Posters

1	**Design of antimicrobial protein-lipid complexes: From purification to delivery of cytotoxic peptides granulysin and granzyme** B Owais Abdul Hameed, Mark Gontsarik, Stefan Salentinig, Michael Walch Infectious diseases are the leading cause of death worldwide. Aside from new infectious diseases emerging regularly, the increasing prevalence of antibiotic resistant strains present a global health crisis. There is an urgent need to develop new alternatives to the conventional antibiotics. The immune response by cytotoxic T lymphocytes (CTLs) and natural killer (NK) cells against various bacterial pathogens involves release of cytotoxic proteins, in particular of the antimicrobial effector protein granulysin and group of homologous serine proteases, the granzymes. These candidates for the targeted immunotherapy with lipid nanocarriers have potential to overcome their limited applications due to the chemical instability in biological environments, low aqueous solubility, and host toxicity when employed at high concentrations. From novel purification system of these peptides to the design and characterization of effective self-assembled nanocarriers with pH-enabled antimicrobial activity providing targeted delivery of such antimicrobial peptides. Biological in vitro assays against Escherichia coli showed high antimicrobial activity of the positively charged oleic acid (OA) and granulysin aggregates at pH 5.0, while negligible antimicrobial activity was observed at pH 7.0 for the negatively charged nanocarriers. The ability to switch their biological activity “on” and “off” in response to changes in pH has potential to focus the antimicrobial peptides’ action to areas of specific pH in the body. This study presents a promising strategy against antibiotic resistant bacteria while protecting the beneficial microbiome in the body and eliminating adverse effects further opening a new door for efficient immunotherapies. University of Fribourg, Faculty of Science and Medicine
2	**Body painting, ultrasound, clinical investigation and peer-teaching: a student-orientated unconventional approach to enhance anatomy learning** Alessandro Bilella (1, 2), Elisabeth Eppler (2), Karl Link (1), Luis Filgueira (1) (1) Anatomy, Department of Medicine, University of Fribourg, Switzerland; (2) Institute of Anatomy, Faculty of Medicine, University of Bern, Switzerland Traditional anatomy teaching to medical students consists in one-way procedure (from teacher to students) of lectures combined with practical courses, such as dissection, prosection or specimen. We present a new approach to enhance the learning of clinical-orientated anatomy of the musculoskeletal system. From 2016 to 2021, five cohorts of second-year medical students voluntarily experimented this new procedure at the University of Fribourg. The new course covered the musculoskeletal anatomy of the upper and lower limb from a clinical perspective, combining clinical investigation and ultrasound examination of the joints and studying the physiological/anatomical conditions and pathological alterations of the musculoskeletal system. The course aimed to enhance the acquisition of knowledge in anatomy and physiology, and to offer a maximum of student-focused and hands-on learning. The students rotated through three activities, i.e. body painting, clinical investigation, and ultrasound, under the supervision of a faculty member or an experienced medical doctor. The students followed an introduction and prepared a PowerPoint presentation related to the clinical anatomy. At the end of the course, the students reported on their own learning experience through a personal journal summarizing the work and topics treated and by voluntarily answering an anonymous online questionnaire (SurveyMonkey). The latter had a special emphasis on the impact of such an optional course on the improvement of students’ knowledge of the musculoskeletal system. The analysis of the journal reports and answers given via the questionnaire revealed that the aim of the course was fully achieved. Overall, the students appreciated the course, with 78% showing very high interest and 22% showing interest. For 58% of the students, the course helped to enhance their knowledge of anatomy, while

	42% of the students the course stabilized it. All the students recommend the course to their younger peers. University of Fribourg, University of Bern, Anatomy, Department of Medicine
3	**Joint hypermobility syndrome: is there any way for experimental modelling?** Andrii Fedotchenko **Introduction.** Joint hypermobility syndrome (JHS) is the most common heritable disorder of connective tissue. Scientists always have faced the challenge of experimental modelling of various pathologies for an in-depth study and to explore opportunities for their treatment. We studied joint morphology using hip joints of Wistar rats in norm and after antigenic stimulation. The last one was modeled through the transuterine intrafetal interscapular subcutaneous injection of antigen (0.05 ml of human normal immunoglobulin) for the rat foetus on the 18th day of its antenatal life under general anesthesia and sterile conditions via laparotomy for the pregnant female (M. A. Voloshyn method (1981). The control group of rats was injected with 0.05 ml of physiological saline in the same manner. Our previous studies have detected a decreased quantity of sulfated glycosaminoglycans and mannose conjugates together with an increased quantity of fucose conjugates in the hip joint and skeletal disproportions (limb segments) of antigen-suppressed rats. **Materials and methods.** Joints were fixed, decalcified and dehydrated. Paraffin-embedded tissue specimens were stained by Mallory and Hart's elastin stains. Components of the joint capsule were analyzed by light-microscope, intersection-point counting method. The obtained data were statistically processed. **Results.** In the joint capsule of antigen-suppressed rats we found also a decreased total quantity of collagen fibers (through the arranged collagen fibers) till the 60th day inclusive together with an increased number of elastic fibers, disarranged collagen fibers till the 45th day inclusive and ground substance till the 14th day inclusive. **Conclusion.** The above-mentioned phenomena might create preconditions for joint capsule hypermobility and joint weakness in general, thereby explaining chronic musculoskeletal pain and fatigue during it. We would suggest the intrafetal antigen injection for Wistar rats as an experimental model to study the morphology of the JHS. Zaporizhzhia State Medical University. Chair of Human Anatomy. Zaporizhzhia, Ukraine. Wilhelmsburger Krankenhaus Groß Sand. Hamburg, Germany
4	**Anatomy learning under COVID-19 measures: A real world experiment.** Luis Filgueira, Nils Lannes, Alexey Larionov Introduction: The best way to teach human gross anatomy to medical students has often been debated, although recent studies emphasize on the benefits of dissection-based courses (DOI 10.1002/ase.1859). Prosection and model-based courses, online and virtual reality approaches have been introduced over time, due to reduction in teaching time, and due to economic reasons with decreasing budget. In 2020, we were challenged by the pandemic of SARS-CoV-2 and corresponding COVID-19 public health measures. We had to change and adapt several times anatomy teaching and learning over the two semesters for the cohort of 112 first/second year undergraduate medical students. With the experience of this student cohort with various anatomy learning approaches, we wanted to know what anatomy teaching method was best perceived by the students. Methods: 112 medical students were asked about their learning experience with various anatomy teaching approaches for 2020, including 1) dissection-based learning, 2) prosections and models-based demonstrations, 3) structured self-directed learning using written instructions and corresponding anatomical material, 4) structured self-directed learning using online instructions via Moodle and an online anatomy program (https://e-learn.anatomy.uzh.ch/Anatomy/Anatomy.html ; German version). All students agreed to participate in the study. The questionnaire was administered by https://www.surveymonkey.com and done anonymously. To date 66/112 (59%) students replied.

	Results: The majority of the students (92%) preferred the dissection course as their main anatomy learning method, 80% of students were happy with a demonstration course using prosections and anatomical models. Only a minority liked to have structured self-directed learning with either anatomical specimens and models (42%) or with online material (36%). Rather few students preferred learning with an online course (18%) or through unstructured self-directed learning using their own resources (12%). For the dissection course the students appreciated the 3 dimensional aspect (92%), the real size appearance of the structures (90%), the active explorative approach (81%), the topographical relationship of the structures (81%) and anatomical variabililiy (72%). During complete lock-down of the COVID-19 measures when the students had to stay isolated at home for several weeks, the majority of students thought that the offered online structured self-directed course was most adequate under those circumstances (55%), but would otherwise not be the preferred replacement of a dissection course (55%). Conclusion: Evaluation of student experience with a variety of anatomy learning methods clearly indicate that a structured dissection-based course with additional anatomical demonstration would be preferred by the majority of our students. Online material would be beneficial as additional learning support. However, the majority of students agreed that, under strict COVID-19 stay-at-home measures, the structured self-directed online course was the best way to learn anatomy. University of Fribourg
5	**Whole genome CRISPR screening to identify novel regulators of desmosomal adhesion** Henriette Franz, Chiara Alessandra Noemi Stüdle, Volker Spindler Desmosomes are essential junctions to facilitate strong intercellular adhesion. They are dysregulated in different diseases such as pemphigus, a blistering skin disease, and arrhythmogenic cardiomyopathy. The mechanisms how desmosomes are regulated, however, are only partially understood. In this study, we applied a CRISPR screen as unbiased approach to identify novel candidates for desmosome regulation. We used a human keratinocyte cell line (HaCat) stably expressing Cas9 and transduced it with a lentiviral pool of small guide RNAs (sgRNAs) targeting each protein-coding gene with four individual sgRNAs. After selection and expansion, the cells were sorted according to the protein levels of the desmosomal adhesion molecule desmoglein 3 (DSG3). $DSG3^{high}$ and $DSG3^{low}$-containing cells were subjected to next generation sequencing and evaluated for enrichment of sgRNA sequences. 56 candidates for positive and 27 candidates for negative regulation of DSG3 were found. One of these candidates for positive regulation of DSG3 levels in keratinocytes is histone deacetylase 3 (HDAC3). CRISPR-mediated knockout of *HDAC3* or treatment with specific HDAC3 inhibitors in an independent cell line confirmed a downregulation of *DSG3* mRNA, a decrease of DSG3 levels at the cell membrane and a reduction of intercellular adhesion. However, deacetylation of histone lysines by HDAC3 leads to transcriptional repression of genes and published ChIP-seq data of the human epidermis revealed no localization of HDAC3 at the *DSG3* promotor. This suggests an indirect regulation of DSG3 by HDAC3 via a two-step process. Interestingly, C/EBP (CCAAT/enhancer binding protein), motifs are significantly enriched in the DSG3 promotor sequence, HDAC3 is located at the promotor of C/EBP family members promotor and C/EBP mRNA is upregulated upon *HDAC3* knockout or inhibitor treatment. Thus, C/EBPs might be directly involved in DSG3 transcriptional regulation and could be new targets for treatment of desmosome-related diseases. University of Basel, Department of Biomedicine
6	**Computed tomography osteoabsorptiometry (CT-OAM) for imaging of degenerative disc disease** Max HP. Gay, Magdalena Müller-Gerbl Lower back pain is a common condition with significant morbidity and economic impact. The pathophysiology is poorly understood but is in part attributable to degenerative disc

	disease (DDD). The healthy intervertebral disc ensures spine functionality by transferring the perceived load to the caudally adjacent vertebrae. The exposure to recurring mechanical load is mirrored in the mineralization pattern of the subchondral bone plate (SBP), where increased bone density is a sign of repetitive localized high stress. CT-osteoabsorptiometry (CT-OAM) is a technique based on conventional CT scans that displays the mineral density distribution in the SBP as a surface-colour map. This study measured and compared the SBP mineral density distribution patterns of healthy lumbar discs and those displaying DDD. The inferior SBP, adjacent to degenerating disc, display an 18-41% higher relative calcium concentration than their healthy counterparts. The opposing superior SBPs the relative calcium content is significantly increased. Whereas it is reasonably consistent for L1-L3 (L1: 132%, L2: 127%, L3: 120%), the increase grows in caudal direction (L4: 131%, L5: 148%, S1: 152%). Furthermore, a change in the areal distribution of excessive mineralization can be differentiated between healthy and diseased motion segments. The acquired data provide in vitro proof of the mechanical and anatomical properties of the SBP in relation to the state of disc degeneration. In conjunction with diagnostic use of CT-OAM, our data provides a basis for a non-invasive and sensitive technique that correlates with disc functionality. This could be promising in various cases, including but not restricted to: • identifying early stages of DDD that would warrant pre-emptive conservative therapies • tracking time-dependent changes in respect to disease progression in patients • assessing the repercussions of surgical procedures (spinal fusion or implant of disc prosthesis) on neighbouring joint segments • evaluating the outcome of experimental regenerative therapies (tissue engineering and cell therapy) University of Basel, Department of Biomedicine
7	**Influence of Macronutrients on Heart Regeneration in Zebrafish** Nick Kirschke According to the WHO, Cardiovascular Diseases are the leading cause of death globally - more than 17 million deaths per year. After an heart attack the wound area is repaired into a scar to avoid any leaking of blood. That's why the heart looses parts of its contractility because there are less cardiomyocytes (CM) which are able to contract. In contrast to humans there are several other animal species such as zebrafish that have the capacity to regenerate the heart in response to an injury by allowing the proliferation of preexisting CMs. A prerequisite that has been found to be very important is that these CMs undergo a metabolic switch from oxidative phosphorylation to glycolysis. Given that metabolic changes are promoting regeneration we wondered whether different diets could lead to a better or worse regenerative capacity. If that might be the case, easy lifestyle change might increase CM's regenerative capacity and leads to a better outcome after a myocardial infarction.Therefore we fed fish with different diets for several weeks before we performed a cryoinjury. One week later we extracted the hearts and performed immunohistochemistry in order to see replicating CMs. So far we were able to perform a few diets in which we could find a slight reduced number of BrdU+ cells in a high fat diet compared to the control diet. The regenerative capacity of other diets are yet to find out. University of Bern, Institute of Anatomy
8	**Human microglia cell in vitro models as a predictive tool for investigating inflammatory-related virus infection of the brain** Nils Lannes[1] Isabelle Fellay[1], Obdullio Garcia-Nicolas[2], Amal Fahmi[3], Benoît Fellay[4], Marco Polo Alves[3], Artur Summerfield[2, 3,] Luis Filgueira[1] [1] Institute of Anatomy, Department of Medicine, University of Fribourg, Switzerland [2] Institute of Virology and Immunology, Mittelhäusern, Switzerland

	[3] Department of Infectious Diseases and Pathobiology, Vetsuisse Faculty, University of Bern, Switzerland [4] HFR Fribourg, Hôpital Cantonal, Fribourg, Switzerland. Microglia are a unique residential immunocompetent cell type of the central nervous system (CNS). Microglia maintain the homeostasis of the CNS and rapidly activate upon insults. Microglia are the major contributors of neuroinflammation, suggested to be one of the most common pathology in CNS diseases. In infection, the engagement of Toll-like receptors (TLRs) leads to microglial activation and production of inflammatory factors. Here, we propose to identify accessible and reliable human microglia in vitro models as a predictive tool for TLR detection of viruses infecting the human brain. To this end, the responsiveness of human blood monocyte-derived microglia models and human brain-derived primary microglia to specific TLR ligands as well as viral infection will be compared. The three monocyte-derived microglia models shared strong homology and were distant from dendritic cells, macrophages and monocytes. Despite heterogeneous population in culture, fully differentiated CX3CR1+ CD11b+ microglia were the main subset. Cells expressed transcripts for all human TLRs (TLR1-10) and were reactive to TLR1-9 agonists by modifying the transcriptome. The most modified genes were inflammatory of the interferon-inducing and -responsive pathways and migratory via chemokine ligands/receptors. Focusing on specific TLR, intracellular proteins were found for TLR7, 8 and 9, but not TLR3 although TLR3 engagement lead to the most distant transcriptome compared to untreated cells. None of the selected TLR ligands modified the phenotype nor the frequency of CX3CR1+ CD11b+ microglia as well as induced cell death. However, TLR ligands influenced the expression of ACE-2, the receptor for SARS-Cov-2. Nevertheless, no evidence of viral infection and propagation of SARS-Cov-2 are yet shown in human microglia. This contrasts with Japanese encephalitis virus where virus replication can take place. In conclusion, human microglia models may be suitable for the study of emerging viruses invading the CNS in order to reveal TLR-dependent pathways and potential therapeutic targets. University of Fribourg, Institute of Anatomy
9	**The anatomy of the piriformis revised – New explanation of the piriformis syndrome** Alexey Larionov, Peter Yotovski, Luis Filgueira Background: The piriformis syndrome is a pathological condition characterized by extreme contraction of the piriformis belly and sciatic nerve compression in the gluteal region leading to pain (radiating down or into the leg) dysesthesias in the buttock area, dyspareunia, and intolerance to sitting long time [1]. The prevalence of piriformis syndrome fluctuated from 0.3% to 36 % among patients with low back pain [2]. The classical treatment approach (steroid injection to the piriformis or area near to sciatic nerve) is not always effective, and genuine sciatic nerve entrapment is found only in 6-8 % of all cases [3]. Anatomic relationships of piriformis origin with the sacral nerves in the pelvic cavity are still poorly understood and needed to be studied in detail. We speculated that relationships between the piriformis origin and sacral plexus portions in the pelvis might be responsible for the initiation and progression of the piriformis and pelvic pain syndromes. Methods: Dissection of 40 unpaired hemipelvises (14 males and 26 females) with lumbar region and leg attached were used for the analysis of the topographic interconnections between the piriformis (from the sacral foramina) and sacral plexus spinal nerves. Results: Based on the muscle position on the sacral vertebrae and around or over the sacral vertebral foramina and also regarding the crossing of the ventral branches of the sacral spinal nerves before forming the sacral plexus, four patterns of piriformis origin and nerve spacing were identified 1) medial pattern of piriformis origin with the ventral branches of the spinal nerves (S2 segment) enclosed by the piriformis (60 %, n=24); 2) the medial pattern of piriformis origin without the piriformis enclosing the nerves (15%,n=6); 3) interforaminal piriformis origin pattern with the free course of the nerves (15%, n=6), and 4) lateral piriformis origin pattern with the free course of the nerves (10%, n=4). The piriformis received innervation in 40% (n=16) of cases from the S1, in 20 %(n=8) from lumbosacral trunk, in 20% (n=8) from S2, and in 20% (n=8) from S1 and S2 sacral

	segments. Conclusions: the cadaveric study revealed that anatomic relations “Piriformis-Ventral branches of the sacral spinal nerves is a potential direct predisposition factor for the development of the piriformis syndrome. 1. Halpin RJ, Ganju A. Piriformis syndrome: A real pain in the buttock? Neurosurgery. 2009; 65: 197–202. doi: 10.1227/01.NEU.0000335788.45495.0C. 2. Siddiq M, Rasker J. Piriformis syndrome: Still unsolved issues. Int J Clin Rheumtol. 2018; 13: 338–40. 3. Probst D, Stout A, Hunt D. Piriformis Syndrome: A Narrative Review of the Anatomy, Diagnosis, and Treatment. PM&R [Internet]. 2019; 11: 54–63. doi: 10.1002/pmrj.12189 University of Fribourg, Faculty of Science and Medicine, Anatomy
10	**The role of TGFβ signaling in melanoma disease progression and treatment response** Benjamin Loos, Julien Debbache, Rishika Pandya, Jelena Vasilevska and Lukas Sommer Due to its vast metastatic potential and resilience to conventional therapy, melanoma remains the most aggressive and deadliest type of skin cancer. Genes that are commonly mutated (gain of function) in melanoma are involved in the MAPK pathway, as for example *NRAS* or *BRAF*. In clinics, targeted therapy, that can selectively inhibit the MAPK pathway is used as golden standard to treat melanoma patients. However, although proven to be effective in many cases, targeted therapy can be overcome by melanoma cells. Following a model which is known as “phenotype switching”, melanoma cells undergo conversions, from proliferative states to invasive states and vice versa, a helpful feature to disseminate the disease across the body. Of note, cells in this invasive state are more resistant to targeted therapy. Transforming growth factor beta (TGFβ) is known for its context dependent mode of action on cellular states and behavior. One of its functions on melanoma cells is to induce epithelial to mesenchymal transition (EMT), a phenotype switch from a residual, proliferative state to a migrating, invasive state. Interestingly we could show in *in vitro* experiments that in a specific context, TGFβ is able to boost apoptosis of melanoma significantly. This context is reflected in scRNAseq data sets of patient biopsies from a longitudinal study of patients on treatment. How this new finding can be exploited as therapeutic modality for melanoma patients is topic of further research. University of Zürich, Institute of Anatomy
11	**The human masseter muscle revisited: first description of its coronoid part** Mezey, Szilvia E. [1], Müller-Gerbl, Magdalena [1], Mireille, Toranelli [1], Türp, Jens C. [2] [1] Anatomical Institute, Department of Biomedicine, University of Basel [2] University Center for Dental Medicine Basel UZB, University of Basel The masseter muscle is considered to be bilayered, consisting of a superficial and a deep part. However, a few historical texts mention the possible existence of a third layer as well, but they are extremely inconsistent as to its position. Here we performed an anatomical study to clarify the presence and morphological characteristics of a distinct third layer of the masseter muscle. We dissected 12 formaldehyde-fixed human cadaver heads, analysed CTs of 16 fresh cadavers, evaluated MR data from one living subject and examined histological sections using methyl methacrylate embedding of one formaldehyde-preserved head. An anatomically distinct, deep third layer of the masseter muscle was consistently demonstrated, running from the medial surface of the zygomatic process of the temporal bone to the root and posterior margin of the coronoid process. Ours is the first detailed description of this part of the masseter muscle. To facilitate discussion of this newly described part of the masseter, we recommend the name M. masseter pars coronoidea (coronoid part of the masseter) as a further reference. The arrangement of its muscle fibers suggest it being involved in stabilising the mandible by elevating and retracting the coronoid process. University of Basel, Department of Biomedicine

12	**Dolichyl-Phosphate Mannosyltransferase (DPM) complex is a novel modulator of desmosomal adhesion** Maitreyi Rathod, Henriette Franz, Chiara Stüdle, Volker Spindler Desmosomes are vital mediators of intercellular cell to cell adhesion and have direct implications in several pathogenic conditions such as pemphigus, cancer metastasis, and arrhythmogenic cardiomyopathy. Desmosomes are complex units, which are differentially modulated depending on cellular context, however the underlying mechanisms in normal and disease context are only partially understood. Thus, in an attempt to identify novel internal regulators of desmosomes, a CRISPR screen was performed in human keratinocytes (HaCaT). Components of the Dolichyl-Phosphate Mannosyltransferase (DPM) complex appeared as significant negative regulators of the desmosomal adhesion molecule desmoglein 3 (DSG3). DPM complex proteins serve as regulators of glycosylation, the latter of which is an important post translational modification dictating intracellular trafficking and protein turnover. To test how DPM proteins could modulate DSG3-mediated cell adhesion, we created CRISPR-mediated knockouts (KO) of DPM1, DPM2 and DPM3 in HaCaT background. DPM1^{KO} cells showed a significant increase in the amount of DSG3 and desmocolin 3 (DSC3) at the cell surface. As a measure of the functional impact of enhanced DSG3 and DSC3, we performed dispase-based dissociation assays to evaluate intercellular adhesion. Surprisingly, we detected significantly reduced cell-cell adhesion in DPM1^{KO} cells. In order to understand this observation apparently contradicting the increased membrane levels of DSG3 an DSC3, we tested the sub-cellular localization of other desmosomal components. Desmoglein2 and desmoplakin showed a dramatic reduction at the cell borders, indicating profound impairment of normal desmosome composition in the absence of DPM1. These data suggest a differential contribution of DPM-mediated glycosylation to distinct subsets of desmosomal molecules. Understanding how DPM1 modulates cell adhesion would add valuable insights into the basic regulation of desmosomes, which could then be used to answer disease relevant questions in the future. University of Basel, Department of Biomedicine
13_1	**Adhesion of Pancreatic Tumor Cell Clusters by Desmosomal Molecules enhances Early Liver Metastases Formation** Camilla Schinner, Niclas Dietrich, Volker Spindler Pancreatic cancer with the most common form of pancreatic ductal adenocarcinoma (PDAC) is one of the most devastating tumor entities with a mean 5-year survival of 10%. Metastatic tumor spreading mainly to the liver is a key step in disease progression. We recently demonstrated that the desmosomal adhesion molecule desmoglein-2 (DSG2) suppresses malignant behaviour of pancreatic cancer cells. In this study, we investigate the role of DSG2 for early liver metastasis formation of pancreatic tumor cells. We established a murine ex vivo liver perfusion model to mimic the initial step of metastasis formation and generated CRISPR/Cas9 based KOs of DSG2 in AsPC1 pancreatic cancer cells. In liver perfusion experiments, less DSG2 KO compared to control cells were detectable in wild type livers. To investigate the relevance of tumor cell attachment to hepatocytes by desmosomal adhesion in this context, liver specific KO mouse models for DSG2 and a second desmosomal adhesion molecule, desmocollin-2 (DSC2) were generated. However, neither depletion of DSG2 nor DSC2 led to altered retention of tumor cells in the liver. Importantly, perfused control tumor cells form multicellular clusters, which were also detectable in the respective livers after perfusion. In contrast, DSG2 KO tumor exhibited reduced cell-cell adhesion with smaller cell clusters up to single cells. This suggests that retention of tumors cells during perfusion of the liver depends on the cell cluster size. Accordingly, perfusion of control cell clusters, which were dissociated into mostly single cells by trypsin treatment, resulted in reduced numbers of cells remaining in the perfused livers. In conclusion, for early metastasis formation with initial attachment of cells, clustering of perfused tumor cells by desmosomal adhesion is more relevant than cohesion to hepatocytes. This suggests that targeting desmosomal adhesion in circulating tumor cell clusters may serve as novel therapeutic option.

	University of Basel, Department of Biomedicine
13_2	**Defective Desmosomal Adhesion Induces Arrhythmogenic Cardiomyopathy by an Integrin-αVβ6/TGF-β1 Signaling Cascade** Camilla Schinner[1], Henriette Franz[1], Aude Zimmermann[1], Marie Wanuske[1], Florian Geier[2,3], Pawel Pelczar[4], Vera Lorenz[5], Lifen Xu[5], Chiara Stüdle[1], Brit-Maria Beckmann[6], Piotr I Maly[1], Gabriela M Kuster[5,7], Volker Spindler[1,*] [1] Department of Biomedicine, Section Anatomy, University of Basel, Basel, Switzerland [2] Department of Biomedicine, Bioinformatics Core Facility, University Hospital Basel, Basel, Switzerland [3] Swiss Institute of Bioinformatics, Basel, Switzerland [4] Center for Transgenic Models, University of Basel, Basel, Switzerland [5] Department of Biomedicine, University Hospital Basel and University of Basel, Basel, Switzerland [6] Department of Legal Medicine, University Hospital Frankfurt, Frankfurt am Main, Germany [7] Division of Cardiology, University Hospital Basel, Basel, Switzerland **Background** – Arrhythmogenic Cardiomyopathy (ACM) is characterized by progressive loss of cardiomyocytes with fibrosis, ventricular systolic dysfunction and arrhythmias. ACM is mainly caused by mutations in genes of the desmosomal cell-cell adhesion complex. Even though the pathological phenotype is known, the underlying mechanisms are not well understood and treatment options are only symptomatic. Here, we investigate the relevance of defective desmosomal adhesion for ACM and its path to disease development employing the DSG2-W2A mouse model. **Methods** – We mutated the binding site of desmoglein-2 (DSG2), a crucial desmosomal adhesion molecule in cardiomyocytes. The DSG2-W2A mutation abrogates the tryptophan swap, a central binding mechanism based on structural data. Impaired adhesive function of DSG2-W2A was confirmed by cell-cell dissociation assays and force spectroscopy measurements. We generated a DSG2-W2A knock-in mouse model, which was analysed by echocardiography and histological and bio-molecular techniques including RNA sequencing, transmission electron and super-resolution microscopy. The results were compared to ACM patient samples and their relevance was confirmed in cardiac slice cultures. **Results** – The DSG2-W2A mutation induced impaired binding and desmosomal adhesion dysfunction on cellular and molecular level. Mice bearing this mutation develop a severe cardiac phenotype recalling the characteristics of ACM, including fibrosis, impaired systolic function and arrhythmia. A comparison of the transcriptome of mutant mice with ACM patient data suggested integrin-αVβ6 and subsequent TGF-β signaling as driver of cardiac fibrosis. Accordingly, blocking antibodies targeting integrin-αVβ6 or inhibiting TGF-β receptor both led to reduction of fibrosis markers in cardiac slice cultures. **Conclusion** – We show that disruption of desmosomal adhesion is sufficient to induce ACM, which is in line with the dysfunctional adhesion hypothesis. Mechanistically, deregulation of integrin-αVβ6 signaling was identified as central step towards fibrosis. This highlights the value of this model to discern mechanisms of cardiac fibrosis and to identify and test novel treatment options for ACM. University of Basel, Department of Biomedicine
14	**A new knock-in mouse model of SCA14 with increased PKCγ activity shows perturbed Purkinje cell maturation and ataxic motor behavior** Etsuko Shimobayashi and Josef P. Kapfhammer Spinocerebellar ataxias (SCAs) are diseases characterized by cerebellar atrophy and loss of Purkinje neurons caused by mutations in diverse genes. Spinocerebellar ataxia type 14 (SCA14) is caused by missense mutations or deletions in the Protein kinase C γ (PKCγ) gene, which is a crucial signaling protein in Purkinje cells. It is still unclear whether increased or decreased PKCγ activity may be involved in the SCA14 pathogenesis.

	In this study we present a new knock-in mouse model related to SCA14 with a point mutation in the pseudosubstrate domain, PKCγ-A24E, known to induce a constitutive PKCγ activation. In this protein conformation, the kinase domain of PKCγ is activated, but at the same time the protein is subject to dephosphorylation and protein degradation. As a result, we find a dramatic reduction of PKCγ protein expression in PKCγ-A24E mice. Despite this protein reduction there is clear evidence for an increased PKC activity in Purkinje cells from PKCγ-A24E mice. Purkinje cells derived from PKCγ-A24E have short thickened dendrites typical for PKC activation. These mice also develop a marked ataxia, even in a heterozygous state corresponding to the human disease situation and signs of Purkinje cell dysfunction making them an interesting new mouse model of SCA14. Recently, a similar mutation in a human patient was discovered and found to be associated with overt SCA14. RNA profiling of PKCγ-A24E mice showed a dysregulation of related signaling pathways like mGluR1 or mTOR. Our results show that the induction of PKCγ activation in Purkinje cells results in the SCA-like phenotype indicating PKC activation as one pathogenetic avenue leading to a SCA. University of Basel, Department of Biomedicine
15	**Uncovering topography and extent of injury to the superior mesenteric plexus at right colectomy with extended D3 mesenterectomy. A composite multimodal 3-Dimensional analysis.** Bojan V. Stimec [3,] Javier A. Luzon [1,2], Dejan Ignjatovic [1,2] [1] Institute of Clinical Medicine, Faculty of Medicine, University of Oslo, Oslo, Norway [2] Division of Surgery, Department of Digestive Surgery, Akershus University Hospital Lørenskog, Norway [3] Anatomy Sector, Teaching Unit, Faculty of Medicine, University of Geneva, Geneva, Switzerland Aims. To describe the superior mesenteric artery plexus (SMAP) and extent of injury after right colectomy with extended D3 mesenterectomy for cancer. Methods. Specimen groups - Group I: Anatomical dissection specimens (formalin-fixed). Group II: Postmortem D3 volume specimens (fresh); Group III: Surgical D3 volume specimens (after division from surgical specimen). Histological analysis consisted of nerve fiber count, diameter and area measurements (Aperio ®), with comparative analysis between groups II and III. 3D-models were generated from both nano-CT and 3D histology reconstructions. Results. 21 specimens were collected. Group I: 5(3 females, age 80-93y). Group II: 7(5 females, age 71-86y) and Group III: 9 (5 females, age 55-69y). Anatomical dissection (group I): the SMAP is a complex mesh-like structure, surrounding the superior mesenteric artery (SMA) within its vascular sheath (thickness 1.04-2.05mm), branching out and following peripheral arteries. Histology (group II): vascular sheath thickness 1.17-2.62mm. Nerve count: 53±12.42 (38-68). Nerve diameter: 648.82±79,17µm (537.88-798.60). Total nerve area: 1.84±0.50mm^2 (1.16-2.29). Histology (group III): nerve count 31.6±6.74 (range 23-43); nerve diameter 504.29±66.52µm (range 417.08-597.65); total nerve area was 0.889±0.45mm^2 (range 0.479- 1.668). 3D-models present numerous nerve fibers within the vascular sheath, while lymph nodes were observed exclusively outside, both in front and behind the superior mesenteric vessels, in all specimens. SMAP injury is 48 and 59% when area and nerve count are analyzed, respectively. Nano-CT imaging show nerve fibers inclination angle of 35º. This indicate an even more extensive injury to the plexus while performing a longitudinal resection along the SMA. Average length of D3-specimens is 4.24cm. Conclusions. The SMAP lies exclusively within the SMA sheath, which does not contain lymph nodes. Nerve fiber inclination along the superior mesenteric vessels, imply a complete injury of the SMAP when performing a right colectomy with extended D3 mesenterectomy. University of Geneva, Anatomy Sector, Faculty of Medicine

16	**Dsc1 sustains keratinocyte proliferation in 3D reconstructed human epidermis** Marie-Therès Wanuske, Chiara Stüdle, Volker Spindler **Objective:** Desmosomes are cell-cell junctions that are indispensable for maintaining tissue integrity in organs such as the heart and skin. In the four distinct layers of the epidermis (basal, spinous, granular and corneal), the expression of differentiation markers and desmosomal molecules follows a complex pattern, suggesting their role in epidermal differentiation. However, the distinct functions of these molecules still remain unclear. Here, we investigated the role of desmocollin 1 (Dsc1), a desmosomal cadherin expressed from the upper spinous layer outwards, in epidermal differentiation using a 3D model of reconstructed human epidermis (3D raft). **Methods:** 3D rafts were generated by culturing adult human foreskin keratinocytes at the air-liquid interphase. shRNA or overexpression approaches were utilized to diminish or increase Dsc1 levels. 3D rafts were studied by HE-staining, immunostaining and FACS analysis. **Results:** Silencing of Dsc1 led to decreased 3D raft thickness. In line with this, expression of the proliferation marker Ki67 was reduced in the basal layer of the epidermis. While the expression and distribution of filaggrin proved regular differentiation towards the granular layer, investigation of the epidermal layers revealed a specific thinning of the stratum spinosum under Dsc1 knockdown conditions. Vice versa, overexpression of Dsc1 resulted in thicker 3D rafts compared to control. As these data suggest an indirect regulation of proliferation in the basal layer by Dsc1, we co-cultured monolayers of primary keratinocytes with 3D rafts. Around the third day of shDsc1 3D raft differentiation, proliferation of monolayer cells was diminished, suggesting that secreted factors in response to Dsc1 expression regulate cell proliferation in the basal cell layer and therefore epidermal thickness. **Conclusions:** Here, we show that Dsc1 sustains proliferation in an epidermal 3D model, which may be explained by paracrine signaling dependent on Dsc1. University of Basel, Department of Biomedicine
17	**Serine/threonine kinase 17b signaling regulates Purkinje cell dendritic development and is altered in multiple spinocerebellar ataxias** Qin-Wei Wu, Josef P. Kapfhammer Serine/threonine kinase 17b (STK17B, also known as DRAK2) is known to be a downstream effector of Protein Kinase C (PKC) in the immune system, in particular in T-lymphocytes. PKC activity also plays a critical role for dendritic development and synaptic maturation and plasticity in cerebellar Purkinje cells. We present evidence that STK17B is strongly expressed in mouse cerebellar Purkinje cells starting in the early postnatal period and remaining highly expressed throughout adult stages and that STK17B is a target of PKC phosphorylation in the cerebellum. STK17B overexpression potentiates the morphological changes of Purkinje cells seen after PKC activation suggesting that it is a downstream effector of PKC. A phosphorylation mimetic STK17B variant induced a marked reduction of Purkinje cell dendritic tree size whereas the inhibition of STK17B with the novel compound 16 (Cpd16) could partially rescue the morphological changes of the Purkinje cell dendritic tree after PKC activation. These findings show that STK17B signaling is an important downstream effector of PKC activation in Purkinje cells. Furthermore, STK17B was identified as a molecule being transcriptionally downregulated in mouse models of SCA1, SCA7, SCA14 and SCA41. The reduced expression of STK17B in these mouse models might protect Purkinje cell dendrites from the negative effects of overactivated PKC signaling. Our findings provide new insights in the role of STK17B for Purkinje cell dendritic development and the pathology of SCAs. University of Basel, Department of Biomedicine