität Halle-Wittenberg), Stefan Frantz (Universitätsklinikum Halle (Saale)), Süleyman Ergün (Julius-Maximilians Universität Würzburg); sueleyman.erguen@uni-wuerzburg.de

Abstract:
Isoprostanes are products of unsaturated fatty acids acting via the thromboxane A2-(TP)-receptor. Our study investigated the effect of isoprostanes on myocardial infarction (MI)-associated processes and the activation of vessel wall-resident stem cells (VW-SCs).

Cardiac angiogenesis assay was performed using left ventricular tissue from WT and TP-KO-mice. MI was induced by ligation of the left coronary artery. Immunhistological analyses were performed by using paraffin embedded cardiac tissue sections.

Cardiac sprouting was significantly reduced in isoprostane-treated mice. By using TP-KO-mice, a TP-dependent inhibitory effect of isoprostanes on cardiac angiogenesis was observed. Furthermore, an isoprostane-dependent alteration of the amount of cardiac VW-SCs was detected. In addition, we observed a TP-dependent inhibitory effect of isoprostanes on the activation of cardiac VW-SCs. MI induced a modulation of the amount of stem cells in the vascular adventitia and the sub-epicardial space exhibiting an inflammatory phenotype. Flk-1+/Islet-1+ cells in the infarcted area indicate the presence of cardiomyocyte progenitor cells in the cardiac vessel wall. Preliminary results indicate no differences between vehicle- and isoprostane-treated mice in respect to morbidity, mortality and infarct size. MI-induced activation of VW-SCs was reduced in isoprostane-treated animals, indicating an inhibitory effect of isoprostanes on the activation of VW-SCs in vivo.

Our study shows that the vascular adventitia and the sub-epicardium serve as a reservoir for stem cells giving rise to inflammatory and cardiomyocyte-progenitor cells after MI. Furthermore, the activation of cardiac VW-SCs is TP-dependently influenced by isoprostanes which might affect cardiac angiogenesis.

Poster-Preview 7:

Titel: The changing face of FIPAT

Autoren/Adressen: John Fraher (University College Cork, Ireland); j.fraher@ucc.ie

Abstract:
Recent changes in the operation of FIPAT, the Federative International Programme for Anatomical Terminology are outlined. FIPAT is a major component of the International Federation of Associations of Anatomists.

It comprises the official international standard set of terminologies of the anatomical profession. Its individual sections cover Anatomy, Histology, Embryology, Neuroanatomy, Anthropology, Odontology and Oral Biology. Each is operated by a Working Group. There are also Informatics and Latin Subcommittees.

FIPAT organisation is flexible, enabling continual modification and refinement in response to feedback, thereby enhancing comprehensiveness through incorporation of new terms from anatomical science and anatomically relevant clinical fields.
All official terms are in Latin, with English equivalents and synonyms in both languages. The Latin term provides the pivotal reference point for translation into any vernacular.
Each Terminology consists of a hierarchically organised, systematised list of terms encompassing pure and applied aspects of Anatomy. The Terminologies are currently undergoing transformation through informatics into ontologies in computer-readable formats, thereby capable of interacting with others in the biosciences field. Previously the Terminologies for anatomy, histology and embryology were published as books. Henceforth, they will be in the form of publications on the FIPAT website.

FIPAT's main aim currently is to have all of its documents universally available on its website. Initially this will apply to the anatomy, histology, embryology and neuroanatomy sections. It will take place in the context of a broadened awareness of the place and value of the FIPAT terminologies in the wider biomedical area.

Poster-Preview 8:

Titel: Establish the novel bioreactor for tendon tissue cultivation

Autoren/Adressen: Johanna Bartsch (Uniklinik RWTH Aachen), Babithra Yoganathan (Uniklinik RWTH Aachen), Wolgang Willenberg (Uniklinik RWTH Aachen), Marcus Stoffel (Uniklinik RWTH Aachen), Bernd Markert (Uniklinik RWTH Aachen), Holger Jahr (Uniklinik RWTH Aachen), Thomas Pufe (Uniklinik RWTH Aachen), Mersedeh Tohidnezhad (Uniklinik RWTH Aachen);mtohidnezhad@ukaachen.de

Abstract:
The mechanical loading the tendon is essential for maintaining the phenotypic stability of tenocytes in both traction and gliding types. The aim of the present study is a) use of a novel bioreactor for physiological cultivation of tendon tissue and b) to improve the in vivo repair capacities of cells and tissue in both traction and gliding tendons.

The tendon of Musculus flexor digitorum longus was isolated from rat and was monoaxial stretched by a novel bioreactor for 6 and 24h at 1Hz. The real time reaction-forces were obtained by the bioreactor. After stimulation the tendons were embedded in paraffin and slides were prepared. Quality of various areas (traction or gliding) was determined using histochemistry and immunohistochemistry analysis

The mechanical result of bioreactor shows that the mechanical strength of tendon was increased in the first half of loading time, but it was decreased in the second part. The alcianblue-staining shows that the content of cartilage in the three areas is statistically different after stimulation. Tenomodulin, fibromodulin, typ I collagen were also expressed in all selected area. We observed a stronger intensity of scleraxis only after 6h stimulation.

We exclude a physiological cultivation condition by current parameters. Wider range of parameter for tendon cultivation should be analyzed in bioreactor. A combination of mechanical and histological data allows us to improve the cultivation condition of tissues

Poster-Preview 9:

Titel: Ultrasound evaluation of conventional and newly designed maneuvers for provoking longitudinal gliding of the median nerve in the carpal tunnel - An anatomical study

Autoren/Adressen: Lukas F Reissig (Medical University of Vienna), Reinhard Beikircher (University of Applied Sciences, Krems), Chieh-Han J Tzou (Medical University of Vienna), Ulrike Hamscha (Medical University of Vienna), Wolfgang Grisold (Kaiser-Franz-Josef Hospital, Vienna), Wolfgang J Weninger (Medical University of Vienna), Stefan Meng (Kaiser-Franz-Josef Hospital, Vienna); lukas.reissig@meduniwien.ac.at

Abstract:
Compression of the median nerve inside the carpal tunnel is a frequently observed pathology. A conservative therapeutic option is to mobilise the nerve by provoking its gliding. This study has been conducted to objectively evaluate the extent of median nerve movement during conventional and newly designed nerve gliding exercises.

We used upper extremities (n=18) still connected to non-embalmed human cadavers. Cadavers with neuromuscular diseases in the medical record or featuring anatomic variations in the wrist were excluded. Gliding exercises were passively performed by an experienced physiotherapist. Distances between markers applied into the nerve and markers in the periosteum were visualised and measured with ultrasound. Microsoft Excel was used for comparing the results achieved by applying different gliding exercises.

Conventional exercises led to substantial gliding of the median nerve proximal to the carpal tunnel and between the heads of the pronator teres muscle (mean distance of 12 and 13.8 mm, respectively). In contrast, the gliding distance inside the carpal tunnel was only 6.6 mm. Exercises, which we newly designed according to anatomical considerations lead to a nerve gliding distance inside the carpal tunnel of 13.8 mm.

Conventional nerve gliding exercises lead to minimal nerve excursions in the carpal tunnel and thus are only of low therapeutic consequence. Our newly designed exercises result in substantial longitudinal gliding. Although clinical trials will have to deliver the clinical evidence, our results suggest that this new approach will lead to better therapeutic results in the conservative therapy of the carpal tunnel syndrome.

Poster-Preview 10:

Titel: Dot1l and histone h3 lysine 79 methylation determine cerebral and cerebellar development by controlling neural progenitor proliferation and cell fate

Autoren/Adressen: Henriette Franz (Albert-Ludwigs-University Freiburg), Venissa Machado (Albert-Ludwigs-University Freiburg), Patrick Bovio (Albert-Ludwigs-University Freiburg), Tanja Vogel (Albert-Ludwigs-University Freiburg); tanja.vogel@anat.uni-freiburg.de

Abstract:
DOT1L histone methyltransferase of histone H3 lysine 79 plays an important role in different cellular processes such as DNA damage response, gene expression and cell cycle regulation. Aberrant H3K79 methylation (H3K79me) is observed in humans with neural tube defects indicating the dependence of correct neural development on H3K79me patterning. DOT1L activity influences progenitors in different neuroanatomical locations, including spinal cord, cerebellum and telencephalon. This study aimed to decipher the role of DOT1L for CNS development.

To unravel the mechanism of DOT1L in regulating progenitor proliferation and neural cell fate we applied genome-wide analysis of the telencephalic and cerebellar expression profiles using RNA-sequencing/microarrays together with H3K79me2 profiling using ChIP-sequencing in different DOT1L-deficient mouse models.

We identified key transcription factors and signalling pathways affecting neural proliferation and cell fate determination as putative direct DOT1L/H3K79me target genes. Our findings delineate DOT1L and the H3K79me modification as novel master regulator of cortical, hippocampal and cerebellar development with regard to proliferation and neuronal differentiation.

Epigenetic modifications are involved in correct neuronal specification, which act at the interface between proliferation and differentiation.

Poster-Preview 11:

Titel: Vasculature of the explanted aorto-coronary venous grafts

Autoren/Adressen: David Kachlik (Third and Second Faculty of Medicine), Josef Stingl (Third Faculty of Medicine), Pirk Jan (Institute for Clinical and Experimental Medicine), Zbynek Straka (Third Faculty of Medicine), Marek Setina (Second Faculty of Medicine), Josef Sach (Third Faculty of Medicine), Vladimir Musil (Third Faculty of Medicine); david.kachlik@lf3.cuni.cz

Abstract:
The vasculature of the wall of autologous grafts for aorto-coronary bypassing is a fundamental feature for the viability of the graft.

The vasculature of explanted aorto-coronary venous grafts, harvested both from preoperatively and autopsies were studied. Samples from 39 patients were studied by methods of qualitative histology using the light microscope techniques. The interval between implantation and explantation of venous grafts was 1 day to 35 years.

One month after implantation the arterialization of the media and a massive intimal proliferation, combined with atherosclerosis begin to occur. Both mechanisms principally contribute to the venous graft lumen reduction. Growing of the vasa vasorum from the adventitia into the external layers of the media is apparent from 7 to 24 months after implantation. Later, the infiltration of the vasa vasorum extends into the complete thickness of the media and further into the hyperplastic intima. Neither a direct communication of the vasa vasorum with residual lumen of the venous graft has been noticed nor a direct relationship between the vasa vasorum neoangionenesis and age/sex of patient/type of graft has been proved.

The authors conclude that the graft disease progression is based mainly on changed hemodynamic relations and endothelial trauma. Proliferation of the vasa vasorum is a secondary consequence caused by reaction on the structural changes of the graft wall.

Poster-Preview 12:

Titel: Cor triatriatum: a heart variation not mentioned in classical embryological textbooks

Autoren/Adressen: Horst Claassen (Martin Luther University of Halle-Wittenberg), Christian Busch (Federal Armed Forces Hospital Hamburg), Martin Schicht (Friedrich Alexander University of Erlangen-Nürnberg), Friedrich Paulsen (Friedrich Alexander University of Erlangen-Nürnberg); horst.claassen@medizin.uni-halle.de

Abstract:
Cor triatriatum is a rare congenital cardiac malformation characterized by a fibromuscular membrane that divides the left atrium into two distinct chambers. It is found in 0.4% of patients with congenital heart disease and in less than 0.1% of clinically diagnosed cardiopathies. This variation is associated with other congenital cardiac lesions in up to 80%.

A cor triatriatum was found in a 60-year-old female cadaver during gross anatomy course. Beside classical dissection, ultrasound, magnetic resonance imaging and computed tomography were applied. Photographs were taken and the variation was illustrated.

The left atrium was partly divided into two chambers by an intra-atrial membrane which took its origin at the upper level of the oval foramen. The membrane was perforated by two very small openings. In addition, the right atrium showed a septum-like structure at its posterior and lateral wall. This septum started at the lower rim of the oval fossa. The oval fossa was more deep than normally seen. However, no opening was seen in oval fossa in direction to the left atrium.

Concerning the pathogenesis of cor triatriatum different oppinions exist: (1) abnormal growth of septum primum; (2) incomplete incorporation of embryonic common pulmonary vein into the left atrium; (3) entrapment of the common pulmonary vein by the left horn of the venous sinus; (4) persistence of the left superior caval vein that impinges on the developing left atrium. The presented case will be discussed in light of this background.

Poster-Preview 13:

Titel: The embryology of the “persitent cloaca”

Autoren/Adressen: Dietrich Kluth (University Hospital Leipzig), Stefanie Märzheuser (Charite - University Medicine Berlin), Martin Lacher (University Hospital Leipzig); dietrich.kluth@medizin.uni-leipzig.de

Abstract:
In general, the malformation female “persistent cloaca” is defined as a structure in which hindgut, uterus/vagina and urethra converge to a common channel. It is believed that this malformation appears in early embryology. However, detailed studies of these developmental processes do not exist. The aim of this study is to determine if a “persistent cloaca” is visible in normal female development.

26 rat and mice embryos staged between embryonic day (ED) 10 to ED15 (mice) and ED12 to ED22 (rat) were investigated using scanning electron microscopy (SEM) and X-Ray micro tomography (µCT). Rat embryos served as normal controls, while mouse Embryos (SD mice) with anorectal malformations served as study group.

In normal female rat embryos the separation of the embryonic urethra and rectum took place at ED16. At this time point neither the uterus nor the vagina were present in female rat embryos. Thus our study could not confirm the existence of a “persistent cloaca” as a feature of normal embryology. In µCT studies, the uterus/vagina were only seen in embryos older than ED18.

In normal female rat embryos a “cloaca” in the actual sense does not exist because uterus/vagina develop after the complete separation of urethra and rectum takes places. Recently it has been shown that a too short cloacal membrane results in the formation of the urorectal fistula which inhibits the normal development of the vagina. Therefor the malformation called “cloaca” is a result of a combined disturbed developmental process.

Poster-Preview 14:

Titel: The x-ray microtomography in pediatric surgery – a new tool for the study of embryos?

Autoren/Adressen: Susann Brosig (University Hospital Leipzig), Jan-Hendrik Gosemann (University Hospital Leipzig), Nicole Peukert (University Hospital Leipzig), Roman Metzger (Paracelsus Medical University (PMU) Salzburg), Martin Lacher (University Hospital Leipzig), Hartmut Schneider (University Hospital Leipzig), Dietrich Kluth (University Hospital Leipzig); Dietrich.Kluth@medizin.uni-leipzig.de

Abstract:
The understanding of congenital malformations is crucial for pediatric surgeons. Thus the discovery and testing of new embryological techniques is essential for pediatric research. In this study we investigate the potential of X-ray micro-CT (µCT) scans which create high-resolution 3-D bodies of specimens studied. The aim was to show (1) can embryos be studied using this technique? (2) What is the impact of the fixation technique?

Chicken, mouse and rat embryos were used in this study. While chicken embryos were harvested fresh, rat and mouse embryos came from our collection of fixed embryos. Fresh, wet samples were fixed in glutaraldehyde. All other samples were dried using the “critical point” technique. The embryos were scanned using the µCT system SkyScan 1172 (Bruker).

Dry probes showed good to excellent contrast. Fixation technique had no impact on image quality in this cohort. Image contrast varied depending on developmental stage and species. Very young embryos (ED 2 in chicken) and mice in general showed less contrast than older chicken (> ED 4) and rats (> ED 13). A variety of embryonic structures were easily shown in these embryos including two embryos with induced diaphragmatic hernias.

In summary, we demonstrate a new technique for embryonic research by using µCT to create 3D-bodies of different embryos. Compared to SEM, surface images showed less detail (diaphragmatic hernias). However, as this technique is non-destructive, it is a good complementary research technique, especially as the probes prepared for the µCT can be used for SEM and vice versa.

Poster-Preview 15:

Titel: Interface morphology and gas production of a refractive ultraviolet femtosecond laser – comparison with established infrared systems

Autoren/Adressen: Christian Hammer (University of Erlangen-Nürnberg), Yao Zhang (WaveLight GmbH), Johannes Lörner (WaveLight GmbH), Christian Wüllner (WaveLight GmbH), Friedrich Paulsen (University of Erlangen-Nürnberg), Kathleen Kunert (Helios Klinikum Erfurt); christian.hammer@fau.de

Abstract:
Gas production and interface quality were evaluated after creation of refractive lenticules. A novel ultraviolet (UV, 345 nm) femtosecond laser was compared with two established infrared laser systems (WaveLight FS200® and Zeiss VisuMax®).

Refractive lenticules were created in 50 porcine corneas with the UV laser (n=22), the VisuMax® (n=22) and the FS200® (n=6). Six corneas from every laser group were fixed immediately after lenticule creation (-5D) to entrap the gas produced. Here, lenticule extraction was omitted. From the remaining 32 corneas (UV laser and VisuMax®: n=16 each) lenticules were extracted. Here, two different refractive corrections (-5D and -10D, n=8 each) were administered. Upon lenticule extraction a flap was created and lifted. In additional 16 corneas, a flap was cut by the FS200® and lifted afterwards. The corneas were processed for light (gas bubble 2D-morphometry) and scanning electron microscopy (interface quality).

The VisuMax® laser generated approximately 2.5 times less gas than the FS200®. The UV laser even undercut the VisuMax® approximately 2.6-fold. The interfaces created by the FS200® appeared smooth and without irregularities. The optical zone margin was always visible in the VisuMax® samples, but not discernible in the UV specimens. Slight irregularities and very small lenticule pieces were found occasionally in both groups. The UV interfaces appeared slightly rougher than the VisuMax® interfaces.

UV laser lenticule extraction seems promising as the interface quality was comparable to the clinical VisuMax® system while gas production was reduced significantly. This should allow for a more gentle and precise procedure to extract refractive lenticules.

Poster-Preview 16:

Titel: Expression of meiosis-associated genes in normal human spermatogenesis and meiotic arrest

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Abstract:
During spermatogenesis, diploid spermatogonia develop to haploid spermatids by meiosis. Patients with non-obstructive azoospermia often show an arrest at the level of spermatocytes (SZA). Failure of meiosis, i.e. regarding synaptonemal complex (SC) formation, homologous recombination (HR) or telomer attachment (TA), might be a reason.

To assess meiotic failure in SZA patients, a panel of ten genes was analyzed in SZA samples (n=14) compared to normal spermatogenesis (NSP) on mRNA (all genes, RT-PCR) as well as protein level (SYCP3 and DMC1, IHC).

All genes analyzed were expressed in NSP. In SZA samples analyzed, SC gene expression was weaker or absent in some samples (SYCP3). Also DMC1 (early HR) was not expressed in all SZA samples. TA genes were expressed in NSP as well as in all SZA samples. IHC showed a stage-specific staining for SYCP3 and DMC1 in NSP. Whereas SYCP3 was localized in pachytene spermatocytes up to round spermatids, DMC1 was detected in type B spermatogonia up to early pachytene spermatocytes. In SZA, IHC matched RT-PCR results. If present, SYCP3 was detected in nuclei of spermatocytes. DMC1 staining, in contrast to NSP, was localized in cytoplasm as well as in nuclei of spermatogonia and spermatocytes.

To conclude, development of SZA is not monocausal. Meiosis-related genes, especially SC genes and DMC1, showed an altered expression in SZA patients. Other genes were unaffected in our samples. Moreover, DMC1 seems to be involved also in transition from mitosis to meiosis, and not only in early HR events.

Poster-Preview 17:

Titel: Vitamin k- antagonist aggravates ckd induced neointimal hyperplasia and calcification in shunt veins: potential role for vitamin k2 to prevent avf failure

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Abstract:
Arteriovenous fistula (AVF) is the common vascular access type for hemodialysis patients. AVF failure is mainly due to neointimal hyperplasia (NIH). Vitamin K antagonists (VKA) are given to lower the thrombosis tendency, but have side effects but they dramatically enhance arterial calcifications. In this study we investigated the effects of VKA and vitamin K2 (K2) supplementation on NIH development and calcification in arterialized veins.

AVF was generated in 190 rats. Chronic kidney disease (CKD) was induced using adenine- enriched diet. Effects of CKD, VKA and K2 on AVF remodeling were evaluated using histology, morphometric analysis and immunohistochemistry. Examination of native and arterialized human veins was performed.

Arterialization, CKD ($p<.001$) and VKA ($p<.0001$) significantly enhanced venous NIH. K2 supplementation additional to VKA reduced NIH in arterialized veins ($p<.05$) in healthy animals. Arterialization, CKD ($p<.005$) and VKA treatment ($p<.002$) increased calcification while K2 supplementation attenuated calcification in healthy and CKD animals. K2 enhanced matrix Gla protein (MGP) carboxylation in control ($p<.05$) and CKD ($p<.002$) animals. Human vein samples contained inactive MGP at calcification and NIH sites, clearly indicating local vitamin K-deficiency.

Our study shows that VKA treatment has detrimental effects on AVF remodeling. K2 supplementation reduced NIH and calcification indicating vasoprotective effects. In arterialized veins, K2 should be considered as therapeutic approach to prevent NIH and calcification.

Poster-Preview 18:

Titel: Embryonic lipid accumulation in response to lipid excess – the difference between embryoblast and trophoblast cells

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Abstract:
The lineage diversification to embryoblast and trophoblast cells is a developmental milestone in embryonic development. Both cell lineages are supplied with nutrients from the mother. Recently, we have shown that an insulin-dependent diabetes mellitus causes a maternal hyperlipidaemia and leads to a non-physiological high lipid accumulation in trophoblast and embryoblast cells in rabbit blastocysts. Our aim was to determine potential mechanisms of intracellular lipid accumulation in rabbit blastocysts derived from an in vivo diabetic and hyperlipidaemic in vitro environment.

To evaluate the effect of an increased environmental lipid level, blastocysts from healthy rabbits were cultured in vitro with a specified lipid mixture. For in vivo analysis blastocysts from diabetic rabbits were collected. Marker molecules involved in lipid metabolism and intracellular lipid accumulation were analysed separately in the embryoblast and trophoblast.

In trophoblast cells intracellular lipid droplets were increased after lipid stimulation. Markers of fatty acid oxidation (CPT1 and PPARα) and fatty acid synthesis (FASN) were down-regulated. In embryoblast cells intracellular lipid droplets and expression of genes encoding for fatty acid uptake (FATP4) and binding (FABP4), as well as PPARγ expression were increased, indicating that two different signalling pathways were activated in both cell lineages. Consistently with the results gained in vitro, embryos from diabetic rabbits revealed also an increased expression of FATP4, FABP4 and PPARγ in embryoblast cells and down-regulation of CPT1 and FASN in trophoblast cells.

We suppose that embryoblast and trophoblast cells adapt to hyperlipidaemic conditions in different ways, leading to the same phenotype in both cell lineages.