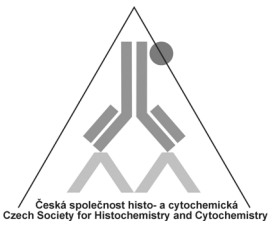
Czech Anatomical Society
Czech Society for Histochemistry and Cytochemistry
Charles University in Prague
First Faculty of Medicine



MORPHOLOGY 2011
46rd International Congress on Anatomy
48rd Lojda Symposium on Histochemistry

Under the Auspices of

Prof. MUDr. Jan Škrha, DrSc., MBA
Prorector of The Charles University in Prague

Prof. MUDr. Tomáš Zima, DrSc., MBA
Dean of The Charles University First Faculty of Medicine

Doc. MUDr. Bohuslav Svoboda, CSc.
Lord Mayor of The City of Prague

Ing. Jiří Paluska
Mayor of Prague 2

Scientific Committee

R. Druga, M. Grim, P. Hach, J. Mokrý, K. Smetana, J. Stingl, L. Vajner

Organizing Committee

V. Báča, H. Brichová, R. Druga, B. Dvořánková, M. Grim, T. Kučera, E. Kluzáková, E. Krejčí, L. Lacina, O. Naňka, D. Sedmera, K. Smetana, P. Šnajdr, E. Vancová

Topics

Stem Cells, Morphogenesis, Neurosciences, Clinical Anatomy, Experimental Medicine, Teaching of Morphology, History, Various Topic

Prague, September 4 – 7, 2011

General Information

Venue

Institute of Anatomy, First Faculty of Medicine, U Nemocnice 3, CZ-128 00 Prague 2,
phone: +420 224 965 780, fax. +420 224 965 770
e-mail: anat@lf1.cuni.cz, web page: cas.lf1.cuni.cz

Registration and Information Desk

Ground level of the Institute of Anatomy, Charles University First Medical Faculty,
U Nemocnice 3, Prague 2, phone: 224 96 5 718
Office Hours:

Sunday, September 4, 2011	15:00 - 17:00
Monday, September 5, 2011	08:00 - 17:00
Tuesday, September 6, 2011	08:00 - 17:00
Wednesday, September 7, 20011	08:00 - 12:00

Languages

English, Czech, Slovak

Oral Presentations

Plenary lectures - 20 min, lectures - 10 min, discussion - 5 min. Windows PC with CD-ROM and USB port are installed in lecture halls. Presentation software is Windows XP/PowerPoint XP or PowerPoint 2007. Videos can be presented only through PowerPoint presentation. Speakers are kindly asked to hand in their presentations directly to the attendant at the lecture hall before beginning of each session. Participants can check their presentations and use the internet access in the e-Point during the congress from 8:00 until 17:00.

Poster Presentation

The size of poster panel is 130 cm (height) and 90 cm (width). All posters will be displayed from Monday to Wednesday in dissection rooms (ground level). Posters with even number will be presented on Monday, September 5 and posters with odd number on Tuesday, September 6. Pushpins will be available. Authors are asked to be present at posters during poster presentation.

Industrial Exhibition

The exhibitions are located at the ground level and open throughout the meeting.

Exhibitors:

BioTech a.s.
Galén, s.r.o.
Grada Publishing, a.s.
Mega Books CZ, s.r.o.
Nikon, s.r.o.
Olympus Czech Group, s.r.o.
Scintila, s.r.o.
Schoeller Instruments, s.r.o.
Sigma-Aldrich, s.r.o.

Welcome Reception
will take place immediately after the Opening ceremony on Sunday, September 4 in the rooms of Institute of Anatomy. It will include buffet and drinks and is free of charge.

Concert in St. Catherine Church
Concert will take in St. Catherine Church, entry in the corner of Kateřinská and Vinična st. (walking distance from Institute of Anatomy - cca 150 m) on Monday, September 5, from 19:30.

Social Evening
Congress dinner will take place at Novoměstská radnice, (The New Town Hall), Karlovo nám. 1/23, 120 00 Praha 2 on Tuesday, September 6 from 19:00 until 22:00 (price 450,- CZK).

Coffee breaks
Refreshments will be served in the rooms of the Institute of Anatomy, free of charge.

Lunch
Lunches will be served in the rooms of the Institute of Anatomy, free of charge.

Transportation
See city maps in this booklet, pp. 170, 171.

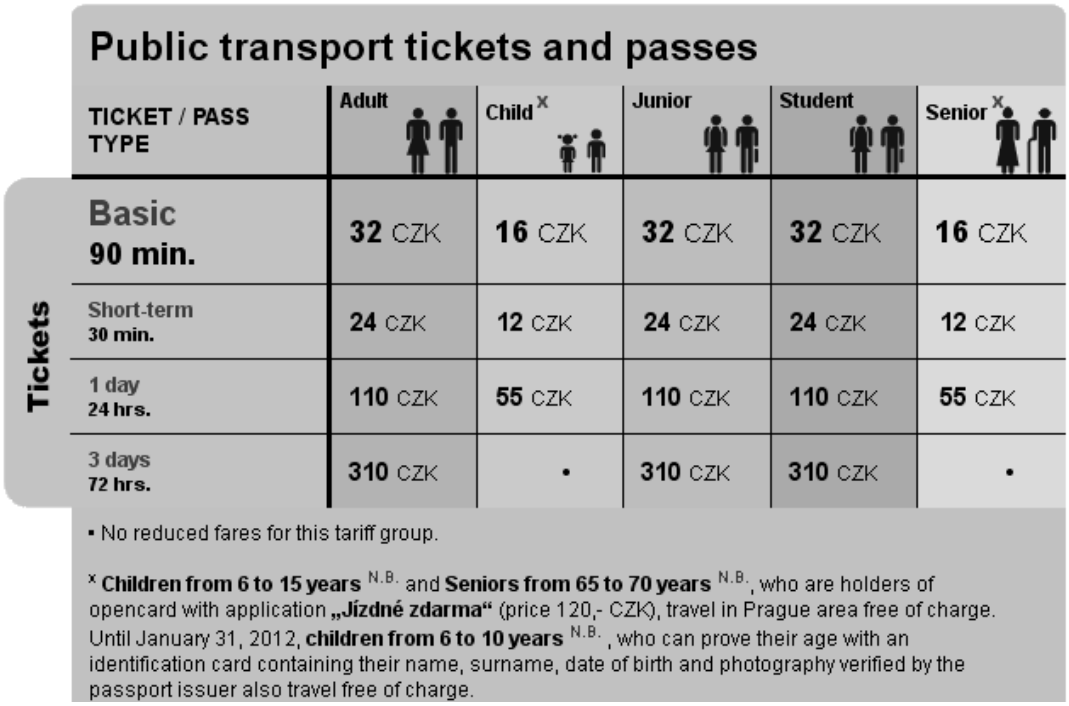
How to reach the Institute of Anatomy, U Nemocnice 3, Praha 2
By tram No. 4, 6, 10, 16, 22, 23, station Štěpánská; by tram No. 3, 4, 10, 16, 18, 21, 24 station Moráň or Karlovo náměstí
By Metro: line C (red) - station I.P.Pavlova; line B (yellow) - station Karlovo náměstí, exit to Karlovo náměstí.
By car: parking in streets of Prague 2 is paid-parking

The New Town Hall (Novoměstská radnice), Karlovo nám. 1/23, 120 00 Praha 2
Is in walking distance from Institute of Anatomy (cca 400 m)

Czech Anatomical Society – member of the Council of Scientific Societies of the Czech Republic
Secretary office: Institute of Anatomy, Charles University of Prague, First Faculty of Medicine, U Nemocnice 3, CZ 128 00 Prague 2
phone: 00 420 224 965 780, fax. 00 420 224 965 770, e-mail: anat@lf1.cuni.cz
http://cas.lf1.cuni.cz

Czech Society for Histo- and Cytochemistry – member of the Council of Scientific Societies of the Czech Republic
Secretary office: Šimkova 870, 500 38 Hradec Králové. phone: +420-495 816 294, fax: +420-495 816 376, e-mail: mokry@lfhk.cuni.cz
http://www.cshc.cz

Transit tariffs



Public transport tickets and passes

TICKET / PASS TYPE	Adult	Child [x]	Junior	Student	Senior [x]
Tickets					
Basic 90 min.	**32** CZK	**16** CZK	**32** CZK	**32** CZK	**16** CZK
Short-term 30 min.	**24** CZK	**12** CZK	**24** CZK	**24** CZK	**12** CZK
1 day 24 hrs.	**110** CZK	**55** CZK	**110** CZK	**110** CZK	**55** CZK
3 days 72 hrs.	**310** CZK	•	**310** CZK	**310** CZK	•

• No reduced fares for this tariff group.

[x] **Children from 6 to 15 years** [N.B.] and **Seniors from 65 to 70 years** [N.B.], who are holders of opencard with application **„Jízdné zdarma"** (price 120,- CZK), travel in Prague area free of charge. Until January 31, 2012, **children from 6 to 10 years** [N.B.], who can prove their age with an identification card containing their name, surname, date of birth and photography verified by the passport issuer also travel free of charge.

Transit ticket vending machines

Ticket vending machines are installed in all metro stations and at selected surface transit stops. They are intended for the purchase of individual tickets.

SMS ticket

Single transfer tickets can be purchased via SMS.

Send SMS	**DPT32**	to purchase regular 90 minute ticket for 32 CZK
	DPT24	to purchase reduced 30 minute ticket for 24 CZK
	DPT110	to purchase 24hour ticket for 110 CZK
	DPT310	to purchase 72hour ticket for 310 CZK

to number **902 06**.
You will receive SMS ticket within approximately 2 minutes.

Advance ticket sales

Sales locations in the metro offer the entire range of tickets, and are intended primarily for the sale of transit passes, both for fixed and sliding validity periods.

Info centres

Information centres sell individual tickets and short-term (tourist) passes.

Tobacconists and wholesalers

Selected tobacconists and wholesalers sell individual tickets.

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Programme

MORPHOLOGY 2011

46rd International Congress on Anatomy
48rd Lojda Symposium on Histochemistry

September 4 – 7, 2011
Prague, Czech Republic

Sunday, September 4, 2011

17:00 - 19:00
Opening Ceremony
Chairs: *Miloš Grim, Eva Mechírová, Jaroslav Mokrý*

Words of Welcome

Prof. MUDr. Jan Škrha, DrSc., MBA
Prorector of The Charles University in Prague

Prof. MUDr. Tomáš Zima, DrSc., MBA
Dean of The Charles University First Faculty of Medicine

Doc. MUDr. Bohuslav Svoboda, CSc.
Lord Mayor of The City of Prague

Ing. Jiří Paluska
Mayor of The of Prague 2

Lecture to the 70th Anniversary of Immunohistochemistry
Denis Corbeil
"New Insights into Cell Biology of Stem Cells by Studying Prominin-1 (CD 133)"

Winning lecture of the 2011 Czech Anatomical Society & Olympus Award
Eliška Krejčí
"Isolation and characterization of neural crest stem cells from adult human hair follicles"

Winning lecture of the 2011 Czech Society for Histochemistry and Cytochemistry & Nikon Award
Pavol Szabo
"Mouse 3T3 fibroblasts under the influence of fibroblasts isolated from stroma of human basal cell carcinoma acquire properties of multipotent stem cells"

19:00 – 20:30
Welcome drink

20:00
Meeting of the National Committees of CAS and CSHC

Monday, September 5, 2011

8:30 - 17:30
Plenary Lectures, Sessions in Section A and B, Poster Session 1

8:30 - 10:10
Plenary Lectures: Stem cells in development and cancer (Lecture Hall A)
Chairs: Aleš Hampl, Birgit Lane

8:30 - 8:55
1 - Lane EB
Failure of the TGF-β pathway leads to multiple self-healing squamous carcinomas

8:55 - 9:20
2 - Smetana K Jr, Dvořánková B, Lacina L, Plzák J, Szabo P, Chovanec M, Strnad H, Kolář M
Epithelial-mesenchymal interaction in cancer

9:20 – 9:45
3 - Hampl A
Interiors and exteriors of human embryonic stem cells

9:45 – 10:10
4 - Peterková R.
Rudimentary structures and their importance during dentition development

10:10 - 10:30 Coffee Break

10:30 - 12:00
Stem Cells (Lecture Hall A)
Chairs: Jaroslav Mokrý, Karel Smetana Jr.

10:30 - 10:45
5 - Lacina L, Kodet O, Smetana K Jr, Dvořánková B, Plzák J, Szabo P, Chovanec M, Strnad H, Kolář M
Relevancy of Skin Cancer in vitro Modelling. (Lesson from Complexity of Epithelial Mesenchymal Interactions in Development and Cutaneous Pathology)

10:45 - 11:00
6 - Dvořánková B, Szabo P, Lacina L, Smetana K Jr., Gabius HJ
Galectins as inducers of production of a 3D extracellular matrix

11:00 - 11:15
7 - Kodet O, Krejčí E, Lacina L, Dvořánková B, Smetana K Jr, Grim M
Intercellular interactions in malignant melanoma

11:15 – 11:30
8 - Mokrý J, Filip S, Čížková D, Vávrová J, Šinkorová Z, Mičuda S, Brčáková E, Fuksa L
Tissue chimaerism following bone marrow cell transplantation

11:30 - 11:45
9 - Soukup T, Barry F, Murphy M, Coleman C, Cancedda R, Gentili Ch, McGongale D, Jones E, Rowan T, Thornton J, Elliman S, Clissmann C
Revolutionising the large-scale production of high quality adult stem cells – PurStem project

11:45 - 12:00
10 - Ecker RC, Rogojanu R, Steiner G
Applications of Slide-Based Single Cell Cytometry by TissueFAXS in Histopathology

10:30 - 12:00
Experimental Medicine (Lecture Hall B)
Chairs: Marian Adamkov, Jitka Kočová

10:30 - 10:45
11 - Adamkov M, Výbohová D
Expression of survivin in benign, pre-malignant and malignant tissues

10:45 - 11:00
12 - Überall I, Škarda J, Janíková M, Radová L, Fridman E
Prognostic significance of LC3 autophagic protein in non-small cell lung cancer

11:00 - 11:15
13 - Novotný T, Krejčí J, Švehlík V, Uhlík J, Vajner L
Effective stabilisation of mast cell granules by sodium cromoglycate leads to changes in percentage of double-laminated vessels during hypoxia and posthypoxic recovery

11:15 - 11:30
14 - Tóth Š, Jonecová Z, Varga J, Staško P, Kovalčinová B, Veselá J
The changes in jejunal mucosa cell populations during mesenteric ischemia-reperfusion injury in rats

11:30 - 11:45
15 - Křížková V, Tonar Z, Houdek K, Eberlová L, Moláček J, Boudová L, Nedorost L, Tolinger P, Kočová J, Korabečná M, Třeška V, Vrzalová J, Pešta M, Topolčan O
Morphology of an Experimental Abdominal Aortic Aneurysm Model

11:45 - 12:00
16 - Eberlová L, Tonar Z, Witter K, Křížková V, Korabečná M, Kočová J, Boudová L, Nedorost L, Třeška V, Houdek K, Moláček J, Topolčan O, Vrzalová J, Pešta M, Valenta J
Quantitative Histology and Proteomic Analysis in Abdominal Aortic Aneurysms

12:00
Congress Photo

12:15 - 13:30 Lunch

13:30 - 14:35
Plenary lectures: Neurosciences (Lecture hall A)
Chairs: Petr Dubový, Darina Kluchová

13:30 - 13:55
17 – Druga R, Salaj M, Cerman J, Kubová H
Status epilepticus results in extensive brain damage in immature rats

13:55 - 14:20
18 – Němcová V, Petrovický P
Correlation of the anatomical structure of the amygdala with its MRI image

14:20 – 14:35
19 - Kučera T, Martínek J
The use of digital virtual microscopy system for creating digital histology slides database as a tool for e-learning

14:35 - 15:00 Coffee break

15:00 - 16:15
Neurosciences (Lecture Hall A)
Chairs: Rastislav Druga, Eva Mechírová

15:00 - 15:15
20 - Birkhead TR, Halata Z
Sensory innervation of the phaloid organ in African Buffalo-Weaver (light- and electronmicroscopic study)

15:15 - 15:30
21 - Dubový P, Svíženská I, Klusáková I, Brázda V, Strejčková L
Neuroinflammatory reaction of peripheral glial cells in two different models of neuropathic pain

15:30 - 15:45
22 - Balentová S, Hajtmanová E, Kinclová I, Trylcová R, Lehotský J, Dobrota D, Adamkov M
Radiation induced long-term changes in forebrain's neurogenesis under experimental conditions

15:45 - 16:00
23 - Mazurová Y, Gunčová I, Astapenko D
Is the neurodegenerative process in the striatum of rats transgenic for Huntington's disease similar to that in HD patients?

16:00 - 16:15
24 - Brichová H, Zima T
Radial glia differentiation and tissue vascularization in rat prosencephalon and telencephalon under normal and experimental conditions (hypoxia, FAS)

15:00 - 16:00
Teaching of Morphology (Lecture hall B)
Chairs: Petr Hach, Yvetta Mellová

15:00 - 15:15
25 - Buchtová M, Horáková D, Matalová E
Experimental embryology - course innovations

15:15 - 15:30
26 - Hubičková-Heringová L, Maňáková E
Chick embryo as a model for normal and abnormal organogenesis - The elective course in the Medical Curriculum at the 3rd Faculty of Medicine

15:30 - 15:45
27 - Hájek P, Slížová D
The dissecting essay - our experience in a seminary work for students of the 1st year

15:45 - 16:00
28 - Lovásová K, Kluchová D, Boleková A
The possibilities of improving the teaching process of Anatomy in study program of Dental Medicine

16:15 - 17:30 Poster session 1 (dissection rooms) + refreshment
Authors of posters with *even numbers* should be present

19:30 Concert in St. Catherine Church

Tuesday, September 6, 2011

8:30 - 17:15 Plenary Lectures, Sessions in Section A and B, Poster Session 2

8:30 - 9:50
Plenary Lectures: Molecular Embryology of Cardiovascular System (Lecture Hall A)
Chairs: Miloš Grim, Robert Gourdie

8:30 - 8:50
29 - Gourdie RG, Ongstad EL, O'Quinn MP, Rhett JM, Palatinus JA, Bowers SLK, Baudino TA, Borg TK
The Connexin43 Carboxyl-Terminus: Regulator of Gap Junction Ultrastructure and Tissue Morphology at the Injury Border Zone Following Myocardial Infarction

8:50 - 9:10
30 - Sedmera D
Ontogenesis and phylogenesis of cardiac conduction system

9:10 - 9:30
31 - Kučera T
Vascular morphogenesis in vertebrates and invertebrates – models of vascular lumen formation

9:30 – 9:50
32 – Naňka O
Physiological role of tissue hypoxia in embryonic development

9:50 - 10:15 Coffee break

10:15 - 11:15
Cardiac Morphogenesis (Lecture Hall A)
Chairs: Jan E. Jirásek, David Sedmera

10:15 - 10:30
33 - Jirásek JE
Prenatal Developmental Stages of the Human Heart

10:30 - 10:45
34 - Šaňková B, Sedmera D
Normal development of cardiac conduction system in the mouse

10:45 - 11:00
35 - Beneš J Jr, Šaňková B, Sedmera D
Effect of connexin40 deficiency on atrial activation in mice on ED12.5

11:00 - 11:15
36 - Peševski Ž, Sedmera D
Endocardial fibrolastosis in chick model of embryonic pressure overload

10:15 - 11:15
Varia (Lecture Hall B)
Chairs: Jiří Ehrmann, Štefan Polák

10:15 - 10:30
37 - Fiala P, Khadang I, Kaiser J, Štepanek I
Mechanical properties of lamellar systems of the human osteon

10:30 - 10:45
38 - Jirkovská M, Niedobová V, Jadrníček M, Moravcová M, Krejčí V, Žižka Z
Comparison of proliferative potential of normal and diabetic term placenta

10:45 - 11:00
39 - Hodorová I, Rybárová S, Mihalik J
Distribution of monoamine oxidase A in normal and tumor kidney tissue

11:00 - 11:15
40 - Rybárová S, Hodorová I, Vecanová J, Muri J
Analysis of expression of drug resistance proteins in non-small cell lung cancer in relation to prognosis

11:15 - 12:15
Poster session 2 - dissection rooms
Authors of posters with *odd numbers* should be present

12:15 – 13:45
Lunch

13:45 - 15:25
Plenary lectures: Developmental Biology (Lecture Hall A)
Chairs: Radomír Čihák, Miroslav Peterka

13:45 - 14:10
41 - Černý R
Vertebrate jaw evolution (Prepattern/Cooption model): genetic, epigenetic or mechanic cause?

14:10 - 14:35
42 - Peterka M
Orofacial development and cleft anomalies

14:35 - 15:00
43 – Šnajdr P, Liška F, Krejčí E, Křenová D, Křen V, Grim M.
Why five fingers ? Genetic control of limb development.

15:00 - 15:25
44 – Valášek P
Development of the limb girdles

15:25 - 15:50 Coffee Break

15:50 - 16:50
Developmental Biology (Lecture Hall A)
Chairs: Milena Králíčková, Ondřej Naňka

15:50 - 16:05
45 - Kralovic M, Horáček I & Černý R
Evolutionary middle ear precursor with the presence of teeth

16:05 - 16:20
46 - Buchtová M, Tucker AS, Matalová E
Degradation and morphology of successional dental lamina

16:20 - 16:35
47 - Zemanová Z, Jirsová Z, Hubičková-Heringová L, Kubínová L
Mitotic activity in the 5-day chick embryonic kidney

16:35 - 16:50
48 - Králíčková M, Čedíková M, Pitule P, Liška V, Zech N, Uher P
Key molecular regulatory factors of developmental potential of human embryo - clinical implications

15:50 - 16:50
Experimental Morphology (Lecture Hall B)
Chairs: Luděk Vajner, Tomáš Soukup (Prague)

15:50 - 16:05
49 - Klepáček I, Pleschnerová M
Spatial changes of vascular patterns in limb buds transplanted on CAM

16:05 - 16:20
50 - Krejčířová L
Morphological manifestations of heavy metals toxicity

16:20 - 16:35
51 - Maňáková E, Hubičková L, Zemanová Z
Mirtazapine embryotoxicity evaluation by Chick Embryotoxicity Screening Test

16:35 - 16:50
52 - Petrásek T, Kopecká K, Zacharova G, Paleček J, Tribulová N, Soukup T
Effect of thyroid status alteration and red palm oil supplementation on motor activity and thermal sensitivity of adult rats

16:50 - 17:15
Plenary meeting of Czech Society for Histo- and Cytochemistry (Lecture Hall A)
Plenary Meeting of Slovak Society for Histo- and Cytochemistry(Lecture Hall B)

19:00 – 22:00
Dinner – Novoměstská radnice, Karlovo nám.

Wednesday, September 7, 2011

8:30 - 12:30
Plenary Meeting of CAS, Plenary Lectures, Sessions in Section A and B

8:30 - 9:00
Plenary Meeting of Czech Anatomical Society (Lecture Hall A)
Plenary Meeting of Slovak Anatomical Society (Lecture Hall B)

9:15 – 10:20
Plenary Lectures: Clinical Anatomy
Chairs: Josef Stingl, Oldřich Eliška

9:15 – 9:40
53 - Eliška O
Clinical anatomy of lymphatic drainage of the breast and parasternal region

9:40 – 10:05
54 - Báča V, Grill R, Otčenášek M, Kachlík D, Báčová T, Bartoška R, Džupa V
Morphological context of pelvic fractures

10:05 – 10:20
55 - Stingl J, Musil V
The synovial bursae in the past and today

10:20 - 10:45 Coffee Break

10:45 - 11:45
Clinical Anatomy (Lecture Hall A)
Chairs: Pavel Fiala, Dáša Slížová

10:45 – 11:00
56 - Kočová J, Tonar Z, Křížková V, Boudová L, Hes O, Opatrná S, Třeška V, Korabečná M, Horčička V, Nedorost L
Structure and changes of the peritoneal membrane at the beginning and during peritoneal dialysis

11:00 – 11:15
57 - Kuchař M
Prevalence and variability of the inferior right hepatic vein

11:15 – 11:30
58 - Tonar Z, Kural T Jr, Kochová P, Nedorost L, Witter K
Quantification of vasa vasorum in human varicose great and small saphenous vein

11:30 - 11:45
59 - Kochová P, Cimrman R, Witter K, Tonar Z
Density and orientation of brain microvessels – a quantitative approach

10:45 – 11:30
History of Anatomy (Lecture Hall B)
Chairs: David Kachlík, Olga Procházková

10:45 – 11:00
60 - Procházková O
Today's state of Hyrtl´s skull collection

11:00 – 11:15
61 - Kachlík D, Čech P
Hyrtl in eponyms

11:15 – 11:30
62 - Čech P
The Prague anatomist Hugo Rex (1861–1936)

12:00 Closing session (Lecture hall A)

Posters

Monday 16:00 - 17:30
Poster session 1 (dissection rooms)
Authors of posters with *even* numbers should be present

Tuesday 11:15 - 12:15
Poster session 2 (dissection rooms)
Authors of posters with *odd* numbers should be present

1. Balková S, Buchtová M
The expression of *Msx2* and *Bmp4* in odontogenesis

2. Bartoš A, Zach P, Tintera J, Ripova D
Comparison of hippocampal and temporal horn areas on brain MRI as indicator of Alzheimer disease

3. Bartoška R, Skála J, Džupa V, Kachlík D, Báča V
Morphological aspects of pertorchanteric and intertrochanteric fractures osteosynthesis

4. Bezdičková M, Filipčíková R, Machálek L, Blažková Z, Laichman S
Vena cava superior sinistra – a case report

5. Bilecová - Rabajdová M, Urban P, Gregová K, Mareková M, Veselá J
Influence of ischemia and transplantation of small intestine to expression of ER chaperones in lungs

6. Bolekova A, Dorko F, Kolesar D, Lovasova K, Kluchova D
New pathways in teaching and learning human anatomy

7. Caisberger F, Skálová M, Pejchal J, Kassa J
A Comparison of Single Oxime versus Oxime Mixture Treatment in Cyclosarin-Poisoned Rats

8. Cedikova M, Pitule P, Houdek Z, Cendelin J, Kulda V, Kralickova M, Vozeh F, Babuska V, Pachernik J, Zech N, Uher P
The comparison of the survival of P19-derived neuroprogenitors and P19 naive cells after intracerebellar application in a mice model with and without neurodegeneration

9. Danielisová V, Burda J, Némethová M, Gottlieb M
Effect of Bradykinin after middle cerebral artery occlusion in rat

10. Domoráková I, Mechírová E, Stebnický M, Žofčák M, Danielisová V, Burda J
Influence of pharmacologic preconditioning and ischemia /reperfusion on rabbit spinal cord neurons

11. Dorko F, Výborná E, Boleková A
Comparison of the Knowledge of Anatomy Students at the Faculties of Medicine in Ostrava and Košice

12. Eberlová L, Křížková V, Tonar Z, Kočová J, Třeška V, Houdek K, Moláček J, Valenta J
Quantitative Assessment of Elastin in the Abdominal Aortic Aneurysmal Wall

13. Fík Z, Valach J, Kodet O, Chovanec M, Plzák J, Gabius H-J, Smetana K Jr
Loss of Galectin-9 from head and neck squamous cell epithelium is an emerging indicator of malignant transformation

14. Filipčíková R, Bezdičková M, Hubáček P, Cigánik D, Veselý B, Blažková Z, Laichman S, Štěpánková M
Partnership network for Clinical Anatomy and Emergency Medicine education – VTEC

15. Gorošová A, Madej JP, Řehák Z, Tichý F, Buchtová M
Skin microscopic structure of the Common Pipistrelle (Pipistrellus pipistrellus)

16. Gregová K, Bilecová - Rabajdová M, Urban P, Mareková M, Veselá J
Correlation of the expression of apoptic genes between donor and recipient in small intestine transplantation model

17. Hájek P
E-learning course Topography of head and neck

18. Halaj M, Kubát M, Joukal M, Strejčková L, Svíženská I, Klusáková I, Dubový P
Invasion of macrophages distal to neonatal and adult nerve crush and its relation to neuropathic pain induction.

19. Haque A, Khan MY, Minhas LA
Effect of prenatal administration of folic acid on retinoic acid induced developmental anomalies in chick thymus

20. Hešková G, Mellová Y, Holomáňová A, Výbohová D
Variations of the pneumatization of the ethmoid cells

21. Horáková D, Buchtová M
The influence of FGF inhibitors on embryonic development

22. Hradilová-Svíženská I, Klusáková I, Brázda V, Dubový P
CB2 receptor protein and mRNA expression in dorsal root ganglia of two rat neuropathic pain models

23. Humlová D, Novotný T, Krejčí J, Švehlík V, Uhlík J, Vajner L
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Abstracts

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Expression of survivin in benign, pre-malignant and malignant tissues

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Survivin is a multifunctional protein that inhibits apoptosis, regulates cell division and enhances angiogenesis. Due to large differences in the degree of survivin expression in cancers and in corresponding normal adult tissues, survivin seems to be a promising diagnostic, prognostic and predictive biomarker.

We studied expression pattern of survivin in 121 cases of epithelial breast lesions and 70 cases of melanocytic lesions by immunohistochemistry using anti-survivin antibody (DAKO). We assessed the percentage of positively stained cells, the intensity of staining and its subcellular localization.

Survivin was detected in 10/19 cases of normal breast tissue - 52.6% (cytoplasmic positivity only), in 28/38 cases of fibroadenomas (19 cases cytoplasmic reaction - 50%, 9 cases combined cytoplasmic and nuclear positivity in small foci of cells - 23.7%), and in 55/64 cases of carcinoma - 86% (12 cases cytoplasmic expression - 18.8%, 8 cases nuclear positivity - 12.5%, 35 cases of combined reaction - 54.7%). In melanocytic lesions, survivin was expressed in 11/18 cases of benign nevi - 61% (cytoplasmic reaction only), in 21/27 cases of dysplastic nevi - 77.8% (cytoplasmic positivity 17 cases – 81%, combined reaction 4 cases - 19%, severe dysplastic changes), in 23/25 cases of malignant melanoma - 92% (nuclear staining 2 cases - 8.7%, cytoplasmic staining 3 cases - 13%, combined staining 18 cases - 78.3%).

Summarizing our results, we detected a gradually increased expression of survivin, starting from normal adult tissues and benign entities through pre-malignant lesions to its overexpression in great majority of malignant tumors. Survivin seems to be valuable diagnostic, differential-diagnostic, prognostic and predictive marker.

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Morphological context of pelvic fractures

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Injury to the pelvic ring is one of the most serious injuries of the skeleton. Increase in the number of pelvic fractures in recent years (200% increase according to the literature), better pre-hospital care and interdisciplinary cooperation in polytraumatized patient care has led to a point where large number of pelvic fractures and serious ones are being treated in comparison with previous years. Significant factor affecting the decision about therapeutic process, timing and subsequent steps, but also the prognosis of the patient with pelvic fracture, are concurrent injuries. Concurrent injuries are defined as injury to the organs and anatomical structures present in the pelvic region. This does not include injury to the distant organs or structures outside the pelvis injured within the polytrauma or combined trauma. Concurrent injuries are present mainly with type B and C injuries and relate in particular to neurogenic structures (most frequently n. ischiadicus, plexus lumbosacralis, n. femoralis, and n. obturatorius) and urogenital tract (most frequently urethra, urinary bladder, vagina, penis, scrotum) farther to gastrointestinal tract (anus, rectum, colon and ileum) and less to gynecologic organs (usually the uterus). Bibliography states occurrence of injury to the neurgenic structures 9-21%, urogenital injuries 5-11%, gastrointestinal 3-17% and gynecologic organs 1%. Severe pelvic injury is linked in almost 20% with urogenital tract injury, but 90% of all traumatic lesions to the urinary bladder and urethra accompany sever pelvic injury.
Urinary incontinence, dysuria or sexual problems (dyspareunia - painful or difficult sexual intercourse) and incontinence of stool can be the result of direct injury to the urogenital tract or terminal parts of the gastrointestinal system, but also as a sequel to neural or vascular structure injury. Moreover, it is showing that very important part in urologic or sexual consequences plays lesion to the pelvic fascia and lesion to m. levator ani in continence failures.
The knowledge of clinical and particularly topographical anatomy of the pelvis can significantly influence physician decision-making early in the therapeutic process.

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Radiation induced long-term changes in forebrain's neurogenesis under experimental conditions

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We investigated the occurence of radiation-induced delayed alterations of proliferation activity in the rat brain neurogenic region. Adult male Wistar rats were investigated 30, 60 or 90 days after whole-body irradiation with fractionated doses of gamma rays (the total dose of 4 Gy). For the study of alterations of the numbers of proliferating cells through the neurogenic pathway, called the rostral migratory stream (RMS), Ki-67, a marker for cell proliferation was used. Quantitative anylysis of cell proliferation in different areas along the migratory pathway, i.e. in the vertical arm, the elbow and the horizontal arm showed time-dependent changes between control and experimental groups. Proliferative activity in rats, evaluated 30 days after exposure remained unchanged in all selected areas of the RMS. In the following two months there was strong decrease in the number of Ki-67-positive cells mostly in caudal parts of the RMS. However, the data from quantitative analysis of the numbers of proliferating cells are still under evaluation, our preliminary results showed, that ionizing radiation clearly affect this neurogenic region.

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The expression of *Msx2* and *Bmp4* in odontogenesis

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Teeth develop through reciprocal interactions between the neural crest-derived mesenchyme and the ectoderm of the oral cavity. Mammalian heterodont dentition is formed of a variety of tooth types with different shapes and cusp numbers. Several hypotheses were proposed to explain the origin of different tooth classes in mammals including the concept of morphogenetic fields, the clone theory and the odontogenic homeobox code (Butler, 1939; Osborn, 1978; Sharpe, 1995) but no one was tested on normodont dentition. We chose to study the development of deciduous teeth in pig embryos as they resemble human teeth in function and shape.

In mouse, *Bmp4* was found to be expressed in rostral rostral area of mandible that induces the expression of transcription factors *Msx2* in the mesenchyme. We designed primers for pig *Bmp4* and *Msx2* based on sequence from public database (www.ensemble.org). Total RNA was isolated by using the RNeasy Mini Kit (Qiagen) from pig embryos (ED 27). SuperScript VILO (Invitrogen) was used for reverse transcription. Gene specific fragments were amplified by PCR and approximately 600bp long PCR products were inserted into the TOPOII vector (Invitrogen). Plasmids were isolated by Plasmid DNA Purification Kit (Qiagen) and the inserts were confirmed by the sequencing (Elisbeth Pharmacon). Later, Dig labeled probe was synthetized and whole mount in situ hybridization was performed on pig embryos (ED 23).

The expression of pig *Bmp4* was localized to the caudal part of limb buds and spinal cord. *Msx2* was expressed in the mesenchyme of the cranial and caudal area of limb buds and also in the spinal cord. Strong signal was localized in the frontonasal mass and cranial part of mandibular arch. The expression was weaker laterally in the face but it is detectable also in the maxillary prominence where simple shaped canine and the third incisors are initiated. In conclusion, the expression pattern in pig limbs and face resembled the expression in mouse embryos.

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Comparison of hippocampal and temporal horn areas on brain MRI as indicator of Alzheimer disease

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AIMS: Mediotemporal atrophy is an early and most prominent brain change in Alzheimer disease (AD). The hippocampus (hipp) contrasts with the surrounding cerebrospinal fluid space in the temporal horn of ventricles (horn) on brain MRI. An easy quantification of these structures would be usefuf in the diagnosis of AD. To quantify hippocampal and horn areas and their relationships on MRI in AD and controls.
PATIENTS AND METHODS: *1.ADgroup***:** 26 patients according to NINCDS-ADRDA criteria**;** *2. Control group:* 29 normal seniors without cognitive deficits
Variables measured: We manually traced and on one coronal T1-weighted image appropriate for each side. We also manually traced one at commisura anterior for normalization to the head / brain size. Variables calculated: We calculated a percentage of the hippocampal area related to the combined area of the hippocampus and the surrounding horns. Besides absolute measures we summed the hippocampal, horn and combined areas for both sides and normalized than to the of a particular subject.
RESULTS: Compared to controls, theAD patients had significantly smaller hippocampal areas and larger horn areas on either side. The hippocampal area occupies significantly smaller proportions of the combined hippocampal+horn areas in the AD patients (dx 63 %, sin 63 %) than those in the controls (dx 84%, sin 83 %). The same results apply for similar measures normalized to the brain area at commisura anterior.

Morphological aspects of pertorchanteric and intertrochanteric fractures osteosynthesis

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From the muscle-ligamentous complex system point of view the morphology of the location of the osteosynthesis introduction for pertrochanteric and intertrochanteric fractures is not elementary, many times, but still unsatisfactorily described. Pertrochanteric fractures belong to the most frequent at all, caused by fall in elderly, by traffic accidents in younger. For the stable treatment is essential the right choice of implant (nail, locking plate, the number of locking screws) and its correct positioning with regard to the arrangement of anatomical structures which might be affected by the surgeon (soft tissue, the structure of cortical and cancellous bone). The aim of the study was to describe morphological aspects of pertochanteric fracture osteosynthesis (PFH, IMHS) with stabilizing of typical fragments.

On 20 anatomical specimens of the proximal femur the region of trochanteric massif with a detailed description of the muscle-ligamentous structures of OS insertion sites and of fracture lines were dissected. Course of osteons in this area was evaluated.

There were identified anatomical structures that may be threatened by the introduction of the osteosynthesis as bone damage and soft tissue damage during surgery. There were described places where no soft tissue covers the bone and where typical fracture fragments arise.

Stable osteosynthesis of pertrochanteric and intertrochanteric fractures must reflect morphological and biomechanical aspects of this region.

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Effect of connexin40 deficiency on atrial activation in mice on ED12.5

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Intercellular coupling via gap junctions is an important determinant of impulse propagation in the heart. Cx40 is the main connexin expressed in atria and ventricular conduction system. The main pacemaker of the heart, sinoatrial node (SAN), is usually located in the right atria near superior vena cava. From this point the activation is spreading through the atria. The objective of our study was to assess the effect of Cx40 absence on atrial activation in murine fetal heart at ED12.5 using optical mapping. According to our data, Cx40 deficiency decelerates the spreading of activation in the atria, comparing the activation from SAN area in WT and in Cx40-/-. In Cx40 deficient mice we also found majority of activation patterns taken place from ectopic focus. And the spreading from this ectopic place is even more decelerated.
Key words: connexin40, cardiac conduction system, sinoatrial node

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Vena cava superior sinistra – a case report

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A persistent left superior vena cava (LSVC) is rather rare anomaly. LSVC was found within the dissection course in the cadaver of 67 years old men who has died from acute peritonitis. The diameter of superior vena cava was 6mm and it originated at the lateral part of the left brachiocephalic vein and passed downward anterior to the left pulmonary veins. Finally it opened to the coronary sinus.

LSVC is an abnormality caused by persistence of the left anterior cardinal vein from the foetal circulation. In case of LSVC persistence, obliteration of the common cardinal and proximal part of the right anterior cardinal vein accompanied this anomaly. In such a case, blood from the right is channelled toward the left by way of the brachicephalic vein. The LSVC drains into the right atrium by way of the coronary sinus.

A double superior vena cava is characterized by the persistence of the left anterior cardinal vein and failure of the left brachiocephalic vein to form. The persistent anterior cardinal vein – LSVC – drains into the right atrium by way of the coronary sinus.

In our case the superior vena cava was doubled and the left brachiocephalic vein was formed too.

Persistent LSVC is known congenital anomaly of the venous field that has already been described in the Atlas of Anatomy by Told. Recently, case reports describe such anomaly as a revelation during the dissection, within the computer imaging methods (MRI or CT imaging) or within the echocardiogram's check-ups. Several cases have been revealed within the heart surgery as the treatment of the congenital disease too. Incidence of LSVC is about 0, 3% in persons without any other proved heart pathology and LSVC is mainly asymptomatic. In the patients with congenital heart disease the incidence is slightly higher – 4, 5%.

In relation to the age of the cadaver where we have described the LSVC and because no heart disease was located, in our case LSVC was evidently asymptomatic.

Influence of ischemia and transplantation of small intestine to expression of ER chaperones in lungs

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The endoplasmic reticulum (ER) is significantly involved after intervention and post-ischemic reperfusion through the programmed cell death. During apoptosis are activated processes which at the end activate Gadd153 expression and caspases induction what cause the cell death or starts the survival mechanisms through production of protein Grp78.

The purpose of this study was to observe the apoptic changes in the lungs, initiated through inflammatory response from the small intestine after it`s one hour ischemic insult with subsequent reperfusion in time periods 1h., 24 h. and 30 days (R1 to R30 group). We were monitoring levels of mRNA expression of pro-apoptotic Gadd153 (Chop) and anti-apoptotic genes Grp78 (Bip) in lung parenchyma. In next step we detected expression changes within the group with the small intestine transplantation and 1 or 6 hours survival period in comparison to control sham operated animals.

In the R1 group we found significantly increased levels of mRNA for gene Gadd153 (21 ± 4 % higher than controls). In opposite to this we detected the lowest mRNA level after 30 days long reperfusion (41,8 ± 10,4 % lower than controls). The levels of anti-apoptic gene Grp78 was lower or equal than controls in all time periods. In the group after intestine transplantation and one hour survival we found significant maximum in Grp78 mRNA level (110 ± 16,2 % higher than sham controls).

For better understanding of the apoptosis alternative pathway it is necessary to analyze the molecular changes after small intestine ischemia–reperfusion, and determine the contribution of this attack to spreading of MODS or to recovery processes in the vital organs.

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Sensory innervation of the phaloid organ in African Buffalo-Weaver. Light- and electronmicroscopic study.

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The Buffalo-weavers */Bubalornis/* spp. are unique among birds in possessing a phalloid organ. This is a penis-like appendage of about 17 mm length in the male, situated at immediately anterior to the cloaca. It contains neither a duct nor a corpus cavernosum. This phalloid organ is employed during copulation but without being introduced into the cloaca of the female. Instead, it is rubbed against the exterior of the female's cloaca. Copulation is unusually protracted in the buffalo weaver and can last up to 30 minutes and may lead to an “orgasm” of the male bird with ejaculation. No other bird is known
to undergo orgasm.

The present study examines the structure and innervation of this phalloid organ using light- and electron-microscopy. It is mainly formed of firm connective tissue covered by skin. The keratinized flat epithelium contains thin feathers. Herbst corpuscles consisting of an inner core and terminal axon are found in the dermis near the bulb of the feathers. They are supplied by myelinated axons of about 8 µm diameter. Bundles of such myelinated nerve fibres are found throughout the entire organ. Their total number is in excess of several hundreds, probably each terminating in a sensory corpuscle. This large number of nerve fibres suggests that the phalloid organ is a highly sensitive structure containing a large number of Herbst corpucles functioning as rapidly adapting mechanoreceptors.

New pathways in teaching and learning human anatomy

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Following new trends in methods of teaching, it is necessary to pay more attention to quality, depth and effectiveness of medical education. In the last years, the particular attention has been paid to the process of teaching and education of Anatomy. Our department has been also focusing on innovative teaching processes with substantial attention on students of dental medicine.

Two anatomical publications: “Guide through Anatomy of Human Body” and “Anatomy of Trunk and Limbs for Dental Students” written by teachers of Anatomy department help students to prepare for anatomy in a better way. Lectures are improved by implementation of 3D virtual models and animations prepared according to our requirements. 3D virtual models serve students to imagine the anatomical structures like in the real body and to achieve as high precision as they need. Visual perception equipped with the comments of teachers brings a big didactic benefit. The virtual materials can be easily transformed into the 2D pictures so these outputs are used in computerized classroom for practical self-study and knowledge assessment as well.

Seeing that the establishment of base of knowledge and its sufficiency for further study and practice of dentist is a very difficult question, our department are endeavour after appropriate education of students of dental medicine for many years. Convenient solution was to structuralize broad themes into the small most important educational parts which can be used in self-study as well as in dissecting room. Both slender books and 3D virtual models were met unhoped-for interest of students.

The new books and lectures facilitate students to study of human body, directing them correctly to draw attention to anatomical structures which are important for their future educational activities.

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Radial glia differentiation and tissue vascularization in rat prosencephalon and telencephalon under normal and experimental conditions (hypoxia, FAS)

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Differentiation of radial glia (RG) and tissue vascularization in prosencephalon and telencephalon has been studied in 3 groups of Wistar rat embryos and fetuses: a) under chronic hypobaric altitude hypoxia treatment and b) under chronic ethanol abuse treatment of pregnant dams c) controls. Tissue samples were studied using biochemical, histological, EM and immunocyto-chemical methods (vimentin, Ox_2, NF01, β-III-tubulin and lectins). In early development RG plays a role of a pool for neural and glial progenitors and of a scaffolding for neuroblast migration, although their precise contribution to neurogenesis remains controversial. At E12 the basal 1/4 of ventricular zone was not yet vascularized. In this layer, in both experimental groups, RG cells and their progeny were damaged due to extracellular oedema derived in avascular part of ventricular zone. The drainage of tissue fluid was decreased and oedema conditioned changes in the metabolism of tissue: a) decrease of O_2 supply and the direct influence of high concentration of lactate and ions and lack of glucose in the tissue fluid, b) tissue hypoxia and primary direct toxic effect of ethanol and its metabolites on the cells. At E16-18 in both a) and b) structural changes in the developing basis of cortical plate were seen. Whilst in the hypoxic tissue retarded cell and blood vessel differentiation mostly conditioned alterations in the corticogenesis, with (perhaps) the prospective possibility of development; in the alcohol treated tissue the basic structure of cortical plate was defective, high reduction of extracellular spaces hindered transport of O_2, growth factors and signaling molecules, vessel sprout growth, neuroblast migration and synaptic contact formation.

Degradation and morphology of successional dental lamina

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There are significant variations in the dental lamina morphology and development in monophyodont, diphyodont and polyphyodont species. However, detailed accounts of the processes, which establish this species-specific pattern, and govern the number of tooth generations, are unknown. The mouse is the main model for tooth developmental study; however, it forms only one tooth generation. The pig lamina is therefore much more amenable for such study, and more similar to that observed in humans. The degradation of the pig lamina, like that of the human, occurs during mid-gestation, creating epithelial pearls during disintegration with potential to cysts formation. Significant morphological differences in the structure of the two sides of the dental lamina become obvious with increasing age of embryos. The disintegration process starts in the side facing the tooth anlagen. We suggest that there are three possible processes that can be involved in dental lamina regression: apoptosis, migration and epithelial-mesenchymal transformation. As the number of cells that make up the lamina appeared to reduce during the development, we analyzed the engagement of apoptosis in dental lamina regression during prenatal period. However, TUNEL-positive cells were rarely located in the lamina of minipig. DiI labeling method was used in order to track groups of epithelial cells migration. Injection of DiI into tissue slices of stage E55 showed the cells to have high migratory potential. Some of cells were already visible out of the dental lamina and appeared to have spread in the adjacent mesenchyme after two days of incubation. In this study, we also tested the hypothesis of involvement of EMT process during odontogenesis and analyzed the presence of EMT markers such as Slug, Mmp2, E-cadherin, vimentin and c-myb at protein level. These markers were chosen to uncover different steps during epithelial-mesenchymal transformation process such as breakdown of basal lamina, loss of cell-cell attachment, degradation of the extracellular matrix, and initiation of a mesenchymal fate. As the dental lamina disintegrated levels of MMP2, Slug and c-Myb were increased on the side of the lamina facing the tooth anlagen, while expression of E-cadherin decreased, starting in those cells with high MMP2, Slug and c-Myb levels. Our results support the migration and epithelial-mesenchymal transformation of cells as processes involved in the break-up of the dental lamina.

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Experimental embryology - course innovations

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Dynamic and effective bridging of preclinical disciplines with practical experience and clinical applications represents one of major challenges in medically oriented universities. At the University of Veterinary and Pharmaceutical Sciences in Brno, the students pass anatomy, histology, with few lessons from embryology and continue with physiology and pathophysiology subjects. Experimental embryology, which has been offered as a new subject since the last academic year, aims to interconnect these disciplines and supplement the knowledge also at molecular and cellular levels which represent the basic approaches in recent development and biomedicine related research. Moreover, the course aims to emphasize understanding of developmental processes in order to understand basis of developmental, hereditary and acquired disorders. In the end, application of obtained knowledge in own experimental design, and in recent treatment strategies are demonstrated, particularly in the area of molecular medicine and stem cells.
Theoretical lectures and seminars are followed by direct hands-on experience in the laboratory. The students start with embryonic stages and species related specificities, become familiar with *in vivo*, *in ovo*, *ex vivo* and *in vitro* systems including analysis and modifications of the embryonic development at DNA level (e. g. PCR, electroporations), RNA level (e. g. *in situ* hybridisation, gene silencing) and proteins (e. g. insertion of inhibitor/activator soaked beads). The students manage their own practical project based on application of retinoic acid into a developing limb *in ovo* to demonstrate and evaluate morphogenetic impact of the substance, they design primers for their molecules of interest and perform PCR reaction and design further experimental approaches in short talks.
This way, the students gradually puzzle their theoretical knowledge and practical applications into a dynamic network to form a more solid background for further particularly practical and clinical education. Electronic materials for the course are available in the intranet university portal, a printed book is just under preparation.

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A Comparison of Single Oxime versus Oxime Mixture Treatment in Cyclosarin-Poisoned Rats

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The reactivating and therapeutic efficacy of two combinations of oximes (HI-6 + trimedoxime and HI-6 + K203) was compared with the effectiveness of antidotal treatment involving single oxime (HI-6, trimedoxime, K203) using *in vivo* methods. Quantitative histochemical method using in this study determined percentage of reactivation of cyclosarin-inhibited brain acetylcholinesterase in poisoned rats. Obtained data showed that the reactivating efficacy of HI-6 + K203 combination is slightly higher than the reactivating efficacy of the most effective individual oxime in all parts of the brain. In respect of brain parts, HI-6 + K203 combination was the most effective in the thalamus, basal ganglia and frontal cortex, all over 90% of reactivation. Based on the obtained data, we can conclude that the antidotal treatment involving chosen combination of oximes brings a beneficial effect for its ability to counteract the acute poisoning with cyclosarin.

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The comparison of the survival of P19-derived neuroprogenitors and P19 naive cells after intracerebellar application in a mice model with and without neurodegeneration.

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Mouse embryonal carcinoma (EC) cells (P19 line) were studied for their survival and developmental potential in the intact cerebellum of B6CBA mice and a model of neurodegeneration, in Lurcher mutant mice.

The P19 cells were cultured without differentiation or differentiated into neuroprogenitors using the retinoic acid. The intracerebellar application was performed in 73 mice - group A consisted of wt mice that received neuroprogenitors (n=21), group B of wt mice that received naive P19 cells (n=22), group C consisted of Lc mice that received neuroprogenitors (n=15) and group D of Lc mice that received naive P19 cells (n=15). The morphology of transplanted cells in the context of surrounding cerebellar tissue was evaluated after three weeks.

Transplantations resulted in survival and further neurodifferentiation of cells in 13 cases in group A, in 7 cases in group B, in 3 cases in group C and in 3 cases in group D which means that the survival rates are 62%, 32%, 20% and 20%. Statistically, there is a significant difference between the groups of Wt (group A and B) and Lc mice (group C and D).

This is a pilot study comparing the fate of transplanted EC cells in the cerebellum with and without a presence of neurodegenerative process. It seems that the differentiated elements do not survive less than undifferentiated and that the neurodegeneration affects the survival of EC derived elements.

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The Prague anatomist Hugo Rex (1861–1936)

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A double anniversary of a Prague citizen and anatomist falls on this year: Hugo Rex was born on January 9, 1861, i.e. 150 years ago, in Prague; graduated July 1, 1878 from the high school in Olomouc and December 1, 1885 from Medical School of German Karl-Ferdinand University, Prague (studied anatomy with professor Karl Toldt); 1880–1883 demonstrator, 1883–1895 1st assistant, German Department of Anatomy, Prague; February 28, 1889 habilitated for anatomy (postdoctoral thesis „*Beiträge zur Morphologie der Säugerleber*“); June 6, 1895 associate professor of anatomy; 1904–1909 substitute head of German Department of Anatomy, Prague between Karl Rabl and Otto Grosser; 1898 substitute professor of zoology, German Institute of Science and Technology, Prague; his field of work: comparative anatomy and embryology; he published 15 papers; retired in 1931, he remained in Prague until his death on August 13, 1936 (75 years ago).

Vertebrate jaw evolution (Prepattern/Cooption model): genetic, epigenetic or mechanic cause?

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The appearance of jaws was a turning point in vertebrate evolution because it allowed primitive vertebrates to capture and process large, motile prey. The vertebrate jaw generally consists of separate dorsal and ventral skeletal elements connected by a joint. How this structure evolved from the unjointed gill bar of a jawless ancestor is an unresolved question in vertebrate evolution. To understand the developmental bases of this major vertebrate evolutionary transition, we examined the expression of key genes responsible for pharyngeal patterning in the modern jawless fish, lamprey. Our analyses revealed, surprisingly, a nested pattern of *Dlx* genes and a combinatorial expression of *Msx, Hand* and *Gsc* genes along the dorso-ventral (DV) axis of the pharynx indicating that the sophisticated gnathostome-type pharyngeal patterning evolved before the appearance of the jaw. In addition, we find that *Bapx* and *Gdf5/6/7*, key regulators of joint formation in gnathostomes, are not expressed in the lamprey first arch while *Barx*, which is absent from the intermediate first arch in gnathostomes, marks this domain in lamprey. Based on these findings a new scenario was proposed in which cooption of "jaw-joint genes" into a pre-existing DV patterning program drove the evolution of the vertebrate jaw by altering the identity of intermediate first arch chondrocytes (Prepattern/cooption model: Cerny et al., PNAS 2010). In this lecture this model will be discussed in the context of mechanism of evolutionary change in order to reveal what can be considered as a cause of evolutionary change in the light of current knowledge.

New Insights into the Cell Biology of Stem Cells by Studying Prominin-1 (CD133)

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Prominin-1 (CD133) is a pentaspan membrane glycoprotein concentrated in plasma membrane protrusions. This cholesterol-binding protein is associated with lipid rafts, and it is expressed by several stem cells including those found in the neural and hematopoietic systems. Remarkably, it was demonstrated that in the developing embryonic mouse brain, CD133 is released from the neuroepithelial cells, concomitant with their differentiation, into the lumen of the neural tube by means of plasma membrane–derived vesicles raising the possibility that these vesicles might contain certain stem cell fate determinants. Does this hold true for other stem cells? We have investigated this issue using human CD133-positive hematopoietic stem cells (HSPCs) growing on primary mesenchymal stromal cells (MSCs) as feeder cell layer. We report here the following observations. First, CD133 is released from HSPCs into the culture medium in association with membrane vesicles that are sedimented after high-speed centrifugation. Second, these CD133-positive vesicles are enriched in cholesterol, and contain Flotillin-1/2 and Syntenin – a PDZ domain containing protein that interacts with the exosomal marker CD63 raising the possibility that these vesicles originate not only from the plasma membrane protrusions but also from intracellular vesicles. Third, the differential immunofluorescence revealed that CD133, in addition to its association with plasma membrane protrusions, is also present in intracellular structures, which at the electron microscopy level appear as multivesicular bodies demonstrating the association of CD133 with exosomes. Fourth, the amount of CD133-positive vesicles found in the culture medium increases upon cultivation whereas the number of CD133-positive cells are decreasing, indicating a link between the loss of CD133-positive vesicles
and cell differentiation in general.

Effect of Bradykinin after middle cerebral artery occlusion in rat

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Bradykinin, an endogenous nonapeptide produced by activation of the kallikrein-kinin system, promotes neuronal tissue damage, as well as disturbances in blood–brain barrier function, mainly through activation of B2 receptors. However, we hypothesized that an adequate dose of exogenous bradykinin used at an appropriate time after ischemia will be an effective stressor finalizing acquisition of ischemic tolerance, and able to turn proapoptotic pathway of delayed neuronal death to antiapoptotic. We investigated the effect of bradykinin used as postconditioning 3 or 6 hours and 1 or 2 days on neuronal cell survival after 60 min of middle cerebral artery occlusion (MCAO) in hippocampal CA1 region. Evaluation of infarcted volume was made using 2,3,5-triphenyltetrazolium chloride (TTC) staining. We demonstrated that focal ischemia by transient MCAO affects the hippocampus which responds bilaterally to the injury. Treatment with bradykinin (150 μg/kg i.p.) used 3 or 6 hour and 1 day after ischemia significantly reduced infarct volume and improved neurological score. Also administration of bradykinin reduces the number of Fluoro-Jade B positive neurons and increases NeuN immunoreactivity. Administration of bradykinin 3 or 6 hours and 1 day after MCA occlusion, ameliorates cerebral ischemic damage. Bradykinin postconditioning markedly reduced the degree of neuronal cell death, suggesting that it provided neuroprotection against delayed neuronal death in most sensitive brain neurons.

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Influence of pharmacologic preconditioning and ischemia /reperfusion on rabbit spinal cord neurons

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Our study aimed to characterized influence of pharmacologic preconditioning to motor neuron survival in the rabbit spinal cord ischemia/reperfusion.

Male rabbits were preconditioned by noradrenaline or bradykinin 48 h before 20 min of ischemia induced by an occlusion of the aorta below the left renal artery. Fluoro Jade B (FJB), NeuN, caspase-3 immunohistochemistry was used for qualitative and quantitative evaluation of motoneurons, as well as Tarlov scoring for evaluation of hind limbs function.

In ischemic group followed by 48 h of reperfusion necrotic changes of neurons were found. FJ B method showed increased number of degenerating neurons and animals were paraplegic (T-0). In the groups with noradrenaline or bradykinin preconditioning followed by ischemia and 24 h of reperfusion, Tarlov scores were better (3-4) than in the ischemic groups. Increased number of surviving cells was found after NeuN immunoreaction. In the groups with preconditioning followed by ischemia and 48 h of reperfusion there was strong correlation between Tarlov score and the number of surviving neurons in the ventral horns. In comparison to noradrenaline, bradykinin preconditioning showed improved neurological status (T 4-5) and more intact motor neurons were found.

Noradrenalin or bradykinin preconditioning reduces ischemic damage of spinal cord motoneurons by induction of tolerance.

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Comparison of the Knowledge of Anatomy Students at the Faculties of Medicine in Ostrava and Košice

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The basis for students of medicine is anatomy, which covers three semesters in the study programme of General Medicine. During the studies of this subject, the students expand their knowledge of human body structure and topographic anatomy.

The first semester is focused on musculoskeletal system and vascular and nervous system in the limbs, so that topographic-anatomical dissection of the limbs can be performed. The second semester concentrates on splanchnology and the area of large vessels and nerves. The third semester continues with the central nervous system. Practical knowledge of the brain and its cross-sections is one of the credit requirements for this semester.

The students' knowledge is being examined by tests and practical exams at the dissecting room in their practical classes. Ninety-nine students took part in the exam in Ostrava and 205 in Košice. The same test focusing on the limbs was used in both the groups. The limit for successful passing of the test was 75 points. In Ostrava, 63 students passed the test successfully, whereas 36 students failed. Therefore, the success rate was 63.6%. In Košice, 116 passed the test successfully, whereas 89 students failed, which equals to a success rate of 56.6%.

The test comprises statements of various facts and the students have to reply with either C – correct, or I – incorrect. It includes 20 questions, each having sub-questions a), b), c), d), and e). Overall, the students were to answer 120 questions within 30 minutes. The test was aiming and examining the students' theoretical knowledge in the area of the limbs and topographic anatomy of the limbs.

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Status epilepticus results in extensive brain damage in immature rats.

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Long – term discussion based on results of experimental studies and observations in children suggest that immature brain is lessly susceptible to seizure-induced damage than the mature, adult brain. In several reports was demonstrated that status epilepticus (SE) showed little or no neuronal damage in rats younger than 3 weeks. In contrast to this recently published clinical studies confirmed neocortical and hippocampal damage in children. These findings are in agreement with results of our experiments demonstrating that (SE) resulted in neurodegenerative changes not only in the limbic structures, but also in the neocortex and in several subcortical structures. Experiments were carried out in Wistar pups 12, 15, 18, 21 and 25 days old. Lithium - pilocarpine model of SE was used. The rats survived for 4, 8, 12, 24, 48 hours and 1 week after SE. Coronal sections (40 μm thick) were processed with a fluorescent stain Fluoro - Jade B (FJB).
Since P12 neuronal damage was consistently found in the dorsal hippocampus (CA 1), amygdala (cortical and basal ncc.), claustrum, in medial thalamic nuclei and in several neocortical areas. Since P 18 the number of affected structures increased and marked neuronal damage was found also in the piriform cortex, septum, ventral and dorsal striatum, lateral thalamic nuclei and in the posterior hypothalamus (mammillary complex). Since P18 neuronal damage was found in all survival intervals and reached peak at 24 and 48 h after SE.
Conclusion.
Lithium – pilocarpine model of SE resulted in immature rats in degeneration of neurons in the limbic and diencephalic structures, in the septum, basal ganglia and in the neocortex. Neuronal damage was found in 12-day-old and older animals, increased with survival interval and reached peak at 24 and 48 h after SE. Neuronal damage in majority structures was age-specific, and the vulnerability progressively increased with the age.

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Neuroinflammatory reaction of peripheral glial cells in two different models of neuropathic pain

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Proinflammatory cytokines contribute to induction and maintenance of neuropathic pain derived from cellular and molecular changes in the dorsal root ganglia (DRG) following a nerve injury. Chronic constriction injury (CCI) and spared nerve injury (SNI) are experimental models of neuropathic pain based on different pathways of signaling from distal nerve segment undergoing Wallerian degeneration to DRG. Adult Wistar rats operated aseptically on unilateral CCI (n=16) and SNI (n=16) of the sciatic nerve were left to survive for 1, 3, 7 and 14 days. Sham-operated (n=16) and naïve (=8) rats were used as controls. Changes of proinflammatory cytokine proteins (TNFa, IL-6) and their receptors (TNFR1, IL-6R) were detected in nerve segment distal to injury and L4-L5 as well as C7-C8 DRG of both sides by immunohistochemistry under the same condition. Activated macrophages were identified by immunostaining for ED-1.
Bilateral invasion of ED-1+ macrophages was observed in L4-L5 DRG of both models with a higher level in SNI rats. An increased invasion of ED-1+ macrophages distal to tibial and common peroneal nerve transection (SNI) than distal to CCI of the sciatic nerve was expected. Expression of cytokine proteins and their receptors was upregulated not only in bodies of the primary sensory neurons but also in satellite glial cells of L4-L5 DRG homonymous with, and C7-C8 DRG that are heteronymous with injured nerve. Schwann cells significantly contribute to elevation of cytokine proteins and their receptors in nerve segments distal to CCI and SNI. The results indicate different neuroinflammatory reaction of peripheral glial cells after distinct nerve injury and support our assumption that signaling for cytokine changes in DRG is apparently by blood stream.

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Galectins as inducers of production of a 3D extracellular matrix

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Adult stem cells, as the tumor cells, require special microenvironment for their survival. A key step to proper understanding of the biochemical aspects of the propagation of these cells is to characterize the "niche" that maintains their stemness. The extracellular matrix is known to play a central role. Studying the microenvironment in cancer stroma and wounded skin, we observed a high incidence of myofibroblasts and presence of an endogenous lectin, i. e. galectin-1.
Aim of the study: To infer a potential role of this certain class in the transition of normal human dermal fibroblasts to myofibroblasts.
Results: Galectins were found to stimulate this transition, galectin-1 being the most potent. The effect of galectin-1 was found to be independent on TGF-β1 (known to activate the fibroblast – myofibroblast transition) but both substances had an additive effect. Examining cell features associated with galectin stimulation, the galectin-exposed fibroblasts/myofibroblasts produced massively an extracellular scaffold rich in fibronectin and also galectin-1. When tested as substratum, it turned out to be beneficial for cultivation of keratinocytes without feeder cells. Interestingly, the keratinocytes cultured on this scaffold changed their phenotype and expressed keratin-19. Giving these observations a perspective, the described approach could in long term be of use in tissue engineering and wound management.

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Quantitative Assessment of Elastin in the Abdominal Aortic Aneurysmal Wall

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Introduction: Aneurysm of abdominal aorta (AAA) is an important disease. The contemporary research aims to decrease AAA mortality. Understanding the pathogenesis could open a new approach for the growth rate pharmacomodulation.

Aim: Elastin decrease and elastic lamellae fragmentation are featuring histological signs. The aim of our study was to quantify the amount of elastin in AAA wall and to find out, whether there is any difference between the elastin content and the symptomatology or size of aneurysm.

Methods: 46 AAAs were assessed (9 ruptures and 37 non-ruptures). The size interval was < 5cm for the small AAA, 5 ≤ middle AAA ≤ 7, big > 7 cm. By means of light microscopy, stereology and Ellipse software area fraction and length density of elastin in tunica media were quantified.

Results: Although there was less elastin in the ruptured AAAs, the difference between the ruptured and non-ruptured AAAs was not statistically significant. Middle sized AAAs were elastin richest, but even here the discrepancy in elastin content and size did not reach statistical significance.

Conclusion: Our findings did not prove any important relation between the amount of elastin in AAA wall, the size and rupture. Elastin decrease as its own does not seem to be able to explain the AAA growth and rupture and needs to be consider in context of other changes, the inflammatory infiltrate above all.

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Quantitative Histology and Proteomic Analysis in Abdominal Aortic Aneurysms

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Introduction: Incidence of abdominal aortic aneurysm (AAA) has been increasing in recent twenty years and so has its mortality. This is why AAA has been an object of an intense research.

Aim: We aimed to quantify the microstructure of AAA wall and thrombus to find out relations between the structure, size and symptomatology and also to correlate the qantitative with the proteomic data.

Material, methods: In 56 AAA wall and thrombi samples and 6 normal aortas, using immunohistochemistry and stereology, degradation of matrix proteins, leucocyte infiltration, phenotype of VSMC, fibrinolytic components and vascularization of AAA wall were quantified. For the statistical analysis correlation matrix, Kruskal-Wallis ANOVA and Mann-Whitney U-test were used.

Results: Some results confirmed either our preliminary hypothesis, or findings previously published in PubMed. We newly proved the microvessel density to continue in increase with the size of AAA, so that the big AAAs are the most vascularized. Secondly, inflammatory infiltrate is more abundant in asymptomatic AAAs. This finding might answer why the asymptomatic AAAs tend to rupture more frequently than the symptomatic ones. Increased levels of MMP-1, TIMP-2 and TIMP-1 in thrombus correlating with area fraction of granulocytes and contractile elements respectively in AAA wall might indicate the influence of thrombus on AAA wall.

Conclusion: Quantitative microscopy seems to offer a new insight into the morphology and pathogenesis of AAA, knowledge of which might enable causal therapy of the small AAAs.

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Applications of Slide-Based Single Cell Cytometry by TissueFAXS in Histopathology

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Background: Automated identification and cytometric quantification of molecular markers (antibodies, histological stains, genetic probes) on the single cell level in tissue sections by means of slide-based cytometry has become an essential tool in biomedical research and routine analysis in histopathology. TissueFAXS is the microscopic equivalent to flow cytometry – applicable to tissue sections, cell culture monolayers and cell smears/cytospin preparations and allows observer independent analysis of all types of cells in all kinds of tissue sections in-situ.
Technology & Method: The TissueFAXS system consists of a high-end motorized fluorescence microscope and sophisticated software for image analysis. The samples can be stained by means of immunohistochemistry or immunofluorescence. By using the nuclear marker (e.g. hematoxilin or DAPI) the location of the individual cells is identified and the system quantifies the staining intensity in each color channel for each and every cell. It allows even to distinguish between and compare between nuclear and cytoplasmatic marker expression in the same sample. For each cell or cellular compartment up to 14 parameters are measured and stored in a database. Representation of cellular parameters may be obtained by dot plots and/or histograms in a FACS-like manner.
Results: Results presented come from several studies and comprise different applications of slide-based cytometry.
(i) Phenotypic characterization of tissue infiltrating leukocytes in tumor biology1, transplantation immunology2 and autoimmune diseases3 exemplifying the quantification of markers CD1a, CD3, CD4, CD8, CD11c, and CD68 in renal cell carcinoma, renal allograft rejection and atopic dermatitis.
(ii) Signal transduction research in prostate cancer4,5 and brain neuropathology6 dealing with signal transducers and activators of transcription (STAT), and transforming growth factor beta.
(iii) Quantification of proliferation marker Ki67 and Her2 in breast cancer.
Discussion: The data above demonstrate novel approaches in research and improved data transparency in diagnosis using slide-based cytometry. Observer-biased visual estimation in immunohistological analysis of tissue samples is replaced by observer independent measurements on the single-cell level, which is especially important in multi-center studies and set-up of clinical studies for the pharmaceutical industry.

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Clinical anatomy of lymphatic drainage of the breast and parasternal region

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Lymphatics of the brest - normal individuals. Our results show that under physiologic conditions the natural dominant drainage for the outflow of lymph from the superficial areas of the breast are the axillary nodes. Although each quadrant of the breast is dominantly drained by one ore two of its own collectors, it is also interconnected via the subareolar plexus and lymphatics running outside plexus with the other quadrants of the breast. This fact is important for increase the risk of developing locoregional recurrences (Pavlišta, Eliška Lymphology 2005).

Parasternal lymphatics - normal individuals. The goal of this part is to describe the natural border between lymphatic drainage of the breast and parasternal lymphatic drainage. The study was performed on 18 female cadavers aged 52- 73 years. After local warming of the anterior part of the thorax to temperature 37 degrees Celsius gradually and slowly patent blue or mixture of Patent blue with India ink was injected cutaneously and subcutaneously on the both sides of the sternum in the level of the second to fifth intercostal space. Results: If the solution was injected in the distance 4 cm from sternum on the left and rigt side, the dye spreads predominantly to the subcutaneus tissue of the ipsilateral breast. If the solution was injected in the distance 1 cm from sternum (parasternal line) the dye spreads predominantly into the deep ipsilateral parasternal lymphatics and contralateral parasternal.

Lymphatics after surgery for breast cancer. After surgery lymph flow in breast region is changed. After total mammectomy and clearing of axilla lymphatics run along the scar and they are linked up to ipsilateral and contralateral axillary nodes ,scapular and abdominal lymphatics as well together with possible development of lymphedema. Partial mammectomy is discussed.

Mechanical properties of lamellar systems of the human osteon

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Mechanical properties of compact bone depend on degree of mineralization of osteons and interstitial lamellae and course of collagen fibers complex. One of the methods analyzing mechanical properties is the nanoindentation.
In the literature there are conflicting results concerning the degree of mineralization and corresponding microhardness of the central and peripheral location of osteon.

The aim of our study was to analyze selected mechanical properties of lamellar systems of the human femur compacta based on nanoindentation, to correlate results with the findings of other authors and to try to answer the fundamental question of hardness and elastic modulus of different parts of osteon.

Our preliminary results show that both the hardness and elastic modulus are higher within the wall of the central canal and cement line.
The detailed architecture of lamellar system is still matter of further investigation.

Loss of Galectin-9 from head and neck squamous cell epithelium is an emerging indicator of malignant transformation

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Galectins (Gal) are potent effectors in diverse cellular activities. Among them, Gal-1, Gal-3 and Gal-7 were already studied in connection with squamous cell cancer. In the context of these observations, we now focused on the first tandem-repeat-type galectin with Gal-9, specifically its presence in normal squamous epithelium of the head and neck, cancer and marginal zone of cancer resecate. Normally, Gal-9 was present in cells of the basal layer of the epithelium. When observing Gal-9-negative tissues, aberrant expression of keratins 14 and 19 was found. Furthermore, some of the marginal zones of tumors showed mosaic-like expression of Gal-9 and aberrant expression of keratins as well. All studied specimens of squamous cell carcinoma were negative for Gal-9. We have found no significant reactivity of tissues for biotinylated Gal-9 or its N-terminal domain. Treatment with neuraminidase generated reactivity to the N-domain, revealing impact of sialylation status. In summary, Ga-9 appears to be a marker of normal epithelium. Its absence in squamous carcinomas could become an indicator for the extent of cancer resection during the surgery.

Partnership network for Clinical Anatomy and Emergency Medicine education – VTEC

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At present, the Czech Army are mainly engaged in foreign missions according to political assignments. The fulfilment of such tasks is characterized by increased demands for training of military professionals not only in the physical and professional spheres but also in the psychological and medical aspects. When on foreign missions, Czech soldiers are expected to adapt to unfamiliar environments and situations. One such situation can even be encountering death, as well as instances when a soldier will have to give help to a colleague suffering from a severe devastating injury. In 2009, the 71st Mechanized Battalion of the Czech Army in cooperation with the Department of Anatomy and the Department of Forensic Medicine and Medical Law Faculty of Medicine and Dentistry Palacky University Olomouc, commenced a project aimed at soldiers' Psychological and Emergency Medicine Training prior to their participation in ISAF mission in 2010.This training consists of soldiers' participation in forensic and pathological autopsy with expert commentary, conducted in detail, and for the soldiers unique anatomical demonstration with expert commentary. In 2010 was establish partnership network between Department of Anatomy Faculty of Medicine and Dentistry Palacky University, Emergency Department of University Hospital and 7th Mechanized Brigade via the 71st Mechanized Battalion of the Czech Army. This working group is concerned with interactive and modern education of emergency medicine with anatomical and clinical relationship, intership education programme, real training programme on emergency medicine for students of Palacky University. In 2011 was this partnership network supported by European Social Fund in the Czech Republic, The Education for Competitiveness Operational Programme much the 24 mil.czech crown.

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Skin microscopic structure of the Common Pipistrelle *(Pipistrellus pipistrellus)*

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The bat wing is the most unusual structure for mammals. Skin covering wing membrane enables flying but also performs multiply functions like thermoregulation, gas exchange, water control and protective border against influences of environmental conditions or microbes. Here, we provide the comparison of microscopic structure of skin and the distribution of mast cells in different body parts of *Pipistrellus pipistrellus.*

Skin samples were collected from the wing membrane, intercrural membrane, dorsum, abdomen, head (between ears), ear and footpad. After fixation in 4% paraformaldehyde, serial transversal sections were prepared and stained with Hematoxylin and Eosin, Green Trichrom, Orcein and Toluidine Blue.

The thickness of epidermis and stratum corneum was the lowest in the wing and intercrural membranes in comparison to other body regions. The wing web skin was highly folded and lacked hair follicles and skin glands. It was composed of two epidermal layers separated by a central core of connective tissue with elastic fibers. The epidermis was consisted of one or two layers of flat epithelial cells and thick stratum corneum with multiple layers of squamous keratinocytes. Melanocytes with pigment granules were situated in the basal layer. The wing membrane was thicker in the area of wing trabecules, and contained numerous elastic and collagen fibers, isolated bundles of striated muscles fibers, larger size vessels and nerves.

The body skin was thicker in the dorsal part with prominent hairs and sebaceous glands. Epidermis was composed only from one or two non-pigment epithelial cell layers covered by outer keratinized layer. Orcein staining revealed the distinct layer of elastic fibers localized between dermis and epidermis. Hypodermis was relatively thin and contained skeletal muscle fibers. Groups of fat cells were situated under this muscle layer in the loose connective tissue.

The mast cells were localized in the dermis; mainly close the epithelium, around vessels and hair follicles. Their shape was an oval or round with relatively small number of granules.

Generally, the bat skin displayed the similar skin morphology typical for other mammals. The thick stratum corneum with multiple layers of squamous keratinocytes is probably a structural adaptation to sustain the continuous air abrasion and shearing stresses during flight as well as limiting percutaneous water loss.

The Connexin43 Carboxyl-Terminus: Regulator of Gap Junction Ultrastructure and Tissue Morphology at the Injury Border Zone Following Myocardial Infarction

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Gap junctions (GJs) are responsible for propagation of electrical excitation between myocardial cells. Connexin43 (Cx43) is the primary GJ isoform in working myocardium, where it localizes at intercalated disks. Previous studies from our lab have established that the interaction of Zonula Occludens-1 (ZO-1) with the carboxyl-terminus (CT) of Cx43 regulates the size of GJ plaques. We have further demonstrated that this interaction controls GJ size by governing the transition of Cx43 hemichannels into GJ intercellular channels. Pathologic remodeling of gap junction organization occurs in the transitional tissues between the infarct scar and surviving myocardium - termed the injury border zone (IBZ). The IBZ is the tissue locus within which re-entrant electrical arrhythmias are generated. We have recently reported that administration of a peptide mimic of the Cx43 CT improved scar morphology, reduced GJ remodeling and decreased arrhythmias after a left ventricular injury. Additionally, IBZ myocytes bordering scars in Cx43 CT mimetic peptide-treated hearts had lower densities of interdigitated collagen, periostin and non-myocardial fibroblastic cells, as well as greater myocyte-myocyte coherence. The changes in IBZ tissue organization prompted by the peptide suggested effects on myocyte-fibroblast interactions. Consistent with this, the Cx43 peptide reduced adherence between myocytes and fibroblasts in monolayer co-cultures. To investigate this further, we generated an in vitro 3D model of the IBZ. This comprised a core spheroid of red-fluor (CellTracker) tagged myocytes surrounded by a layer of green-fluor tagged fibroblasts. To do this, percoll gradient-purified myocytes from rat neonates were seeded into concave recesses in 96-well micromolded non-adhesive agarose hydrogels, where they aggregated into spheroids. After myocytes had formed a cohesive spheroid at 24 hours, fibroblasts were added to the myocyte spheroids. The sorting of the two cell types by differential adhesion was then followed by confocal microscopy of red and green live cells in the aggregates. Similar to IBZ in vivo, migratory invasion of the fibroblasts into the core of myocyte spheroid was reduced by peptide treatment relative to controls. The data suggests that one mechanism by which the Cx43 CT mimetic peptide reduces arrhythmia following myocardial infarction is via effects on a fundamental morphogenetic process. Specifically, the peptide may modulate patterns of differential adhesion between myocytes and fibroblasts in the IBZ following injury.

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Correlation of the expression of apoptic genes between donor and recipient in small intestine transplantation model

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Transplantation of the small intestine is on the one side very serious and invasive surgery but on the other it is able to prolong the life of the patient suffering from „short Bowell syndrome“ for instance. The small intestine is the source of production of proinflammatory mediators leading and contributing to multiorgan failure. The group of organs which show the most common potential dysfunction comprises mainly the lungs, liver and kidneys. The endoplasmic reticulum (ER) highly influenced by transplantation is significantly involved in the apoptosis activation of different small intestine cell populations.

The aim of this work was to analyse and compare the changes of expression in proapoptic and antiapoptic genes of ER chaperones after heterotrophic allotransplantation of the small intestine in rats for time periods 1 hour and 6 hours after transplantation.

Total RNA was isolated from the complete jejunal wall. After isolation of mRNA, RT-PCR was used with consequent electrophoresis and data evaluation.

The mRNA levels of both proapoptic gene Gadd153 and antiapoptic gene Grp78 was after 1 and 6 hours survival significantly higher in grafts (by 155 ± 20,5 %) as well as in recipients intestine (by 102 ± 15,8 %) against control group. There were also significantly higher levels of Grp78 mRNA in recipient`s intestine (37,5 ± 8,2 %) and grafts (42,6 ± 10 %) after 6 hours of surviving in comparison with both group after 1 hour survival.

Studying the gene expression in the early stages of the small intestine transplantation model can help to understand apoptotic processes triggered by endoplasmic reticulum in the different cell populations.

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The dissecting essay - our experience in a seminary work for students of the 1st year

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A seminary work as credit condition in anatomy for study program General medicine was firstly required in the academic year 2004/05. This written work concerned a description of dissecting procedure and has become known under a title "Dissecting essay". Authors of this presentation would like to share their several-year experience with assignment of topics, evaluation of works, and feedback acquired in form of an inquiry from students.

Increased number of students in previous years results in problems with want of dissecting regions. It was impossible to exercise all students by manual work at the same time. Students waiting for their opportunity often wasted their time and were frustrated.

Now, while a part of a study group works at one intact cadaver, the other students get simultaneously a body undergone one-year routine pre-graduate dissection. There the instructor selects a small region to complete the dissection. This manual work is not intended for fast progress, but for detailed study. It results in the dissecting essay - written work in extent of 2-4 pages containing a theoretical introduction, description of actual situation on a cadaver, and a report of an advance of the student's work. Further, we require a hand-made picture - at least a simple scheme, of course with preference of found variations. The final mandatory item is the list of the sources used for the dissection and text of the essay including eventual copied pictures. The evaluation is only in two levels: accepted / not accepted.

In the dissecting essay, a student gives evidence of integration of his manual skill, theoretical knowledge, bibliographic search, own invention, interpretation and presentation of his own work. The essay should reach expert, stylistic, and graphic levels of pre-graduate seminary work.

Asset of this "duty" appears to be high. Even students themselves concede this fact in the inquiry. Work on this brings a motivation to dissect in the time when students only remove fatty tissue in subcutis in the intact cadaver. The work refines written style, instils graphic layout of a printed document. A narrow topic makes the student to use more then one textbook. It supports more responsible approach in the dissection. Last but not least, the cadavers after related dissection prove to be excellent study material for demonstration of muscular, vascular, and nervous structures for the next year's students.

E-learning course Topography of head and neck

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The poster presentation shows the work on the E-learning course Topographic anatomy of head and neck using LMS Moodle on server of Faculty of Medicine in Hradec Kralove. This new course is further of serial of similar courses presented before. All of them are induced by need of our students faced with our subject in 2 semesters only. The time stress makes students to focus on systematic anatomy and to neglect the topographical anatomy. For this reason the courses aspire to complete current contents of practical classes and to clear blind spots in syllabi, as well as in motivation of students.

Specific way of our solutions results from consequent procedure. The first step is a diligent dissection of the selected region of the head or the neck, occasionally adjustment of older dissected specimens followed by photo making and drawing. After data capturing there is time for work with graphic editors or video studio in case of video files. The other used software includes html editors and text editors. The records are then loaded into modules of LMS Moodle or they are made in Moodle directly on-line (quizzes, questionnaires).

The presented course is composed from own texts and remarks, authorized pictures and photos of our dissection specimens. Authorial approach should guarantee an absence of any extraneous copyrighted materials.

Our course is fully equipped by feedback components built both for students and also the creators, so the course can be improved and completed continuously. Students need to confirm after their study of any lesson by a quiz whether they understood the topic and they are ready to continue by following lesson. On the other hand, the author of a course has a feedback from questionnaires or discussion forums which can be either generalized or aimed to any specific problems of studies by appropriate question.

In conclusion, we effort to create a sofisticated source of informations but it does not mean that we plan to replace regular education in practical classes. The study of anatomy belongs into dissection rooms seriously. We would like to enable students to repeat the contents of classes after education or to prepare better for following classes in comfort of computer rooms. Of course, such courses can be very useful for the time of vacation when is not possible to study in the dissection rooms for incomplete staffing. Moreover, the course motivates the students and offers them to cooperate on the education by participation on feedback.

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Invasion of macrophages distal to neonatal and adult nerve crush and its relation to neuropathic pain induction.

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Peripheral nerve injury induces an invasion and activation of active marcophagesmacrophages in thea distal nerve stump of sciatic nerve. The goal of our work was to investigate invasion of macrophages distal to nerve crush in relation to duction and conditions for neuropathic pain induction in neonatal and adult rats. Ten neonatal rats (Wistar) were divided into two groups: neonatal crush injury (n=5) and control group (n=5). Three 3 rats from the control group were subsequently used for adult crush injury, another two rats for sham-operation. Mechanoallodynia and thermal hyperalgesia were measured 3 weeks after the neonatal and adult crush in both neonatal and adult rats. Nerve sample 5 mm dDistal to crush stumps of the sciatic nerve and contralateral counterparts were removed from neonatal and adult experimental rats after measurement of behaviouralbehavioral tests and perfusion with . Rats were deeply anesthetized with a lethal dose of CO2, perfused transcardially with Zamboni´s fixationve. Immunofluorescence staining of transversal distal stump sections for gap43 and ED-1 wasere used for detection of activated macrophages. ED1+ macrophages were quantified by means of image analysis (LUCIA G). Despite of thermal hyperalgesia mechanoallodynia and thermal hyperalgesia mechanoallodynia observing in adult rats, thermal hyperalgesia was no´t detected observed in neonatal rats but mechanoallodynia was. Ratio of ED1+ macrophages in nerve samples of ipsilateral and /contralateral sides was 2:1 in neonatal rats, but . Otherwise the ratio was 40:1 in adult rats. From our results we can conclude that an increased invasion amount of activated macrophages occuredoccurred in the distal stumps correspond with increased elevated behaviouralbehavioral signs of neuropathic pain in adult than neonatal rats.

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Effect of prenatal administration of folic acid on retinoic acid induced developmental anomalies in chick thymus

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We examined the possible protective effects of prenatal administration of folic acid on retinoic acid induced, defective thymus model in chick. Chicken eggs were divided into two experimental groups. First group was injected with, 0.3µg of retinoic acid (R2625) via yolk sac. Second group was concomitantly administered additional 0.5µg of folinic acid. Groups were compared with each other and with sham injected matched control. Total volume of thymus was calculated by employing software Image J on serial histological sections. Data was analyzed quantitatively for total volume of thymus, number of lymphocytes in cortex and medulla and the size and number of Hassall's corpuscles in a unit area. The intensity of staining for collagen and reticular fibres were analyzed qualitatively. Our results show that administration of retinoic acid resulted in statistically significant decrease in the volume of thymus. In addition there was marked decrease in the number of thymocytes and epithelial cells, loss of prominent interlobular septa and increase of reticular fibres. There was also increased mortality, behavioral effects, delayed hatching, limb deformities and failure of yolk sac retraction. Folinic acid improved hatching significantly, but failed to completely reverse the damage induced by retinoic acid in thymus and other structures studied. We conclude that retinoic acid teratogenesis is mediated via more than one pathway. The partial protection imparted by folinic acid and failure to completely rescue defects, could be due to multifactorial aetiology of retinoic acid. Moreover a higher dose of folinic acid may be required to significantly increase the level of signaling molecules to counter the effects mediated via disturbing neural crest pathway.

Variations of the pneumatization of the ethmoid cells

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Variations of the pneumatization of the ethmoid cells mean expansion of the cells to the surrounding structures, which may lead to changes in their relationship. The aim of this study was to assess the incidence of some variations of the extensive pneumatization of the ethmoid cells - concha bullosa, infraorbital ethmoid cells, sphenoethmoid cells by CT scans. 56 CT scans of paranasal sinuses of patients older than 18 years were reviewed retrospectively. Images were assessed in coronal and axial planes with 1 mm section thickness. 3 CT scans of the patients with presellar type of the pneumatization of the sphenoid sinus were excluded from evaluation of the sphenoethmoid cells. The extensive pneumatisation of the ethmoid cells into middle nasal concha which forms the concha bullosa was detected in 58.9% of patients. The pneumatisation was evaluated in the lamellar, bullous and both portions of the middle turbinate in 35.7%, 19.7% and 14.3% of cases, respectively. There was a significant correlation between the presence of the concha bullosa and contralateral nasal septal deviation ($p<0.05$). The other variations - infraorbital ethmoid cells and sphenoethmoid cells were found in 14.3% and 22.6% of patients, respectively. CT is adequate method for visualisation of paranasal sinus anatomy and successfully detects the anatomical variations of the sinuses. It is essential in preoperative planning. Coronal CT scans were preffered for detection of concha bullosa and infraorbital ethmoid cells while the observation of sphenoethmoid cells was evaluated in both coronal and axial planes. Awareness of these different variations can help surgeons in their orientation during endoscopic surgical procedures and can decrease the risk of intraoperative complications.

Distribution of monoamine oxidase A in normal and tumor kidney tissue

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Identifying consistent changes in cellular function that occur in many types of cancer could revolutionize the way cancer is treated. Previous work has produced promising results such as the identification of p53 in tumor tissues. Recently drugs that affect serotonin reuptake were shown to reduce the risk of colon cancer in man.

Here, we analyzed expression of monoamine oxidase A (MAO-A) in 63 samples of renal cell carcinoma (RCC). To demonstrate MAO-A, we have used indirect enzymatic method by following primary antibody: rabbit polyclonal antibody MAO (H-70): sc-20156 (Santa Cruz Biotechnology, Inc.). We compared expression of MAO-A between cancerous and normal tissues of kidney. Our analysis found significant downregulation of MAO-A, the mitochondrial enzyme that degrades monoamine neurotransmitters including 5-hydroxytryptamine (5-HT, or serotonin) and norepinefrine (NE) in tissue samples of RCC. MAO-A expression was decreased in 52 (82.53%) cases of RCC compared to the non-cancerous controls.

Future studies should investigate links between MAO-A supression and the development of cancer and determine that MAO-A supression contributes to increased cancer risk.

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The influence of FGF inhibitors on embryonic development

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The family of fibroblast growth factors (FGFs) regulates many developmental processes such as brain patterning, branching morphogenesis and limb development. The FGF ligands carry out their functions by binding and activating the fibroblast growth factor receptors (FGFR). There are 22 fibroblast growth factors and three FGF receptors. Deregulated FGF signalling can contribute to pathological conditions either through gain – or loss-of-function in ligands or receptors. The aim of our recent project is to compare the effect of two FGF inhibitors - PD161570, PD173074 (Tocris Bioscience) on limb development. While PD173074 is inhibitor of FGFR1 and FGFR3, PD161570 antagonizes all of the FGF receptors. We used chicken embryos where limb development is initiated at stage HH17. FGFR inhibitors were injected into the right wing limb anlagen at developmental stages HH20–22 when the limb already protruded as the bud from the body. The embryos were kept in humid incubator for other ten days. Samples for skeletal analysis were fixed in 100% ethanol and stained with Alizarin Red and Alcian Blue. Wing bones were analyzed for possible abnormalities and measured in programme AxioVision Rel. 4.8. Expression of FGF receptors was analyzed by whole mount in situ hybridization and QPCR method. We determined that both inhibitors induced shortening and deformation of limb with reduced autopodium. Limbs treated by higher concentration (100mM) stopped their development at early bud stage and right wing did not proceeded further despite of normal development of control left wing.

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CB2 receptor protein and mRNA expression in dorsal root ganglia of two rat neuropathic pain models

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Agonists of cannabinoid receptors (CB1R and CB2R) modulate nociceptive signal transmission and seem to be promising in treatment of neuropathic pain. Importantly, CB2R activation inhibits pain responses without adverse, most often psychotropic effects that are produced by CB1R agonists. CB2R synthesis and protein distribution of protein in the dorsal root ganglia (DRG) were investigated using immunohistochemistry and in situ hybridization in two rat neuropathic pain models– chronic constriction injury (CCI) and spared nerve injury (SNI).

Both types of nerve injury induced symptoms of neuropathic pain which were measured as a decrease of paw withdrawal threshold (mechanical allodynia) and withdrawal latency (thermal hyperalgesia). CCI and SNI resulted in up-regulation of CB2R in both neurons and satellite glial cells of the rat DRG related to injury. Unilateral nerve injuries always induced bilateral increase of both CB2R protein and mRNA comparing with sham-operated animals. Bilateral CB2R up-regulation was found not only in DRG associated with damaged nerve but also in those of spinal cord segments remote from the operation (C7, C8 levels). Increased CB2R protein in DRG of both neuropathic pain models was confirmed by expression of CB2R mRNA using in situ hybridization.

Our results suggested an activation of endogenous cannabinoid system expressed by CB2R during neuropathic induction.

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Chick embryo as a model for normal and abnormal organogenesis - The elective course in the Medical Curriculum at the 3rd Faculty of Medicine.

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The introduction of the new teaching course , The Embryology and the Developmental Toxicology, to the Medical Curriculum aims to extend the way of the Embryology teaching . The demonstration of the clinical and experimental applications of embryology may attract the students interest to the study of Embryology . The experimental application of embryology is demonstrated on the model experiment for embryotoxicity assessment in the population of chick embryos. The embryonic exposure during pregnancy and its risk assessment is another, clinical and epidemiological application of embryology. Both applications are presented practically. Students have the opportunity to take active part in the real experiment and in the standard practice in the Czech Teratological Information Service teratological counselling. The course of the experiment (manipulation with fertilized eggs, incubation, application of a tested drug, embryo examaination) is documented in photographic and video material step by step. Demonstration of normal and abnormal development during organogenesis is documented as well. The existing electronic material is accessible in electronic tools (Moodle) and can be used for the distant study by the pregraduate and postgraduante medical students from different medical faculties.

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Morphological changes of large conduit pulmonary arteries during posthypoxic recovery

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Except our interest in pulmonary resistive arteries and hypothesis that the effective stabilization of MC granules by sodium cromoglycate (SC) affects hypoxic remodelling and post-hypoxic re-remodelling of the pulmonary arteries, we have also noticed interesting and not published changes at the level of large conduit arteries (LCA) Ø >100 μm. These arteries are not dominant in the determination of pulmonary blood pressure, but their morphology is changing during hypoxia and post-hypoxic recovery.

We assessed the changes of tunica media thickness of LCA depending on the duration of recovery phase and variant timing of SC administration. Total 56 Wistar-Han male rats were divided into 7 groups by 8 individuals, exposed to chronic (21-day) normobaric hypoxia (10 % of oxygen) and allowed to recover 4 days or 21 days with variant timing of the intraperitoneal SC administration. On the lung slides, histological changes were assessed by a light microscope and the LCA tunica media thickness was measured using image analyzer NIS elements 3.0 AR.

In both 4 days and 21 days recovery group, we revealed hypertrophy of LCA tunica media persisting since the hypoxic period with no re-recovery processes and no influence of MC stabilization, either.

We suppose that there are also another factors than MC products, which play role in pulmonary arteries remodelling.

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Topography of the posteromedial complex of the human knee joint

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There are different opinions on localization, course, attachments and portions of the medial collateral ligament (further MCL), which is essential stabilizing component of the human knee joint
The purpose of this work is to analyze the variability of the course , shape, attachments , dimensions and topographical relations of MCL. Twenty two intact human knee joints (age 60 – 78 years) were used in this study.
MCL has two portions , superficial (this is the second layer of the medial compartment) and deep (this is the third layer of the medial compartment). Both portions are variable, the deep one more than superficial. The findings show 68% of common proximaol attachment and 32% of two site attachment. The portions differ in dimensions: the superficial portion is 9 - 14 mm wide, 1,8 – 2,2 mm thick and 80 - 110 mm long, deep portion is 8 – 9 mm wide, 1,6 – 2,1 mm thick and 42 – 70 mm long.
The distal attachment of the superficial portion lies 50 – 60 mm bellow the level of articular fissure and that of the deep portion just 10 – 18 mm . Distal attachment of the deep portion varies in three different locations.
Our findings are important for safer orientation on medial joint wall in total knee arthroplasty.

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Topographic anatomy of lumbar sympathic chain regarding lateral trans-psoatic approach to the lumbar spine.

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Extreme lateral interbody fusion (XLIF) is a novel technique for the anterior disc replacement. Patients who presented with symptomatic degenerative disc disease or failed back surgery syndrome are candidates for this surgery. The aim of this report was description of the anatomy of the sympathetic chain of lumbar spine regarding this XLIF surgery. The patient is placed in a true right lateral decubitus position and small (6 cm) left lateral skin incision is performed. Access to the lumbar spine is achieved by approach that passes through the retroperitoneally fat and psoas major muscle, using peroperative fluoroscopy. Expandable retractor is inserted, discectomy and replacement by Oracle cage (Synthes, USA) with synthetic cancellous bone to the interbody space is performed. XLIF represents save surgical method with maximally careful approach and spacious working portal. The new benefit of XLIF is based on the minimally invasive spine surgery technique through retroperitoneal space. The lateral access to the disc avoids the major vessels and nerves and implant placement in the anterior
and bilateral position provides sagital and coronal plane imbalance correction.

Apoptosis in Placental Vasculature from Pregnancies of Mothers with Diabetes Mellitus Type I

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Increased level of angiogenesis was reported in placentas from pregnancies complicated by DM type I. While neovessel formation is manifested by an increased proliferation rate, parts of microvasculature might be also eliminated via apoptosis.

We decided to quantify the number of apoptotic cells in placentas from normal pregnancies and pregnancies complicated by DM type I. The main goal of this quantification was to determine whether increased angiogenesis in placentas from pregnancies of DM type I mothers is associated with an increased level of apoptosis in the placental vasculature.

The placentas from normal pregnancies (n=8) and placentas from mothers with DM 1 (n=18) were obtained at the time of delivery. The number of active caspase-3 immunoreactive cells (apoptotic cells) was quantified and normalized to the cross-sectional surface of villi.

In the diabetic group, an average number of apoptotic cells in vessels was 18, 1±18, 7/mm^2. In placentas from normal pregnancies, this value was 8, 3±5, 7/mm^2. The difference was not statistically significant. Placentas from pregnancies with good metabolic compensation had 17,0±15,2 apoptotic cells in vessels per mm^2 of villous cross-sectional area, while in those from pregnancies with poor compensation this value was 18,8±22,1/mm^2. Neither this difference was statistically significant.

Regarding vascular changes previously observed in DM type I placentas it could be concluded that these arise as a consequence of neovessel formation, but without significantly increased apoptosis of cells within the vessel wall.

The work was supported by the Grant GACR No.304/09/0733.

Prenatal Developmental Stages of the Human Heart

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Using original techniques the morphology of the heart tube, heart loop, embryonal heart and fetal heart will be presented.
Original findings: Such as the muscular bridges of the heart loop and the glucosominoglycan (GAG) connective tissue complex will be presented. The GAG complex of the heart has an atrioventricular portion, septal portion and aortopulmonal portion. Developmental significance of the GAG complex of the heart is following:
1. division of atrioventricular canal and formation of cuspid valves,
2. formation of membranous interventricular septum,
3. aortopulmonary separation,
4. separation of atrial and ventricular myocardium.

Comparison of proliferative potential of normal and diabetic term placenta

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In order to fulfill fetal requirements the placenta grows during the whole pregnancy. Placental growth is based on a continual proliferation of all kinds of cells constituting the organ. The aim of this study was to compare the proliferative potential of normal and diabetic placenta at term.

Specimens collected by systematic random sampling from eight normal placentas and eighteen placentas of mothers suffering from type I diabetes mellitus (DM I) were fixed in formaldehyde and embedded in paraffin. Histological sections cut from five randomly chosen blocks per placenta were used for immunohistochemical detection of Ki-67 antigen as a marker of proliferation. The numbers of Ki-67 positive nuclei were counted in trophoblast, stroma and vascular wall and the areas of stem, intermediate and terminal villi were separately measured in 20 fields of view per section. Mean numbers of labeled nuclei per squared millimeter of villous tissue were calculated and data were statistically analyzed.

In both examined groups the Ki-67 positive nuclei occurred in cytotrophoblast, stromal cells and cells of vascular wall in all types of villi. In DM I placentas we observed conspicuously lower frequency of those nuclei in pathological forms of villi. Counted for all types of placental villi overall the mean number of labeled nuclei of cytotrophoblast just like the mean number of labeled nuclei of endothelium were significantly lower in DM I group (25.8 ± 12.2 vs. 12.3 ± 6.6 and 7.7 ± 4.0 vs. 3.9 ± 3.0 respectively). The same parameters counted separately for terminal villi were also significantly lower in DM I placentas (32.2 ± 14.7 vs. 19.9 ± 12.2 for cytotrophoblast and 10.1 ± 4.6 vs. 5.2 ± 4.5 for cells of capillary wall.

We conclude that maternal diabetes may cause lower ability of placenta to enlarge the area of syncytiotrophoblast and capillary wall in terminal phase of pregnancy and thus adversely influence fetal well-being.

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The effect of mesenteric ischemia-reperfusion on the intestinal wall morphology in rats

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The progress of jejunal damage and restitution in the course of mesenteric ischemia-reperfusion injury at different time periods was investigated. Adult male Wistar rats were randomly assigned to one of the experimental (IR) or sham groups (S). IR animals underwent occlusion of the superior mesenteric artery by microvascular clamp for 1 h followed by 1 h (IR1), 24 h (IR24) or 30 days (IR30, each n=10) of reperfusion periods. The sham-operated rats (S) underwent midline laparotomy and tissue samples were taken in the same time periods as for the IR groups (SC1, n=5; SC24, n=5; and SC30, n=5). Mesenteric ischemia lasting 1 h followed by 1 h of reperfusion caused significant disintegration of the mucosa, reduction of the muscular layer and diminution of the wall thickness. Intestinal specimens from rats after 1 h of reperfusion (IR1) exhibited the most prominent damage (HII = 5.27±0.30), ranging from multifocal lifting of the epithelial layer to disintegration of the villous tissue locally with crypt layer destruction and transmucosal infarction. The HII was significantly higher than in sham-operated group SC1 ($p<0.001$). Reduction of villous height ($p<0.01$) as well as intestinal crypt depth ($p<0.001$) between IR1 and control group SC1 was observed. In the IR1 group, significant reductions of thickness in the muscular layer (IR1 vs. SC1, $p<0.05$) and the intestinal wall (IR1 vs. SC1, $p<0,001$) were found. After 24 hours of reperfusion, restitution of the mucosa was found, expressed by normal villous morphology and re-epithezation, although the reduction of smooth muscle layer of intestinal wall was still prominent (IR24 vs. SC24, $p<0.01$; IR24 vs. IR1, $p<0.001$; IR1 vs. IR30, $p<0.001$).

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Penetration of dextran from subarachnoid space to the dorsal root ganglion and its stimulation of macrophage invasion

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Neuroinflammatory reaction is spread bilaterally alongside neuroaxis from lumbal to cervical dorsal root ganglia (DRG) following unilateral nerve ligature used as neuropathic pain model (1). We hypothesize that signal molecules may diffuse from liquor into DRG. The goal of our experiments was to confirm hypothesis on intrathecal (IT) pathway of neuroinflammatory signal molecule diffusion into DRG.
Six adult male rats (Wistar) were divided into 3 groups. Rats of group A with IT application of FluoroEmerald (FE) (n=2), unilateral sciatic nerve ligature was applied before IT application of FE in rats of group B (n=2) and rats of group C were sham-operated (n=2). FE was injected via the cerebellomedular cysterna and rats were left to survive for 16 hrs (group B, C) and 48 hrs (A). After the time of survival, the rats were deeply anesthetized, perfused transcardially with Zamboni´s fixative and cervical and lumbal DRG were removed and immersed in the same fixative overnight. The FE staining was observed in DRG cryostat sections under fluorescent microscope. A part of sections was incubated with GFAP and ED1 primary antibodies to detect the satellite glial cells (SGCs) and activated macrophages, respectively.
We found intense FE staining in ED1+ macrophages invading both cervical and lumbar DRG removed from rats of group A and B. Particles of FE diffused into SGCs of all DRG and into the neuronal bodies only in DRG of cervical segments. Penetration of FE was higher in cervical DRG caused by a gradient of concentration. Amount of macrophages containing FE was higher in both cervical and lumbal DRG of contralateral side in group B rats.
We confirmed that FE penetrates from liquor into DRG and causes strong immune reaction.

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Clinical anatomy of arteries of upper extremity

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Introduction: The vasculature of the upper extremity is a heterogeneous system of vessels. Eve the more constant arteries show extreme differences in variations incidence. Moreover, the principal trunks can serve as a graft for cardiosurgery, as a conduit for the intervention cardiologist, as a site for shunt application for the vascular surgery or as a source vessel of flap for plastic/reconstruction surgery.
Material and methods: Cadaveric study of 135 dissected extremities and radiodiagnostic study of 3609 catheterization videos.
Results: More than 50% of patients/cadavers showed variable arrangement of arteries in the axilla, more than 23% showed variable arrangement of the radial artery but less than 5% showed variable arrangement of ulnar artery. The median artery (*arteria comitans nervi mediani*) is a real problem for the correct incidence determination due to permanent inaccuracies in definition of this vessel, but when present in both forearm and hand, its incidence is approximately 12 % of cases.
Conclusions: The particular knowledge of the vascular anatomy of the upper extremity is fundamental for any clinical intervention. Nowadays, the radial approach is the intervention technique of choice in procedures of coronary arteries thanks to lower incidence of complications, better comfort to the patient and lower financial support.

The anniversary of publication of Anatomiae Pragae

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Anatomiae, Pragae, anno M.D.C. abs se solenniter administratae historia (*History of the Dissection Done By Him Solemnly in Prague A.D. 1600* is a fantastic book describing in a very detailed way the performance of a public anatomical dissection. Both the performed dissection and the issued book are to be attributed to Johannes Jessenius (Ján Jesenský). He was of Slovak-German origin, born in Wrócław in 1566, studied in Leipzig, Wittenberg and Padua. Although he worked in Wittenberg, he was strongly tempted by chance to join Prague court of Emperor Rudolph II. In 1600, thanks to Tycho Brahe, he arrived to Prague to draw attention to his personality by the dissection. Then he moved to Prague and work here as physician but disapointed left for Vienna to serve Emperor Mathias. But in 1616 he finally returned to Prague, where he was twice elected rector of Prague University. He joined the Czech protestant side and therefore was after the battle at Bílá hora punished (being beheaded). But his work survived as a witness of late-Renaissance level of anatomical knowledge, although we are not able to trace any personal Jessenius´ discovery in the field of anatomy at all.

Hyrtl in eponyms

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Last year, we commemorated 200-year-anniversary of Joseph Hyrtl (1810-1894) birth. He was a famous anatomist who studied medicine in Vienna and spent 8 years of his carrier at Karl-Ferdinand University in Prague (1837-45). Then he left for Vienna to be the head of the Anatomy Department at Vienna University and later a rector (1853).

Due to his unique personality of excellent anatomist, his surname is connected (thanks to both famous textbooks and quality articles) with many anatomical structures throughout the human body. Some of them are homonymous and some synonymous, however, we can find 16 different eponymous terms in human and 1 in animals, together encompassing 25 different structures. Fortunately, we can conclude that only 2 are actively applied in current science: Hyrtl´s fissure (fissura tympanomeningea) in embryology and otology throughout the world, and Hyrtl´s canal (canalis musculofibularis) in education of anatomy mainly in the Central Europe. Both terms are not part of Terminologia Anatomica.

A case report on accessory brachial artery

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Introduction: Although anatomical variants are frequent matters, some of them can gain its clinical relevance during introduction of new diagnostic and/or treatment methods. Such case can represent the accessory brachial artery.
Material and methods: During routine dissections two cases of this variant were noticed.
Results: In first case, the accessory brachial artery originated from the axillary artery, descended along the arm and rejoined the brachial artery within the medial bicipital groove. It was approximately 18.5cm long and its outer calibre was 5mm.
In second case, the accessory brachial artery originated from the axillary artery, descended along the arm ventrally to the median nerve, giving little branches to surrounding structures and rejoined the brachial artery above cubital fossa. Its outer calibre was 2mm.
Conclusion: This quite rare variations (less than 1% of cases) can cause failure of catheterization, if present, due to its too narrow caliber.

The body composition of the young school 7-9 year-old boys and girls

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Overweight and obesity are found in the present population of children more frequently. The aim of our research was to identify anthropometric parameters and body composition in healthy children of both gender aged 7-9 years. 116 boys and 98 girls from three elementary schools in the Olomouc region were measuring. Basic anthropometric data were collected by the help of conventional anthropometric methods. We have determined the body height and weight, counted the body mass index and assessed the ratio of skeletal muscle and fat by method of bioelectrical impedance (In Body R20). For statistical evaluation we used paired t-test. Results: The height and weight of both girls and boys is corresponding to values from 6th Nation-wide Anthropological Survey of Children and adolescents 2001 Czech Republic. According to the percentile zones of body mass index was 10.34 % of the boys and 5.18 % of the girls in the zone of overweight (90th - 97th percentil) and 6.03 % of the boys and 4.08 % of the girls in the obesity zone (over 97th percentil). Average percentage of body fat increases from 7 to 9 year-old: at girls 16.80 %, 18.30 % and 19, 06 %; at boys 13.18 %, 15.04 % and 17, 32 % for each age group.

Spatial changes of vascular patterns in limb buds transplanted on CAM.

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It is accepted that the ZPA (zone of polarizing activity) described as an area of proximal and postaxial mesenchyme in limb primordium, influences posteroanterior arrangement of limb tissues while the complex of AER/SZ (apical ectodermal ridge/subectodermal mesenchymal zone) regulates proximodistal limb growth. Further, characteristic limb vascular pattern (exhibiting the preaxial, postaxial outflow stems, interskeletal plexi, and marginal sinus), as well as continuous blood circulation are essential for normal limb outgrowth, too.
The questions are: 1) How the posteroanterior differentiation of limb transplants occurs due to renewed circulation and 2) Are there some changes in expression of ZPA in relation to the vascular rearrangement?
To examine revascularization of vascular pattern two groups of transplantations using White Leghorn embryos as donors (HH 21-23) and hosts (HH 35-37) were done: first, the wing limb buds (composed of prospective zones of stylopodium, zeugopodium and autopodium) and second, the wing buds with adjacent lateral plate mesoderm (prospective limb girdle area), were transplanted into chick chorioallantoic membrane. The vascular patterns in the developed transplants were studied using intravital injection of sonicated and filtered Indian Ink solution. Further, an immunohistochemical detection of Sonic hedhegog was used to display ZPA activity in whole mount embryos.
It is shown that postponed outflow of blood, postaxially located extravasates and circulation disturbances in limb transplants correlate with hypoplastic changes in the ZPA areas. Zeugopodial bones are short and ulna is less differentiated than curved radius. The limbs are tortuous and bent postaxially; these findings are frequent namely in transplants containing adjacent lateral plates, where activity of ZPA was not well-established.

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The expression of NADPH-d in developing and adult rat hippocampus

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This study describes the expression of NADPH-diaphorase (NADPH-d) activity in hippocampal neurons of the rat during postnatal life and in the adulthood.

Nitric oxide (NO) is an intra- and extracellular messenger with multiple functions in the developing and adult brain. NO is produced by several isoforms of nitric oxide synthase that can be detected by NADPH-d histochemistry. The hippocampal formation contains a population of morphologically, neurochemically and functionally diverse nitrergic neurons.

Wistar rats of both sexes were used in this experiment. The animals were anaesthetized and perfused transcardially on 14. postnatal day and in the adulthood. Coronal sections of hippocampuses were processed for NADPH-diaphorase histochemistry.

NADPH-d positive neurons were present in the hippocampus proprius from the first week of postnatal life. However, in the dentate gyrus is a period of NADPH-d expression more delayed and positive neurons are present since the second postnatal week. NADPH-d staining on 14. postnatal day is seen to be similar as in the adulthood. Single layers of hippocampus proprius and dentate gyrus significantly differed in their staining for NADPH-d. Pyramidal neurons of CA1-3 regions and dentate granular cells were generally unstained. Light or moderately stained interneurons were scattered in all hippocampal layers, only. In the dentate gyrus, many of larger NADPH-d positive neurons were located in the hilus and at the hilar border of the granular cell layer.

We can presume that NO could play specific role in different regions of hippocampal formation and the presence of NADPH-d reactivity is changing during the postnatal development.

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Invasion of macrophages in the dorsal root ganglia of two neuropathic pain models

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Activation of macrophages in response to the peripheral nerve injury is essential for both nerve regeneration and neuropathic pain induction. Increased number of activated macrophages was described in the distal stump and dorsal root ganglia (DRG) associated with injured nerve. In this study we investigated whether intensity of macrophage invasion depends on type of neuronal injury. Invasion of ED-1+ macrophages was detected and quantified bilaterally in lumbar DRG using image analysis system 1 week after i/ chronic constriction injury (CCI) and ii/ spared nerve injury (SNI) of rat sciatic nerve.

ED-1+ cell areas considered to be macrophages were found bilaterally in DRG 1 week after both types of nerve injury. Significantly higher proportion of ED-1+ cell areas was observed in the ipsilateral than contralateral DRG in both types of neuropathic pain models. In contrast to the CCI, SNI model displayed significantly higher proportion of ED-1+ suggesting massive infiltration of DRG by activated macrophages.

Retrograde transport of signal molecules from distal nerve stump undergoing Wallerian degeneration is interrupted in SNI model of neuropathic pain. Our results suggest that signals for activation of hematogenous macrophages in DRG after SNI of rat sciatic nerve are transported by blood circulation.

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Structure and changes of the peritoneal membrane at the beginning and during peritoneal dialysis

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Introduction: The extensive area of the peritoneal membrane and quick turnover of peritoneal fluid have led to use the peritoneum as a filter to replace the kidneys function during peritoneal dialysis (PD). Our aim was to analyze the structure of the peritoneal membrane at the beginning and during peritoneal dialysis.
Material and Methods: We studied the structure of peritoneum at the beginning and during PD by means of light and electron microscopy, immunohistochemical detection of smooth muscle actin , CD 31, CD 68, and the fibrinolytic markers: PAI-1, uPA, tPA. Quantative histology was used for estimation of microvessel density and average peritoneal thickness of the submesothelium. At the time of the first insertion of the catheter for PD, small biopsies were obtained from 29 men and 13 women.
Results: The morphology of peritoneum shows a great local as well as individual variability. Specific changes similar to effects of the long-term PD, e.g.: damage of mesothelium, fibrosis of submesothelial connective tissue, vascular degeneration, the presence of myofibroblasts and macrophages, were identified even at the beginning of PD. In a case study of peritoneal dialysis with biocompatible PD fluid, an increase in submesothelial compact zone thickness but not vessel density was observed.
Conclusion: Pathological changes of peritoneal structure visible at beginning of PD probably have been caused by primary diseases (diabetes mellitus, hypertension, nephrosclerosis) resulting in renal failure.

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Intercellular interactions in malignant melanoma

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The incidence of malignant melanoma, a malignant skin tumor in the Czech Republic increased from 1970 until now 4 times (approx 19.5 per 100 000 inhabitants, 2007 SZÚ). Despite progress in cancer treatment, the prognosis of patients with malignant melanoma is poor, especially for metastatic disease, which may play a significant role in complex tumor environment.

Nodular melanoma is a prognostic poor form, here we observed in the periphery of the tumors and above the tumors, significant hyperplasia of the epithelium, there were even present in significant changes in the level of specific keratins, mainly of keratin 14 and 10. Malignant melanocytes induce expression of keratin 19, keratin 8 and epithelial-mesenchymal transition in cultured keatinocytes. In contrast, these changes do not cause healthy melanocytes. These results are comparable with the experiment when keratinocytes are exposed to the influence of neural crest stem cells from which melanocytes originate. Tumor environment does´t consist only of the tumor cell, but from tumor associated fibroblasts too. These stromal fibroblasts isolated from skin metastases of malignant melanoma can also affect the healthy keratinocytes, but only at the level of keratin 14 and the presence of epithelial mesenchymal transition. Stromal fibroblasts can´t make changes such as malignant melanocytes or tumor stromal fibroblasts from other skin tumors.

This research demonstrates to the complexity of the tumor microenvironment, where the tumor melanocytes induce stem cells like phenotype of keratinocytes and epithelial mesenchymal transition. This effect is similar to the results of neural crest stem cells from which melanocytes originate.

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Density and orientation of brain microvessels – a quantitative approach

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Microvessels in 32 samples of white matter, cortex and subcortical grey matter of human brain were detected immunohistochemically using anti-laminin antibody. Samples were cut into 20 μm thick histological sections. We determined rose of directions, level of anisotropy, preferential directions, length density of microvessels and numerical density of microvessel profiles per area unit.

No significant differences in vascular anisotropy were found between cortex, subcortical grey matter and white matter ($p=0.05$). There were no significant differences in anisotropy between cortex and subcortical grey matter ($p=0.22$) and between white matter and subcortical grey matter ($p=0.28$). Significant difference was found between white matter and cortex ($p=0.03$). Length density of microvessels was higher in subcortical grey matter (652.5 mm^{-2}) and in cortex (570.9 mm^{-2}) than in white matter (152.7 mm^{-2}). Numerical density of microvessels was higher in subcortical grey matter (3782.0 mm^{-3}) and cerebral cortex (3160.0 mm^{-3}) than in white matter (627.7 mm^{-3}).

Microvascular bed in white matter appeared to be anisotropic and direction-sensitive system while microvessels supplying cortex were closer to isotropic network.

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Extreme case of bilateral talipes equinovarus from archaeological records

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The main purpose of this study was to describe the nature of presented feet deformity, reconstruct the morphology of the feet, offer differential diagnosis and also describe the changes, possibly connected with the deformity.

An almost complete skeletal remains of an adult female (determined according to the morphological structures of the cranium) from slavic burial site Pagan field (Pohansko near Břeclav) show malformed feet bones. The age at death was estimated according to the dental abrasion as being from 39 to 44 years of age. Bones attributed to the skeleton include both pelvic bones, femora and tibiae, calcanei, right talus, all left cuneiforms, medial and lateral right cuneiforms, both cuboid bones, all metatarsal bones and some phalanges. Both fibulae are missing. Some bones of upper limb are also present.

The talus and the calcanei are reduced in size and flattened. Both tibiae had articulated with the calcanei posteriorly from the tali. Lateral sides of the cuboid bones are contracted and the articular surface for lateral cuneiform bones are displaced plantarly.

Bones of the feet and tibie present different state of deformation which are the most probably consistent with the diagnosis of bilateral talipes equinovarus. Thanks to the good preservation and the completeness of the bones of the lower limb is possible to study changes of almost whole lower limb (e.g. position of articular facets). Skeletal remains of this individual can provide the possibility to study development of the congenital deformity in an untreated individual.

Differential expression of ABC transporters (MDR1, MRP1, BCRP) in developing human embryos

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Three ABC transporters (MDR1, MRP1, BCRP), belonging to the family of multidrug resistance (MDR) proteins, play a crucial role in the protection mechanisms during embryogenesis and mediate drug resistance in cancer cells. The distribution of these transporters in the serie of human embryonal/fetal intestine, liver and kidneys of various stages of IUD by indirect two-step immunohistochemical method was investigated. The organ- and age-specific expression patterns of these transporters were depicted and compared with the expression in adult organs. The evaluation of intestine and liver samples demonstrate differences in expression pattern of ABC transporters during IUD. On the contrary, in kidneys the age-specific localization was not observed. However, the increasing positivity from the kidney surface towards deeper, more differentiated parts was observed. We also speculate that the differences in distribution of these transporters during IUD reflect their vulnerability to different drugs and toxins. Hopefully, our findings may contribute to elucidation of the role of multidrug resistance (MDR) pathways during IUD.

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Pre- and/or post-surgical administration of estradiol reduces skin flap necrosis in ovariectomised rats

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Previously, it has been well demonstrated that estrogens have protective effects against tissue ischemia. Therefore, in this investigation the influence of systematically administered estradiol-benzoate on skin flap surviving was studied in ovariectomised female Sprague-Dawley rats (n=43). Thirty-three rats underwent ovariectomy, while the other ten rats were shame ovariectomised. Three months after ovariectomy all rats were subjected to skin flap surgery with 2 x 8 cm flap dimensions located on the dorsum. Rats were randomly divided into 6 groups (2 control – treated with saline and 4 treated – treated either with 10 or 100 μg/kg of estradiol-benzoate). Treatment started either at the day of flap surgery or three days prior the surgery. All rats were sacrificed seven days after flap surgery and skin samples wre removed for macroscopic and histological examination. Our results showed that administration of estradiol-benzoate significantly decreased skin flap necrosis with the highest surviving rate in group where treatment started three days prior flap surgery. In conclusion, here observed protective effect of estradiol on skin flap surviving in ovariectomised rats could be potentially applied in plastic and reconstructive surgery in post-menopausal women. Nevertheless, further research is needed to explain the exact underlying mechanism and to find the optimal treatment protocol.

Is there any association of induced hyperhomocysteinemia in global ischemia/reperfusion injury (iri) of rat brain?

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Ischemia/reperfusion injury of the brain as a result of cardiac arrest frequently represents a leading cause of death worldwide. Ischemic preconditioning (IPC) represents an important phenomenon of adaptation of CNS, which results in increased tolerance of CNS to the lethal ischemia. The mechanisms underlying ischemic tolerance are rather complex and not yet fully understood [1]. Otherwise, hyperhomocysteinemia (hHcy) is one of the risk factor, which could have negative impact on the onset/progression of IRI [2]. There has been an explosion of research documenting the role of variety signaling molecules in IRI and IPC [1]. The MAPKs (mitogen activated protein kinases) are a signal transduction routes that couple intracellular responses to cell surface receptors. ERK protein is part of the cascade leading to survival of neurons after injury. In contrast, high levels of p38 protein are associated with degeneration and/or death in many studies using neuronal injury models [3]. The present study was designed to characterize changes in MAPK pathways and related enzymes after IPC and hHcy on IRI-associated alternations. Global forebrain ischemia was induced by 4-vessels occlusion. Rats were preconditioned by 5 min of sub-lethal ischemia and 2 days later, 15 min of lethal ischemia with reperfusion period of 1h, 3h, 24h and 72h was induced. hHCy was induced by twice a day subcutaneous injection of Hcy (0.45 μmol/g). Immunohistochemical as well as Western blot analysis identified ERK protein as well as p38 protein in injured areas. The highest level of ERK protein and the lowest protein level of p38 were detected in the reperfusion time after IPC. Converse effect was observed in the reperfusion time after induced hHcy. This suggests that adaptive mechanisms in the MAPK signal transduction machinery might have a potential role in tissues response subjected to IRI and in the phenomenon of tolerance. Our results showed that IPC as well as hHcy affects post-translational changes in the rat brain induced by ischemia.

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Key molecular regulatory factors of developmental potential of human embryo - clinical implications.

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The traditional systems using morphological criteria for assessing gamete and embryo viability have limited ability to accurately select those with best developmental potential. To overcome the still insufficient implantation and pregnancy rates following IVF in humans, more than two embryos are commonly replaced, potentially leading to high number of multiple pregnancies. The aim of our study is to contribute to the understanding of key molecular regulatory factors of developmental potential of human embryo and then, on the basis of that to reform the evaluation criteria by adding to morphometry the detection of certain molecular markers (in follicular fluid, blood or culture media) that will be proved to have a predictive value.
The evaluated population consisted of 279 infertile women and of 143 healthy fertile women that conceived spontaneously and delivered successfully at least once in their history. The mutational status of LIF and IL-11 genes and parameters of oxidative stress in the follicular fluids pool were assessed. The results were statistically evaluated by the the Fisher's 2 by 2 exact test. The LIF gene mutations don't decrease the implantation rates of assisted reproduction techniques in most patients but they interfere with the immunopathologies that are associated with endometriosis or idiopathic infertility. The levels of homocystein in the follicular fluid have a predictive value for the embryo developmental as well as implantation potential.

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Evolutionary middle ear precursor with the presence of teeth

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Spiracle is mostly known as an opening on the head surface situated behind the eyes in some sharks or basal bony fishes, but from the developmental point of view it in fact represents the first pharyngeal slit between mandibular and hyoid arches. The presence of the spiracle is generally considered a primitive character of jawed vertebrates; in early tetrapods, however, the spiracular canal was evolutionary transformed into the middle ear cavity. In the Senegal bichir, the basal extant bony fish, we identified several toothed bony plates located around a base of the spiracular canal. The development of spiracular teeth is, however, delayed when compared to other teeth and this relative late appearance was shown to correlate with differentiation of palatal dermal bones. We suggest a homology of spiracular dental plates to those of the branchial apparatus, which, however, are situated on viscerocranial elements instead. The presence of spiracular teeth in bichir might represent an ancient developmental potency to produce teeth alongside the entire oropharyngeal cavity; in this particular case, interestingly, within the middle ear precursor.

Endoderm outside the head: striking pharyngeal morphogenesis in a basal fish, bichir

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Bichirs (Polypteriformes), the most basal group of extant ray-finned fishes, possess a unique set of features. Among these, the presence of external gills is of a special interest since this larval adaptation develops so early that outer gills constitute in fact the earliest structure visible on the embryonic head. As external organs located on the head surface, the epithelium of external gills is generally considered ectodermal in origin. In the Senegal bichir, however, we found a strong contribution of pharyngeal endoderm to outer gill epithelial lining, which to our best knowledge represents the first evidence of endoderm being situated out of the head. The bichir outer gills develop exclusively from the hyoid arch and our developmental analyses revealed that a heterochronic shift in the pharyngeal pouch differentiation is responsible for the very early morphogenesis of outer gill structures. We suggest that the striking contribution of the pharyngeal endoderm to external gill morphogenesis can be understood in evolutionary terms of embryonic adaptation and we will discuss **possible alternative hypotheses.**

Isolation and characterization of neural crest stem cells from adult human hair follicles

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Neural crest (NC) is a transient embryonic tissue, whose cells are motile and multipotent until they reach their destination and differentiate according to microenvironmental cues into variety of cell types. However, a subpopulation of these cells remains multipotent. They were found, among other locations, in a bulge of adult murine whisker follicle and were designated epidermal neural crest stem cells (EPI-NCSCs). The aim of this work is to ascertain whether the EPI-NCSCs could be isolated from human hair follicles as well. Due to their exceptional properties, they could represent potential candidate for stem cell therapy. The presented work focuses on the isolation and characterization of EPI-NCSCs from human skin. We obtained population of cells that expressed markers of NC, NC progeny and general stem cell markers. After prolonged cultivation, subpopulation of cells spontaneously differentiated into some of NC derivatives, i.e. neurons, smooth muscle cells and Schwann cell progenitors. Targeted differentiation with neuregulin-1 highly increased the number of Schwann cells in culture. Human EPI-NCSCs were also able to grow under non-adherent conditions and form 3-dimensional spheres. Microarray analysis was performed and gene profile of human EPI-NCSCs was compared with the list of key genes of murine EPI-NCSCs, and the list of genes upregulated in newly induced NC cells. This revealed 94% and 88% similarity, respectively. All the presented results strongly support the NCSCs identity and multipotency of isolated human cells. These cells thus could be a good candidate for stem cell therapy, especially because of the easy accessibility of donor tissue.

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FGF signaling mediates adaptive response to pressure overload in the embryonic myocardium

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Fibroblast growth factors (FGFs) and their receptors play an important role during embryonic induction and patterning, as well as in modulating proliferative and hypertrophic growth in fetal and adult organs. Exogenous FGF2 (also known as basic fibroblast growth factor) was found to be able to induce proliferation of chick embryonic and fetal cardiac myocytes in vivo. Pressure loading resulting in myocyte stretch is a powerful physiological stimulus for embryonic myocyte proliferation. Here we tested whether FGF2 signaling is involved in transmission of mechanical stretch to myocyte growth in vivo.
Pressure overload was induced by constriction of the outflow tract in Stage 21 chick embryos. Hearts were sampled at 24 and 48 h intervals, and anti-bromodeoxyuridine labeling was used to assess proliferation. Western blotting and RT-PCR to measure FGF2 protein and mRNA levels was performed on extracts from ventricles sampled at 48 h interval. Blood was collected at 48 h as well to assess the amount of FGF2 in serum by ELISA.
Proliferation was increased significantly at the 48 h sampling interval. Neither Western blotting, nor immunohistochemistry performed on sister paraffin sections revealed any changes in the amount of myocardial FGF2. However, ELISA showed a significant increase of FGF2 in the serum. Increased amount of FGF2 mRNA was confirmed by real time PCR.
We thus conclude that FGF2 synthesis is increased in embryonic ventricular cardiomyocytes in response to increased stretch due to pressure overload. However, increased stretch causes its release into serum, making it act in endocrine, rather then usual paracrine manner.

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Morphological changes of large conduit pulmonary arteries in hypoxic conditions

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The environment with low oxygen concentration causes development of hypoxic pulmonary hypertension (HPH) as well as remodelling resistive pulmonary arteries. This is mediated by products of the mast cells (MC). Remodelling processes can be modified and slowed down by MC stabilization with sodium cromoglycate (SC). Having tried to affect morphologic image of HPH, we encountered some interesting changes of large conduit arteries (LCA) with the diameter over 100 μm. These vessels do not affect the final blood pressure in the pulmonary vascular bed; however, their structure changes substantially under hypoxia.

Total 64 Wistar-Han rat males were grouped by 8 animals in 8 groups and exposed to 4-day (H4) or 21-day (H21) normobaric hypoxia (10 % of oxygen) accompanied with variant timing of intraperitoneal administration of SC (first 4 experimental days in groups H4K and H21KZ; last 4 experimental days in H21KK group). We assessed the average thickness of tunica media (TM) of LCA using the image analyser software NIS elements 3.0 AR.

Significant increase in TM thickness was evidenced in groups H4 and H4K vs normoxic controls. Difference between both hypoxic groups was not significant.

In groups H21, H21KZ, and H21KK, we observed continuing significant increase in TM thickness vs normoxic controls; no significant difference between these hypoxic groups was revealed.

The environment with low oxygen concentration causes increase of TM thickness of LCA vs normoxic controls. Obviously, such remodelation changes are not mediated mostly with MC products since they have occurred in spite of efficient stabilization of MC granules with SC. We hypothesise some mitogenic hypoxia-inducible factors could be in the background of such changes.

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Morphological manifestations of heavy metals toxicity

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Distribution of heavy metals and subsequently induced structural changes in some organs of female mice and mouse foetuses after peroral administration of moderate doses of lead, mercury, and cadmium (0.03 mg of metal per mouse and day) was studied in this study.

Heavy metals (HM) were administered to female mice on days 9 – 20 of pregnancy. There after animals were euthanised by cervical dislocation. Samples of organs of female mice and foetuses were subsequently collected and processed by standard protocol for light and electron microscopy. Histochemical reaction based on metal conversion into appropriate sulphide that conjugates with silver was used for detection of HM.

By the light microscopy, deposits of HM, were found: in the placenta in labyrinthic and spongiotrophoblast region, namely in erythrocytes and trophoblast cells; in the liver of pregnant mice at the periphery of hepatic lobules in hepatocytes and Kupffer cells; in the liver of foetuses in hepatocytes, endothelial cells of capillaries and erythrocytes; in the renal cortex of females and their foetuses, namely in the epithelial lining of proximal and distal tubules.

By the electron microscopy, deposits of the reaction product were identified in compartments of cells reacting in the light microscope as follows: in the nuclei and nucleoli, lysosomes and ribosomes of rough endoplasmic reticulum in the cells of the placenta; in the nuclei and nucleoli, lysosomes, ribosomes of rough endoplasmic reticulum (RER) and mitochondria of hepatocytes and Kupffer cells in the liver of females and their foetuses; in the nuclei and nucleoli, lysosomes, ribosomes of RER and mitochondria in the kidneys of females and their foetuses.

Structural changes caused by HM were manifested: in the trophoblast cells of placenta as a desintegration of nuclear envelope, decrease of nucleoplasm electron dension with the clumps of heterochromatin, obliteration of cisternae of RER and separation of ribosomes; in the hepatocytes by ruptures of nuclear envelope, obliteration of cisternae of RER, separation of ribosomes, edematous mitochondria and destruction of lysosomes. Vacuolization of the cell cytoplasm was also a frequent phenomenon. Damage of hepatocytes was more expressive in the fetal than in the maternal cells; in the kidneys of foetuses already at the light microscopic level with: dilatation of uriniferous tubules, decrease of height of their epithelia and defects of basement membrane, decreased size of glomeruli and enlargement of their urinary spaces, intersticial connective tissue extravascular clumps of blood cells; at the electron microscopic level: sparse of nucleoplasm with the clumps of heterochromatin in the cell nucleus, obliteration of cisternae of RER and separation of ribosomes, destruction of lysosomes and vacuolization of cell cytoplasm seen in the kidneys of females and their foetuses.

Observed structural changes show that some cells are destroyed by necrosis. High accumulation of HM in organs of pregnant mice and their foetuses was verified in our study. It is concluded that placental barrier does not provide against HM penetration into the foetal organism. Higher accumulation of HM in foetal than in mother organs can be explained by intense metabolism of developing embryonic cells.

Inferior vena cava duplex. A Case Report

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On dissection of an adult 70 years old male cadaver at Anatomy Department, Charles University, Faculty of Medicine in Pilsen presence of double inferior vena cava (further VCID) accompanied by some other vascular anomalies was found .
The purpose of this study was to understand the actual anatomical nature of the congenital malformations,to describe their developmental view, to correlate with the preexisting works of other authors and to analyze their clinical significance.
Detailed dissection of both abdominal and pelvic area was done.

In the abdomen both right and left inferior vena cava were found parallel to vertebral column in range of L1 – L5. Left renal vein was the vein connecting both sides. Second larger connecting vein was found below the aortic bifurcation at L5 level, forming the venous network. Development of the connecting veins can be explained by developmental prenatal anomalies.

Described pelvic connecting vein of both vena cava inferior has not been published in literature; it is unusual founding which can be of clinical significance in vascular surgery.

Morphology of an Experimental Abdominal Aortic Aneurysm Model

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Introduction: The abdominal aortic aneurysm (AAA) is a serious disease because of its incidence and mortality.
Aim: In our study we assessed the impact of statin administration on histopathological changes in an porcine AAA model.
Material, methods: AAA was created by an infusion of elastase and application of a plastic cuff. The group 1 (n=13) – animals received no postoperative treatment. In group 2 (n=14) pigs were treated with atorvastatin (20 mg per day for 4 weeks). Using histological staining and immunohistochemistry, we applied stereology for quantitative analysis of the area fraction of elastin collagen, actin, desmin and vimentin, the length density of elastin, the density of microvessel profiles within the aortic wall.
Results: In group 1, we found lower fraction of elastin, actin and a higher fraction of vimentin, the intima-media thickness was higher and the density of vasa vasorum was lower when compared with group 2.
Conclusion: After administration of statins, the AAA wall contained more elastin and actin-positive cells, both essential for the proper mechanical function of the tunica media. Without statins, more cells were positive for vimentin, a marker of rather synthetic phenotype of vascular smooth muscle, the wall was thicker and the thickening has not been accompanied by formation of new vasa vasorum. We conclude that progression of experimental AAA was mitigated by administration of atorvastatin.

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Feasibility of fractal analysis for evaluation of rat lung tissue in experimental model of sepsis treated by gentamicin

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Bioavailability of aminoglycoside antibiotics may be critical for treatment of septic patients. That is why we tried to assess the development of morphological changes in acute phase of sepsis in experimental rat model in order to prove validity of this model from morphological (besides the classical histopathology) point of view. For description of complex and irregular structures, there is possible to use methods of fractal analysis (e.g., box-counting), based on self-similarity of structures.

Lung tissue (paraffin sections, haematoxylin-eosin) of control group rats (n=6) was compared with (1) rats administered gentamicin (n=6), (2) rats administered LPS/IL-2 (endotoxinaemia was induced by administration of Pseudomonas aeruginosa lipopolysaccharide and recombined mouse interleukin-2) (n=6), and (3) rats given LPS/IL-2 followed by gentamicin (n=6). Light microscope Olympus BH-2 with Olympus MicroPhoto v2.2 software was used for microphotography (10 pictures per each slide). Pictures in file format bmp were processed (10 box-counts with random starting point per each picture) with HarFA - Harmonic and Fractal Image Analyzer, Demo version 5.4. Comparisons among groups were made via one-way analysis of variance (ANOVA) and appropriate post-hoc tests.

Rat lung tissue has fractal properties. Fractal dimensions (D) oscillated between 1.5 and 1.8. We found differences among groups, but none of them were statistically significant ($p < 0.05$). There may be many factors influencing accuracy of fractal analysis, we consider the adjustment of sampling pattern to be the most important. We plan to go on with fractal analysis, eventually using some other methods to study the complexity of diseased tissue.

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Endoneurial extracellular matrix influences regeneration and maturation of motor nerve axons – a model of acellular nerve graft

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The growth and maturation of regenerating axons are influenced by Schwann cells and endoneurial extracellular matrix. The aim of our experiments was to study the role of endoneurial extracellular matrix in regeneration and maturation of motor axons. Morphometric features (number and diameter of myelinated axons, thickness of myelin sheath) of motor nerve axons regenerated into the microenvironment of acellular grafts prepared from the cutaneous and muscular branches of the rat femoral nerve were evaluated. In addition, motoneurons regenerating their axons through different acellular grafts were counted.
No differences were found in the numbers of regenerated axons and related motoneurons through the motor and cutaneous nerve grafts 1 month after operation. Two months from grafting, however, the numbers of motoneurons and regenerated axons were increased significantly in the motor grafts while these were decreased after regeneration through the cutaneous grafts compared with 1 month. Axons' diameter and thickness of their myelin sheaths were similar in the cutaneous grafts 1 and 2 months after grafting. In comparison to 1 month, axons had larger diameter and thicker myelin in the motor than cutaneous nerve grafts 2 months from their application. Results of morphometric analysis indicate more beneficial extracellular conditions for regeneration and maturation of myelinated motor nerve axons in the acellular motor than cutaneous nerve graft. Generally, the results revealed that the endoneurial extracellular matrix of motor fibers has a positive effect on regeneration and maturation of motor nerve axons after lesion.

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Vascular morphogenesis in vertebrates and invertebrates – models of vascular lumen formation

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The blood vascular system in vertebrates consists of endothelium-lined tubes that carry oxygen and nutrients to tissues and take away carbon dioxide and waste products. Blood vessels are of different diameter and they differ in composition of their walls but all originate as simple endothelial tubular structures. Opinions on how the lumen of these tubes is created can be traditionally summarized into two competing theories. Intracellular theory claims that vascular lumens arise initially inside endothelial cells via fusion of vesicles (vacuoles). On the other hand, the intercellular theory is based on the idea that lumens are formed in the intercellular space and this process is associated with junctional rearrangement and cell shape change. Although these theories are not necessarily mutually exclusive, a general consensus has not been reached yet. Experimental support for these theories often comes from different *in vitro* and *in vivo* models which could contribute to conflicting conclusions.

Studies on mouse aorta morphogenesis have revealed that the vascular lumen formation consists of the following steps: 1) angioblasts assemble into solid cords held together by intercellular junctions, 2) cells establish apicobasal polarity and intercellular junctions move to the peripheral location, 3) luminal membranes deadhere as a consequence of electrostatic cell surface repulsion, 4) nascent lumen expands as the cells elongate and flatten, which is mediated by an actomyosin complex at the luminal plasma membrane.

In the last years we have observed acceleration of research on vascular morphogenesis and we might expect further interesting findings on molecular mechanisms behind this process in the near future.

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The use of digital virtual microscopy system for creating digital histology slides database as a tool for e-learning

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After completing syllabus of general histology, microscopical anatomy and embryology, students have to understand the structure and composition of tissues and organs together with their functional properties. As an integral part of the study process students learn the structure of cells, tissues and organs practically at the level of the light microscope, which is a prerequisite for understanding of pathologically altered structures as observed in histopathology slides during pathology lessons.
For the histology exam there are more then 100 slides, a student has to identify and describe. However, for the self-study and for revising the slides it is inconvenient when students are limited by working hours and location of the slide room, especially during the exam period. As a method of e-learning there is now a new tool available to create a virtual histology slides database that students can access (preferably from home PC). In our institute we began to utilize a digital virtual microscopy system dotSlide (Olympus) for creating a database of histology slides. This system is not only useful for teaching but it has many potential applications elsewhere. Virtualization provides a procedure for creating databases with remote access. These can be shared during teleconsultations or teleconferences.
We started to collect virtual database from slides required for histology exam. The images in a special format are stored on the server. These preparations can be accessed via either a web browser or using a special programme called OlyVia that can be downloaded from the Olympus company web site. Annotation of the structures is possible and we plan to expand the database to add more staining variants to different tissue sections and to include some special slides (eg. embryology slides, special techniques etc.).

Prevalence and variability of the inferior right hepatic vein

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Objective: This study aims to present the prevalence, variations, and age distribution of the accessory and inferior right hepatic veins (IRHV) on the abdominal MR scans.
Methods: The routine abdominal MR images of 345 female pacients were reviewed for presence of IRHV. After exclusion of the patients with improper opacificated hepatic veins or with right hepatic lobe lessions, 305 scans were evaluated (range 0–86 years, mean age 42,3 years). Forty (range 0-29 years, mean age 18,3 years) men scans served as a control group. Presence, positions and diameters of the hepatic veins were noted. On other 127 patients (61 men, 66 women) hepatic veins patterns and sex distribution was examined.
Results: Of the 305 females, fifty-six (18,36%) has one or more large IRHV´s (≥ 0,4 mm). Their highest prevalence was in group of young women (18–29 years, 25,64%), lowest was in the range of 40-49 years (13,51%). At a group of young men (18-29 years) large IRHV was found in 33,33%. Branching of the right lobe hepatic veins is very variable and description highly depends on observer and imaging technique used. Finding of any possible patterns was rather difficult.
Conclusions: Magnetic resonance imaging proved to be very reliable method for this type of study. Preliminary results show tendency of a higher prevalence of any accessory hepatic veins at a women and younger persons regardless of sexes.
Destription of IRHV patterns was not beneficial.

Variations of the seventh cervical vertebra and their clinical significance

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This paper is part of a larger study in which we study the variations of vertebral column. The anatomical variations of the cervical vertebrae include various types of changes, for example cervical ribs, non fusion of the halves posterior arch of the atlas, the presence of a third occipital condyle, etc. In this section, we focused on the vertebra prominens and their clinical significance.

A cadavers were preserved by means of the routine embalming techniques following the completion of dissection were used for bone maceration. The soft tissues were removed and the cervical vertebrae were studied for variations.

The seventh cervical vertebrae were examined to determine the morphometric variations: the cervical canal anteroposterior nad transverse diameters, vertebral body width, height. The second purpose of our study was to evaluate the variations of the foramen transversarium. This foramen sometimes has the same dimensions as the foramina of the other cervical vertebrae, but it can also be smaller or absent. We also studied the variations of the vertebral processes. We observed the changes in the shape of the processus spinosus and its length, the shape and the position of the processus transversarius and the position of the processus articularis.

The implications of these variations and abmornalies in the anatomy of this region should be noted by neurologist, neurosurgeon, orthopaedics. Clinicians themselves have called for a better understanding of variations in the human spinal column in their quest for enhanced treatments for a range of spinal problems.

Relevancy of Skin Cancer in vitro Modelling. (Lesson from Complexity of Epithelial Mesenchymal Interactions in Development and Cutaneous Pathology)

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Skin cancer is the most frequent type of cancer in developed countries, but it represents due to its high incidence a serious burden worldwide. Based on developmental similarities histogenetic relationship of different skin tumors to normal skin structures was proposed. We have isolated cancer associated fibroblasts (CAF) from different types of skin melanoma, non-melanoma skin cancer and mesenchymal skin tumors in order to investigate the function of such stromal cells in tumor development and spreading. We have elucidated multiple differences between normal dermal fibroblasts and CAF based on gene expression profiles. Certain features of their bioactivity seem to be relevant for cancer stem cells maintenance. For routine screening of CAF bioactivity we have employed methods of direct and indirect cocultivation with normal kertinocytes. This exceedingly simple procedure appoints to enormous plasticity of normal human keratinocyte phenotype and its profound dependence on environmental factors. Thus we can demonstrate significant degree of agreement between in vivo tumors (e.g. basal cell carcinoma, dermatofibroma, melanoma) and their in vitro models. These in vitro models could be also relevant in future cancer treatment development. Finally the lecture emphasizes the importance the appropriate complexity preservation in such efforts.

Failure of the TGF-β pathway leads to multiple self-healing squamous carcinomas

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Following our interest in causes of susceptibility to skin cancer, including epidermolysis bullosa, we studied a rare cancer condition called multiple self-healing squamous epithelioma, or Ferguson-Smith disease (MSSE/FSD), which causes many keratoacanthoma-like skin cancers to develop in sun-exposed sites. The disorder was reported over 70 years ago in Scotland. With collaborators in Dundee, California and Singapore, and clinicians from Scotland to New Zealand, we discovered that the genetic defects causing MSSE are mutations in the TGFβ-R1 receptor. *TGFBR1* mutations were found in 3 unrelated MSSE families using high-throughput sequencing with exon array capture over an extended 24 Mb region on chromosome 9q. Sanger sequencing then determined *TGFBR1* mutations in 18 MSSE/FSD families altogether, confirming *TGFBR1* as the causative gene for MSSE/FSD. TGF□ signaling pathway defects have often been reported in various cancers, but this one is unusual because the skin tumours regress spontaneously. Mutations in *TGFBR1* have previously been reported in Loeys-Dietz Marfan-related syndrome, a developmental abnormality with blood vessel dysmorphology but no cancers. The reason for one gene causing two diseases seems to be that the MSSE mutations cause loss of function of *TGFBR1*, whilst Marfan-related disorders with gain-of-function mutations lead to developmental defects, but no increased cancer incidence.

Canalis fibularis morphology

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Introduction: Lesions of nervus fibularis communis is one of the most common neurological affections (15% of all adult mononeuropathies). Canalis fibularis is one possible site of the entrapment syndrome to occur but its morphology is neglected both in anatomical books and articles in the recent periodicals.
Material and methods: Sixty limbs from cadaverous material (collections of Department of Anatomy, Charles University in Prague) were dissected and structures were measured from reference point at caput fibulae.
Results: Nervus fibularis communis alwyas passed between m. fibularis longus and fibula. The canal entry was always formed with tough connective tissue arch between the origins of musculus fibularis longus and musculus soleus. Proximal border of canalis fibularis entry was 4–9 mm a distal 7–27 mm from reference point. The nerve ranched within the canal in 90 % of cases.
Conclusion: The canalis fibularis syndrome is not a rare neurological unit due to both traumatological and anatomical causes of the nerve entrapment.

The possibilities of improving the teaching process of Anatomy in study program of Dental Medicine

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Anatomical knowledge is fundamentally important to the study and practice of medicine. Despite the offer of wide range of high quality anatomical texts and atlases on the medical current market, there is no publication with detailed, colorful figurative photographs of the head areas that can serve as an visual aid for practical and also theoretical teaching of the Anatomy for Dentistry and General Medicine students. Following new trends in methods of anatomy teaching it is necessary to pay more attention to quality, depth and effectiveness of medical education. There is a need to prepare the interesting publication in order to point out the connection between this important morphological discipline and clinical medicine.

The anatomy of the head is mainly of interest to students early in their studies. Clinically, it is mostly relevant to surgeons who are performing interventions and repairing the maxillofacial region, skull base and the orbit. This work maps the significant areas of the head that are not normally prepared for practical exercises in anatomy because of the time and difficulties with the preparation. Thus the aim of study was focused to present predominantly the detailed photodocumentation of the orbit, ear, structures inside the infratemporal and pterygopalatine fossae, parasellar region and hypophysial fossa. Also the areas of upper and lower jaw teeth are depicted. The important chapter is dedicated to the head parasympathetic ganglia to specify their sensory, sympathetic and parasympathetic roots.

The detailed photodocumentation enriched by schemes provides a view of structures until now only poorly documented. We believe that this "Topographical Anatomy“ and above mentioned details will be beneficial for the further medical education.

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Mirtazapine embryotoxicity evaluation by Chick Embryotoxicity Screening Test

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Mirtazapine is a new antidepresant with low acute toxicity. However, there is only little information about its embryotoxicity. Aim of our study was to contribute to the knowledge on possible risks. For embryotoxicity testing we used an alternative method - Chick embryotoxicity screening test (CHEST) . Fertilized eggs of outbred Grey Leghorn stock (AVČR farm Koleč) were incubated at 37,5 ± 0,2° C and relative humidity 55-65%. Access to the embryos through a opening of eggshell was performed on embryonic day 2 for CHEST I or 4 for CHEST II. Mirtazapine (Sigma) was diluted in DMSO (Sigma) and was administered subgerminally for CHEST I and intraamniotically for CHEST II. Embryos were incubated till 9ED, when they were weighed and examined under stereomicroscope. Summing the proportions of dead and malformed embryos, the beginning of the embryotoxicity dose range was estimated. The results were approximated to the doses in mammals. The lower administered dose corresponded to therapeutic dose in human (40mg/day). In doses, that were comparable with terapeutic, no diference from control group were found as well as in doses 10x time higher. Our results are in agreement with experimental findings on rats and rabbits reported by producer, in which the risk of resorption and decreased pup weight and viability at term were demonstrated at doses 20 times higher than therapeutical.

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Intersexual differences some anthropometric parameters in the group of children in the 3 rd month of age.

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The nutrition at the begining of life has important influence on the individual health during the next years. The mother's milk is unique and not reccurent by specific composition, content of the safety substance and the growth factors. Grant not only protect against infection, but also stimulates the development and the increase of the baby's organs.The monitoring of the anthropomethric parametrs of the babies since the born permit evaluating of the body growth like the main indicator of the health condition, nourishment and socio-economic state of the individual and the groups of the populations.

We have examined 216 babies (111 boys and 105 girls) regularly in tree month intervals, from the birth till the twelfth month of their life, using the Martin-Saller's examination methodology. The children were born in University Hospital in Martin. The groups of children, according to the sex and duration of the breastfeeding, in the subgroups were separated. The values of the measures were calculated to arithmetic value and standard deviation. In statistical analysis the t- test was used.

The boys in our groups were heavier and higher than the girls. The value of the abdominal circumference, chest circumference and the head circumference were bigger in the boys in all subgroups, differences were not statistically significant.

The aim of this study was to examin the group of children, which are three month old and to evaluate intersexual differences between the somatic parameters of this children´s group. The literary sources state, that the boys usually have higher values of the anthropometric parameters. The results in our group of the 3 rd month children confirm this study.

A role of CD163 and iNOS in development of acute respiratory distress syndrome: A case report

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Adult Respiratory Distress Syndrome results from a variety of different initial insults, including trauma, sepsis, pneumonia and aspiration, and represents a severe form of acute lung injury. This condition is characterized by diffuse pulmonary microvascular injury. The purpose of this study was to evaluate the histopathological alterations and to determine the role of iNOS and macrophages development of this injury. The necrotic lung sample of patient suffering from car accident with developed ARDS were harvested immediately fixated in 4% paraformaldehyde and embedded in Paraplast wax, sectioned (4-5 μm) and stained with H&E, and by immunohistochemical means using anti-CD163 and anti-iNOS (DB Biotech) antibodies for detection of macrophages and iNOS cells population. Expresion of CD163 was observed mainly in the cells of monocyte-macrophages system, primarily in macrophages. Majority of these macrophages were located in the lumen of alveoli possible as a result of chemotaxis. Smaller populations of macrophages were located in interalveolar septa. Positivity iNOS cells were mainly alveolar macrophages, but some positivity was also present in some smooth muscle cells of lungs blood vessels. Immunoreactivity of iNOS was detected in leucocytes and possibly also in pneumocytes type II. Our findings indicate that monocytes-macrophages system plays an important role in development of ARDS.

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Glutamine effect in development of lung damage after intestinal ischemia-reperfusion injury.

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Intestinal ischemia-reperfusion (IRI) may induce lung damage which can result into development of acute respiratory distress syndrome. Aim of this project is to determinate the effect of glutamine pretreatment on development of lung damage after IRI of jejunum. Wistar rats (n=36) were divided into experimental groups: In first group (Gln+I/R, n=15) glutamine pretreatment (i.v. Dipeptiven con inf, 0,75 g/1kg) was perform prior to ischemia insult of superior mesenteric artery (60 min) followed by reperfusion periods (R1,R4, R24). In I/R group (I/R, n=15) only ichemia (60 min) and reperfusion (R1,R4, R24) was performed. In control group (n=6) no ischemic insult was performed. After reperfusion sample of lung parenchyma was taken for histological (H&E) and immunohistochemical (anti-PCNA, anti-CD163 antibodies) study. Our results pointed out on significant decrease of interalveolar septum thickness in glutamine pretreatment groups. Thickness in glutamine pretreatment group was comparable with control group. Also decreased population of macrophages was decreased in Gln+I/R group in comparison with ischemic/reperfusion group. Highest proliferation rate was in I/R groups, which may be associated with highest damage of lung parenchyma. In Gln+I/R groups proliferation rate was decreased in comparison with I/R group. Our results indicated that glutamine may have a positive effect on decreasing damage, cellular infiltration, macrophages population in lung parenchyma induced by IRI.

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Arterial system of poultry, visualisation and variations

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In poultry distinct differences in each organ system are observed. In case of cardiovasular system these differences include arterial and venous ramification, differences in vena cava, and by existence of kidney portal system. Aim of our work was detailed observation of arterial system in poultry. We used domestic poultry (*Gallus domesticus,*n=6,♀) and visualization was determited by corrosing cast (Duracryl Dental). We observate branches of *aorta descendens*, which in poultry crosses between lungs, kidneys and *synsacrum* bone, because body cavity in poultry is not intergrated. Branches of *aorta descendens* are divided by segments of vascularisation on *aa.intersegmentales* (localized bilaterally), bilateral and unilateral visceral branches for digestive tract and organs. As well as in other animals first branches of *aorta descendens* are *a.celiaca* and *a.mesenterica cranialis.* In case of *a.celiaca* we observed branches for stomach, spleen and liver and these branches differs in its thickness.This observation was not detected in case of autopsy, because of internal organs, which covered arteries. Variations of *aa.renales craniales* were observed, in this case arteries were individual branches of *aorta descendens.* Observation of others branches of *aorta descendens* showed differences in localisation as well as in size. Increased thickness of arteries was observed in *a.iliaca externa*, which in poultry is localised between cranial and medial segment of kidneys and *a.ischiadica*, which is situated in caudal segment of kidney. We also observed differences in aa. *intersegmentales synsacrales.*

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The effect of cadmium on Sertoli cells and germ cells communication

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Electron-microscopically was studied the effect of cadmiun on the Sertoli cells contact with the developing germ cells in the rabbit testes. Cadmium was applied during four weeks per os. In experiment we used adult rabbits (n=10) in two groups. In experimental group cadmium (n=5, 50 g/l). After experiment samples of testis were used for electron microscopic study. Observations presented here demonstrate that cadmium damage the Sertoli cell communications with the developing germ cells, namely the spermatocytes. Cell communications were preserved in some cells, but contact of cytoplasmatic membrane between Sertoli cells and developing germ cells, except specialised cells communication was impaired. In these places the cytoplasmatic membrane of both cell types was extensively dilateted, thereby vacuolar spaces were present between these cells. Integrity of cytoplasmatic membrane in both cell types was not damaged. We suppose that cadmium impact on Sertoli cell communication is performed by its effect on actin filaments in places where connection with germ cells occur. We propose, that cadmium treatment would affect the organization of microfilaments related with ectoplasmic specializations.

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Anatomical variabilities of pudendal nerve.

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Basis: An objective of our work was to clarify variations in pudendal nerve formation, as well as their possible impact on the clinical picture.
Set: *B*ilateral pudendal nerve course and formation was studied on 20 adult cadavers. Anterior approach was used in 15 of them; both posterior and anterior approaches were used in 5 of them.
Results: In the prefixed type of lumbosacral plexus: S_1+S_2 roots were dominant in its formation in 8 cases. In the postfixed type: S_3 and S_4 roots were dominant in 3 cases, S_2 and S_3 roots in other cases. In 8 cases pudendal nerve run below sacrospinous ligament (in vicinity of ischial spine) and was pressed by its inferior part. Some other observations include: the sacrospinous ligament calcifications (4 cases), narrowing of the space between the sacrospinous ligaments (3 cases), and division of pudendal nerve before entering the pudendal canal (15 cases).
Conclusion: Our study was aimed on description of some anatomical variations in the pudendal nerve formation, depending on the type of the lumbosacral plexus.
Key words: pudendal nerve, sacral plexus, sacrospinous ligament, sacrotuberous ligament

Is the neurodegenerative process in the striatum of rats transgenic for Huntington's disease similar to that in HD patients?

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The rats transgenic for Huntington's disease (tgHD rats - von Hörsten et al, 2003) represent a new animal model for HD, similar to the late-onset form of the human disease. Surprisingly, except two marginal studies, exact morphological study of the development of striatal neurodegenerative process in these rats has not been performed yet.

In our study, we compare morphological changes characteristic for the progression of degenerative process within the striatum of 2, 6, 12, 18 and 22-24 months old homozygous tgHD rats and their age-matched wild-type littermates. Three representative cases of HD brains were used for comparison with our experimental data. The immunofluorescent double-staining method (using the array of antibodies) and quantitative image analysis were used for the assessment.

In summary, the degenerative process in tgHD rat striatum develops very slowly and the typical morphological signs, i.e. neuronal degeneration with concomitant reactive gliosis, appear at 12-month-old rats at the earliest. Unlike in human, the degeneration of striatal neurons is only scatter and very slow. However, we have first described gradual decrease in size of neuronal bodies and nuclei (with maintenance of nucleo-cytoplasmic rate) in tgHD rats which process we have also found in HD autopsies. Furthermore, we have confirmed the important participation of age-related changes (in wild-type rats) in a reduction of grey matter.

In conclusion, morphological changes in the striatum of tgHD rats correspond essentially to the pathology in brains of HD patients, however, in diminished form principally due to very slow degeneration of striatal neurons which induces only mild reactive astrogliosis.

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HSP and antioxidant enzymes participate in acquisition of delayed pharmacologic preconditioning in the spinal cord

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The present work investigated whether application of pharmacologic preconditioning prior to lethal ischemia can induce delayed tolerance of neurons in lumbar spinal cord of rabbits and improve neurological status of hind limbs. Rabbits were preconditioned with noradrenaline and bradykinin 48 h prior to 20 min of ischemia followed by 24 and 48 h of reperfusion. The activity of SOD and catalase was measured, and HSP immunohistochemistry was evaluated. Neurological status was assessed by Tarlov scoring system. Increased total SOD activity after noradrenaline preconditioning in the ventral horns (118%-24 h of reperfusion and 111%-48 h of reperfusion) was caused mainly by increased CuZn-SOD activity (340%-24 h of reperfusion and 290%-48 h of reperfusion). Total SOD activity after bradykinin preconditioning was moderately increased (102%-24 h of reperfusion and 108%-48 h of reperfusion) and was caused by increase of CuZn-SOD. In both groups the activity of Mn-SOD showed appropriate decrease. Catalase activity in the ventral horn neurons was highest in the group of rabbits with bradykinin preconditioning and 48 h of reperfusion (118%). No animal of these preconditioned groups was paraplegic. Immunohistochemic evaluation showed in the bradykinin preconditioned groups marked HSP positive cytoplasm and nuclei in the ventral horns. In the noradrenaline groups the positivity of neurons was less pronounced and in the pure ischemic group dead neurons were not HSP positive. These results indicate that endogenous antioxidant enzymes and HSP are involved in the mechanism of pharmacologic induced tolerance acquisition in the rabbit spinal cord.

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Nonkeratinizing skin cells and their pathological lesions

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The epidermis is composed of keratinocytes and other less numerous cell types not participating in continuous replacement - melanocytes, Langerhans cells and Merkel cells. A rare type of skin cancer can rise from Merkel cells, Merkel cell carcinoma, the most aggressive skin cancer. Morphological and biological similarities between the Merkel cells and Merkel cell carcinoma on one side and some differences between them on the other side, causing problem in establishing the correct diagnosis led the authors to recapitulate the latest knowledge of Merkel cell carcinoma as well as Merkel cells themselves. It was revealed, the problem of origin of Merkel cells is still not resolved. It is proved by many works bringing as evidence of neural origin as the epidermal origin of Merkel cells. The authors summarize the results of a study of this issue over the past ten years. It can be concluded new informations may clarify the biological properties and pathology of Merkel cells.

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MAO-A and MAO-B in rat female genital organs during early period of pregnancy

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Monoamine oxidases (MAO) are enzymes located in the mitochondrial outer membrane. They exist as two forms, MAO-A and MAO-B, which are different gene products and have different substrate specificities. The objective of our study was to determine the localization of MAO in rat female gonads during preimplantation period of pregnancy.
Pregnant rat females were killed on the first, on the third, and on the fifth day of pregnancy and animals were transcardially perfused with PBS. Ovaries, oviducts and uteri were immediately removed and embedded in fixative solutions. They served for the determination of MAO localization employing the immunohistochemical methods.
MAO-A activity in ovary was visible in corpora lutea and in interstitial gland cells, while MAO-B was detected predominantly in blood vessels. Both MAO enzymes were seen in the smooth muscle fibers of the ovarian hilum. However, the presence of MAO enzymes was not detected in follicles at any stage of their development. In oviduct and uterus both MAO enzymes were visible in the similar places, it means in smooth muscle fibers, in mast cells and in blood vessels. To our knowledge this is the first paper describing detection of both MAO-A and MAO-B enzymes in female genital organs employing mimunohistochemistry.

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Dorsal hand arterial variation

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Introduction: Arteria radialis gives off arteriae metacarpes dorsales for the dorsum of hand which are both used as pedicle vessels in plastic surgery for island cutaneous flap. Before it merges into the musculus interosseus dorsalis primus, a thick artery can branch from it running superficially across the muscle into the palm to anastomose with the arcus palmaris superficialis. Information on this vessel is very rare both in anatomical books and articles in the recent periodicals.
Material and methods: Ninethy limbs from cadaverous material (collections of Department of Anatomy, Charles University in Prague) were dissected and fifty volunteers were examined by ultrasound.
Results: The incidence of the variation was 17.8 % in dissected limbs and 10.0 % in ultrasound examination. The diameter of the artery was 0.7-1.5 mm. The vessels below 0.5 mm-diameter could be examined with difficulties and were in accordance with their minimal clinical relevance excluded from total numbers.
Conclusion: This artery seems to be a good alternative (if present) for other arteriae metacarpales dorsales in thumb reconstructions with flap plastics.

Tissue chimaerism following bone marrow cell transplantation

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Bone marrow (BM) cell transplantation represents a life-saving treatment for syndromes involving BM failure and thalassemia. To trace distribution of transplanted cells in the recipient organism, we utilized transgenic eGFP mice as donors whose cells were tagged to endogenous vectors.
BM ablation in recipient C57Bl6/J mice was induced by whole body lethal irradiation 9.5 Gy that severely damages haemopoiesis. Irradiated splenectomized or non-splenectomized mice were rescued by i.v. transplantation of $5x10^6$ bone marrow cells performed 3 hours following irradiation. Recipient tissues were examined by independent methods (incl. quantitative PCR, flow cytometry, immunohistochemistry and fluorescent microscopy) to determine the rate of colonisation of tissues in non-splenectomised and splenectomised mice by transplanted cells at different survival periods (8 to 180 days).
A successful transplantation led to establishment of a stable donor-cell chimaerism in BM (approx. 80%) and in peripheral blood. Initial low engraftment in the thymus led to 100% chimaerism after a 180-day survival in non-splenectomised mice whereas in the small intestine and liver the highest levels were reached by day 180 in splenectomised mice (21% and 14% respectively). Results obtained from histological examination correlated with data obtained from qPCR analysis and provided detailed information on changes in local distribution of transplanted cells. In the small intestine, the highest positivity was observed in the mucosa while the epithelial lining contained only sporadic GFP^+ intraepithelial lymphocytes. Stroma of in intestinal villi contained up to 43% GFP^+ cell because of a high turnover and production of new connective tissue cells. Inside the liver parenchyma, cell chimaerism reached 26% after 180 days; GFP^+ cells participated in a turnover of stromal elements incl. endothelial and Ito cell while hepatocytes remained GFP-negative.
BM cells after systemic i.v. administration do not necessarily engraft homotopically back to BM. Our data demonstrate BM transplantation induces a stable post-transplantation chimaerism in several recipient tissues.

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The Evaluation of Osseointegration of Nano Titanium Screws

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Study Objective: Nano titanium is a quite new material consisting of extremely small particles. This material is thought to have good biological properties and thus can be used for medical purposes. The objective of this study is to evaluate the osseointegration of screws made of nano titanium.
Materials and Methods: In total, six nano titanium screws were implanted into the distal femur and proximal tibia of a rabbit under general anesthesia. The animal was treated with antibiotics and analgesics postoperatively. The correct position of implants was controlled by using a dental X-ray machine 14 days after the surgery. Two doses of the Tetracycline, was applied at two different periods of time (2 and 6 weeks after surgery) to demonstrate regions of active bone formation, mineralization and to demonstrate the quantity of newly formed bone at the implant interface with the help of a confocal laser scanning microscope (CLSM) due to its fluorescent property. The rabbit was euthanized after 12 weeks and the femur and tibia were removed. The position of the screws and bone apposition was checked by taking a series of radiographical images, using a dental X-ray machine, 3D CB CT and bone densitometry. A histological examination was performed and the quality of osseointegration of the nano titanium screws was evaluated.
Results and Conclusion: After bone processing and staining for eventual histological analysis, a series of microphotographic images were taken under both light and CLSM showing the bone-implant interface. This experiment determined the extent of osseointegration and the rate of healing of nano titanium implants. The results show that nano titanium implants are excellent at osseointegration and therefore can be a material of choice for implantology.

Project TIP FR-TI1/415

Volumetric analysis of pons, cerebellum and hippocampi in older patients with Alzheimer disease.

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We have performed manual volumetric MRI analysis of pons, cerebellum and hippocampi in 20 healthy controls and 20 patients with Alzheimer disease (both groups average age was above 70 years). Our goal was to find out whether decrease of hippocampal volume is accompanied by similar volume decrease of pons and cerebellum. Interestingly, we have not found at all statistically significant decrease of the volumes of selected brain structures in Alzheimer disease patients compared to age matched healthy group. Also, we have not found statistically significant right/left laterality in both groups as well as laterality differences between groups. This is in contrast with many studies, reporting generally hippocampal volume loss. The explanation of our finding could be in relatively higher age of control group, where shrinkage of brain tissue may take place for many other reasons. This study may indicate that volume loss of brain areas in Alzheimer disease may be diagnostically attributable to only certain age interval of patients and can not be used without age limitation.

Keywords: Alzheimer disease, laterality, pons, cerebellum, hippocampus, volumetry, MRI

Physiological role of tissue hypoxia in embryonic development

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Our experiments demonstrated that normal embryonic development of Japanese quail (*Coturnix coturnix japonica*) involves physiological tissue hypoxia during embryologic days (ED) 4-17 and the first 12 hours after hatching. Hypoxic regions marked by Hypoxyprobe-1 were present in the brain, liver, myocardium, limb primordia and metanephros. The extent of hypoxic regions decreased with the age of embryos and they disappeared within 12 hours after hatching. In the group of hypoxic embryos (incubated at 16% 0_2between ED4-ED9) the hypoxic regions became larger and more intensely stained and their vicinity showed higher capillary density. We found that quail embryos incubated under hypoxic conditions died at ED9 with signs of perturbed organization of vascular plexus, leading to impaired connection of coronary arteries to the aorta followed by heart failure and embryonic demise.

The expression patterns of HIF1 followed in general the Hypoxyprobe marked regions. All studied factors (HIF1alpha and beta, VEGF A) were proportionally elevated under hypoxic conditions; however, they remained restricted to their original sites, without being expressed at ectopic sites.

In conclusion, we have presented evidence that the local tissue hypoxia represents the physiological stimulus participating on embryonic morphogenesis of blood vessels. Experimentally induced hypoxia enhances this microenvironmental effect. At early developmental stages the embryo tolerates hypoxia well, responding by augmented vasculogenesis. In the subsequent period there is a failure in organization of coronary vascular plexus, leading to reduced connection of coronary arteries to the aorta followed by heart failure and embryonic death.

Another set of experiments showed accelerated conversion of ventricular activation sequence during hypoxic incubation and correlated with increased apoptosis in the atrioventricular canal. We hypothesized that this slight increase in apoptosis could lead to an accelerated pruning of myocardial atrioventricular connections, which would manifest as earlier appearance of mature apex-to-base activation patterns. Accordingly, hearts of embryos treated with caspase inhibitor zVAD showed a significantly increased proportion of immature (base to apex) activation patterns, including ventricular activation pattern from the anterior right atrioventricular junction never observed in control hearts. The number of apoptotic cells in atrioventricular canal myocardium was decreased in the treated hearts.

Apoptosis in the atrioventricular canal myocardium is thus an important determinant of ventricular activation pattern. Its inhibition at critical stages can lead to persistence of accessory atrioventricular connections, which form a morphological substrate for ventricular pre-excitation.

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MRI of the brain in congenital hypothyreosis in rats

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To investigate the impact of prenatal hypothyreosis on brain development, we performed macroscopic, MRI, and histological analysis of postnatal rat brains subjected to pre- and/or postnatal hypothyreosis.
Hypothyreosis was induced in pregnant rats since the time of conception by addition of propylthiouracil or methimazole in drinking water. Several groups of pups (prenatal only, prenatal and postnatal or postnatal only and controls) were obtained by cross-fostering on postnatal day 1. Sampling for MRI and histological analysis was performed at postnatal day 1 and weaning (day 30). The pups were weighed on postnatal days 1, 14, and 30.
Micro-MRI analysis and brain weights showed relative sparing of brain volume compared to stunted body growth and reduction in hippocampal volume apparent even after correction for brain weight in both postnatal only and prenatal/postnatal hypothyreosis. Histological analysis did not reveal any gross alteration in brain, hippocampal or cortical structures. Cross-fostering of pups exposed to prenatal hypothyreosis only produced animals that did not differ from controls in any analyzed parameter.
The correction of prenatal hypothyreosis in rats thus seems to result in full gross morphological compensation. However, functional deficit detectable by behavioral tests are not excluded; it should be also noted that the rat newborn is considerably less mature than the human one, corresponding thus more to the prenatal correction. This model thus provides basis for more precise definition of critical point of no return.

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Correlation of the anatomical structure of the amygdala with its MRI image

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Fast development of the MRI makes magnetic resonance image of the amygdala more and more similar to its anatomic counterpart. Some larger cerebral structures look in the MR image like in low magnification microscope. More precise delineation of the amygdala in the MRI is still not done.
Best known amygdalar subnuclei are: cortical, basal, lateral, medial and central. Recently are the „extended amygdala“ and „expanded amygdala” often mentioned as well. The extended amygdala is the part of the amygdala, which continues to the substantia innominata and the expanded amygdala is a „cap”on the rostral pole of amygdala, (preamygdalar area) related to olfactory areas.
Main amygdalar connections are organized as three circuits: Intrinsic circuit connects its subnuclei (step by step: cortical – baso-lateral and centro-medial subnucleus). Afferents of the cortical subnucleus come mainly from the olfactory areas. The baso-lateral subnucleus is connected via the bidirectional cortical circuit (for the neocortex, hippocampus and entorhinal cortex) and the centro-medial subnucleus is connected in reciprocal subcortical circuit (for the hypothalamus and brainstem structures, mainly the parabrachial nucleus and the „chemical“ brainstem nuclei).
Information about the MRI density of amygdalar subnuclei is not yet available. Simple comparison of the cytoarchitectonic and the MRI picture shows too much difference. We therefore introduced some „halfway“ steps to bring closer these two aspects. Unfixed „anatomical“ human brain is examined by MRI. After the shortest possible fixation we cut the block with amygdala out of the brain, and this block is examined again by MRI. After that it is processed and stained by Nissl reaction to reveal cytoarchitecture. By such technique we get identical sections – one in the MRI picture, and the corresponding section in the cytoarchitectonic picture. By comparison or transposition of both sections is possible to find and determine precisely the amygdalar borders in the MRI picture and to use thus gained experience for the routine MRI amygdalar evaluation.
Comparison of the normal amygdalar volume in the MRI picture with its volume in neurological patients can help to obtain more accurate diagnosis or prognosis of some diseases.

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Application of histological methods for distinction of tumours osteolytic lesions

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Nowadays we have the possibility to use some of the following examination methods for palaeopathological specimens: scanning electron microscopy (SEM), classic light histology using semi-thin Epon sections, stained by toluidine blue (LH), and laser confocal scanning microscopy (LCSM). Samples of two cases are demonstrated to show microscopic distinction between osteolytic metastases of carcinoma of a 50-60 year old male from Borovce, district Piešťany, Slovakia, (end of the 8th century – early 9th cent. AD) and multiple myeloma of a 30-40 year old female from Wien-Mauer,district Wien 23, Austria (beginning of the 4th millennium BC).
Their differences proved to be minute, concentrated in the changes of the osseous trabeculae, their morphological structure semi - thin sections (LH) and number and form of the lytic foci afflicting them. There are evidently different processi of destruction which can be found in the spongy trabeculae and compact layers [SEM,LCSM] .
From our results we conclude that SEM, semi - thin sections and LCSM are methods applicable to archaeological and historical specimes of adequate but also poor preservation.

Immunohistochemical aspects of bradykinin postconditioning in rat brain hippocampus

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This study deals with postconditioning (post-C) issue in the postischemic rat brain. We used this procedure to show some features of the different tissue protecting against ischemia-reperfusion injury. Post-C refers to a broad range of sublethal stimuli performed 2 days after cerebral ischemia. Delayed resistance to ischemic injury can be induced also by a variety of conditioning stress stimuli. This study is devoted to examining of bradykinin (BK) post-C impact on immunoreactivity (IR) of several different proteins (superoxide dismutases and proteins associated with apoptosis development) in the rat hippocampal CA1 vulnerable neurons and in the relative resistant dentate gyrus (DG). Adult male Wistar rats underwent 8 min of experimental ischemia and 3 days reperfusion and some of them was administered BK (post-C group) in a dose of 150 um/kg after 2 days of reperfusion. For immunohistochemistry animals were deeply anesthetized and perfused with 250 ml of saline followed by 250 ml 4% paraformaldehyde. Coronal sections were cut on a cryostat and then processed using indirect immunoperoxidase procedure with the ABC system with using of primary polyclonal antibodies against superoxide dismutases (MnSOD, CuZnSOD) and against proteins from apoptotic bcl2 family (Bcl-2 and Bax) too. Our study demonstrates that ischemia/reperfusion and BK post-C induced a substantial increase of MnSOD expression, but CuZnSOD-IR was decreased after BK administration. On the other hand, BK administration did not affect Bax-IR, but Bcl-2-IR was significantly increased in both brain areas. Our findings are, mainly in CA1, very interesting according to known persisting strong inhibition of protein synthesis during reperfusion. Application of adequate post-C stress is probably a condition of preventing of some degree of apoptosis and allowing more widespread cell survival. In advance of the postischemic neurodegeneration, increased immunoreactivity of all followed compounds could prevent the process of delayed neuronal death.

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Ultrastructural changes in pancreatic cancer cells

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We compare ultrastructure of normal pancreatic tissues with nonmalignant part of cancerous organs and from primary ductal pancreatic adenocarcinoma.
We take a spatial interest in ultrastructure of mitochondria. The mitochondrial morphology is continuously modified by functional requirements to adapt to different cell demands. This organelle has been implicated in the process of carcinogenesis, which includes alterations of cellular metabolism and cell death pathways. The mitochondria in cancer cells, independently of histogenesis, are seen with dense matrix or condensed configuration and with lucent-swelling matrix associated with disarrangement and distortion of cristae and partial or total cristolysis.
Functionally, the structural alterations suppose the presence of hypoxia-tolerant and hypoxia-sensitive cancer cells. Possibly, hypoxia-tolerant cells are related with mitochondrial condensed appearance and are competent to produce adequate amount of ATP by mitochondrial respiration. Hypoxia-sensitive cells are linked with lucent-swelling and cristolysis mitochondria profile and have an inefficient or null oxidative phosphorylation,
which consequently use the glycolytic pathway to generate energy.

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Effective stabilisation of mast cell granules by sodium cromoglycate leads to changes in percentage of double-laminated vessels during hypoxia and posthypoxic recovery

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The hypoxic experimental conditions induce the development of the hypoxic pulmonary hypertension, which is mostly caused by remodelling of pulmonary resistive arteries (PRA) (Ø <100 μm). This remodelling is characterised by neomuscularisation in the level of prealveolar arteries and hypertrophy of tunica media at the level of peripheral arteries. Having returned to the normoxic environment, the pulmonary resistive arteries are re-remodelled back to the normoxic shape. Both these processes are mediated by mast cell (MC) products depending on changes in the MC distribution and production of the interstitial collagenase.
The effective stabilization of MC granules by sodium cromoglycate (SC) at the beginning of hypoxia leads to the restriction of hypoxic remodelling and conspicuously enables recovery re-remodelling of the PRA back. The effective stabilization of MC granules at the beginning of recovery period leads to the restriction of re-remodelling of the PRA during the recovery period.
Hypoxic remodelation processes of PRA are also morphologically characterised by formation of membrana elastica externa (MEE). MEE with existing membrana elastica interna makes an image of “double laminated vessels” (DLV).
We assessed the changes in percentage of DLV during hypoxia and posthypoxic recovery, depending on the duration of hypoxia and recovery phase and variant timing of SC administration. Wistar-Han male rat lung slides were assessed in the orcein staining.
This exact method was able to confirm our previous morphological results as a significant.

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Structure of Vertebral Arch in Human and in Selected Mammals

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Fissured discontinuation of the vertebral arch (spondylolysis) can occur in various forms and segments of human spine. Caudal lumbar vertebrae are most frequently damaged as a result of the most intensive mechanical strain. Spondylolysis has been described in humans only, therefore the relationship between the origin of the disease and erected posture and bipedalism can be assumed. The etiology of spondylolysis is not very clear. It is usually considered a fatigue fracture, however its congenital origin cannot be excluded. Multifactorial influences are also taken into consideration, such as hereditary predisposition to fractures occurrence due to slight morphological spinal abnormalities. The presented study deals in depth with morphological analysis of the interarticular region of vertebral arches of the most distresses part in humans and with the comparison of vertebral arch structure in selected species of mammals. For the purpose of the study cross-sections, bone ground sections, X-ray images and CT scans of caudal thoracic and lumbar vertebrae of the man, horse, cattle, sheep, pig, dog, tiger and baboon. Due to different spinal strain in tetrapods, the cranial thoracic vertebrae and the sacrum were also studied in the above noted animal species. In contrast to the man where compact bone tissue is present in the vertebral arch of the last lumbar vertebrae in the region between superior articular process and inferior articular process, in all examined mammalian species in the corresponding region of monitored vertebrae – between cranial articular process and caudal articular process – coherent cancellous bone tissue covered with a corticalis layer can be found.

Newly opened elected subject for students of the Faculty of Veterinary Medicine Brno

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Within the framework of the project for the consolidation of the preclinical and clinical subjects at the VFU Brno, there was new elected subject for students of VFU, dissection course, opened.

The dissection course is intended especially for students who study anatomy, it is the students of the 1st year during summer semester and the students of the 2nd year during winter semester. Under the supervision of assistants, the students dissect mammals which are subject to the studies of anatomy (dogs, cats, ruminants, horses and pigs). Besides these kinds of animals, there is an occasional possibility to dissect exotic mammals. The dissection is targeted not only at the deepening of the knowledge of anatomy, but also at the preparation of fixed preparates for lectures of anatomy. The collection of preparates is necessary to be renewed and completed continuously. The preparates are prepared either as individual organs, parts of bodies, or, in case of muscle preparates, the whole bodies of animals. These are the so called wet preparates, which are fixed in formalin and stored in cuvettes. These preparates are used during the lectures for demonstrations.

Another outcome of the dissection course will be a publication composed of photographs of topographical regions of dog including detailed description. This publication should serve students as an auxiliary studying material not only in the 1st and 2nd year of their studies, but also during the studies of clinical branches and for graduates in veterinary practice. The dissection of dogs will be mostly focused on the topography of organs and cavities of bodies which are, from the surgery point of view, the most frequent ones.

Changes of cytoskeletal proteins during prolonged subcultivation of human "glia-like" cells

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Intermediate filaments (IF) are one of three types of cytoskeletal proteins. They are often used as differentiation markers for normal and neoplastic cells. The aim of this study is to show the differentiative properties of human "glia-like" cells.

The tissue cultures were derived from 20 adult human brain biopsies. The cells in primary and secondary passages were examined by immunofluorescence methods using specific antibodies to glial cell types and two IF: glial fibrillary acidic protein (GFAP) and vimentin. The immunofluorescence staining revealed the presence of small amount of morphologically and immunochemically distinct astrocytes, oligodendrocytes and microglial cells in early passages which disappeared by 4th passage. The secondary well proliferaing cultures contained only GFAP-/Vim+ "glia-like" cells. Spontaneous growth deceleration occured in cultures within passage 8 to 15. Formation of processes and enlargement of cell bodies were characteristic for this period. The cells became very heterogeneous in size and shape. Some of them were astrocyte-like in shape. These morphologically changed cultures with successive arrested growth became exclusively GFAP+/Vim+ astrocytes.

Cells cultured from adult human brain have been poorly defined until now and are often termed "glia-like" cells. Our results demonstrate that these flat initially GFAP-negative cells became GFAP-positive during the differentiative period at the end of their life span.

Immunofluorecence analysis demostarate the different expression of both IF. Vimentin is expressed in all glial and "glia-like" cells, however GFAP staining is dependent on differentiative properties of cultured "glia-like" cells. Based on this study we suppose that these cells are glial precursor cells.

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Endocardial fibrolastosis in chick model of embryonic pressure overload

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The term endocardial fibroelastosis(EFE)refers to a pronounced,diffuse thickening of the ventricular endocardium, causing myocardial dysfunction and presenting as unexplained heart failure in infants and children.The endocardial thickening is believed to be caused by persistent and increased wall tension in the ventricles.The EFE can be primary or secondary to various congenital heart diseases, most notably hypoplastic left heart syndrome.We have tested the hypothesis that increased pressure loading in the developing heart will lead to EFE in a chick model of conotruncal banding.The eggs were incubated in a force draft incubator at 37.5°C and high humidity.Conotruncal banding of chick embryonic hearts was performed at the ED4,leading to increased pressure in the ventricle. At ED12 modifications of myocardial architecture were studied by histology and immunoconfocal microscopy.Histology of pressure-overloaded hearts showed chamber dilatation and compact layer thickening.However, H&E with Alcian Blue staining did not reveal any significant fibrosis with the exception of cardiac skeleton and valves.Immunohistochemistry with anti-periostin antibody clearly showed fine layer of subendocardial fibrous tissue that was more prominent in the pressure-overloaded hearts.It appears that increased pressure loading stimulates fibrous production in the subendocardium.Further experiments involving later sampling points and proliferation studies will be necessary to confirm these findings.

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Orofacial development and cleft anomalies

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Orofacial clefts belong to frequent developmental defects in human population. The critical period for development of orofacial clefts in man lasts from embryonic day 30 till 60. Cleft lip results from failing fusion between the medial nasal and maxillary facial processes, while the isolated cleft palate is a consequence of the absence of fusion between the palatal processes of maxillae. In majority of cases, the non-fusion is caused by hypoplasia of the respective facial or palatal processes induced by epigenetic or genetic factors. The isolated cleft palate can also follow the growth retardation of the lower jaw, whose forward growth normally redraws tongue from the space between the palatal shelves (Pierre-Robin syndrome).
The incidence of orofacial clefts in the Czech population, and its annual and district variations were analysed on the basis of data on pregnancy history of mothers of 5000 children born with an orofacial cleft during 1964 - 2009. The patients were collected at the Clinic of Plastic Surgery, University Hospital Kralovske Vinohrady. The incidence of the cleft anomalies was correlated with natality data in the Czech Republic. The long-term incidence of orofacial clefts in living newborns was 1:530. However, this ratio progressively decreased during the last 10 years to 1:750. This decrease can be explained by increased quality of ultrasound examination allowing early detection and induced abortion of the most serious orofacial cleft anomalies.

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Rudimentary structures and their importance during dentition development

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The knowledge on the factors that promote or inhibit organogenesis is important not only from the aspect of normal and abnormal development. It can also help in understanding evolutionary loss or reappearance of a structure, as well as in designing the methods of regenerative medicine aimed to biological organ replacements.

Development of the mouse dentition with strongly reduced tooth number represents a very useful model for studies of organ progression and regression, and of their molecular control. Two types of tooth primordia occur in mouse embryos: some primordia will form functional teeth, whereas the other primordia are rudimentary, and mostly stop their autonomous development at a bud stage with aid of apoptosis. The rudimentary tooth primordia in mouse embryos have been related to the ancestral teeth lost during muroid evolution.

In course of mouse prenatal development, the rudimentary tooth primordia become either extinct - to determine the prospective toothless diastema, or incorporated during morphogenesis of functional teeth - to be integrated in the new context. The mechanism of rudiments suppression in course of ontogeny might help in understanding the way of disappearance of the corresponding functional structures during phylogeny.

Under specific conditions, rudiments can express a retained ancestral developmental potential. A revivification of the rudimentary (ancestral) tooth primordia, resulting from decreased apoptosis and increased proliferation, is involved in origin of supernumerary teeth in some mutant mice. Such supernumerary teeth can be interpreted as atavisms.

Rudimentary structures also occur during odontogenesis in other animal species, including human, and represent an integral component of the developing dentition in general. These data support the view that rudiments are no superfluous structures, but have important ontogenetic and evolutionary implications. Furthermore, studies of the revitalization of rudimentary tooth primordia in mutant mice can help to lay the foundation for tooth regeneration therapies.

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Tumor infiltrating lymphocytes as a prognostic factor after the radical surgery of colorectal carcinoma

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Aim of the study: Correlation of the broadly used clinical and histopathological factors with the presence of tumor infiltrating lymphocytes (TIL) in the tumor. The goal of the study was to identify patients with the high risk of the poor overall survival (OS) and early reccurence (short disease-free interval, DFI) of the malignity after the radical surgical treatment.
Methods: The set of 150 tumor samples was analysed for pre-operational leukocytosis, radicality of the surgery, post-operational complications, oncological treatment, tumor grading and staging, morphological features of the tumor, characterisation and quantification of the TIL and the reactive changes in the lymp nodes (LN). All of these factors were statistically correlated with the OS and DFI of the patients.
Results: Included patients had the 1, 3 and 5 years OS 92,2%, 76,5% and 70,2% respectivelly, and 1, 3 and 5 years DFI 85,3%, 64,3% and 49,4% respectivelly. Endolyphatic infiltration and metastatic inflitration of the LN were confirmed as a negative prognostic factor for OS. N2 stage decrease the OS 9,3 times and DFI 5 times. Crohn-like peritumoral lymphocytic infiltration and follicular hyperplasia were confirmed as a protective factor. CD8+ and CD4+ TIL were confirmed as a positive prognostic factor and their presence leads to the increase in OS and DFI. Perineural infiltration was confirmed as a prognostic factor of early reccurence. Higher infiltration by CD8+ TIL was found to be positive prognostic factor which increase the DFI.
Conclusion: TIL are important factor which corresponds with the ability of the organism to attenuate the development of the malignity.

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Chordin and follistatin in craniofacial development

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The development of human face is the complex process where frontonasal mass fuses with the maxillary prominence to form the upper lip. Failure of any step of fusion results in the cleft lip and/or palate. Bone morphogenetic proteins (BMPs) are signaling molecules that regulate many aspects of development including skeletogenesis and craniofacial patterning. Furthermore, the application of BMP antagonist Noggin in combination with retinoic acid (RA) leads to the transformation of the maxillary prominence into the frontonasal mass (Lee et al. 2001). Here, we decide to test other BMP antagonists (follistatin and chordin) and their potential to transform or change the development of the maxillary prominence in chicken embryos. We used developmental stages 14 - 24 (Hamburger and Hamilton, 1951) and applied inhibitors together with retinoic acid into the maxillary prominence. Furthermore, each antagonist was also injected separately to reveal their influence on the development. Embryos were incubated for ten to twelve days after treatment. Chicken heads were collected and fixed in 100% ethanol for skeletal analysis. In case of early embryonic dead, samples were collected into 4% formaldehyde for macroscopic analysis of craniofacial malformations. Later, samples were stained with Alizarin Red and Alcian Blue for visualization of bone and cartilage deformations. Follistatin treatment leaded to only occasional disruption of craniofacial development (chordin alone - 8/40, follistatin with RA - 3/23) with upper beak deviation, unilateral cleft, hydrocephalus or microophthalmia. Chordin treated embryos exhibited mostly shorter mandible and abnormal maxillary bone (chordin alone - 2/5, chordin with RA 0/6). However, embryos did not exhibit any sign of frontonasal mass duplication similar to noggin-retinoic acid treatment.

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Today's state of Hyrtl´s skull collection

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The aim of this paper was to learn more about current state of Josef Hyrtl´s skull collection. Josef Hyrtl (1810 -1894) was a famous anatomy professor at Prague and Vienna universities. During his life he collected about 2000 skulls, which had a rich historical, educational, anthropological and clinical value.

The best information about the number of the skulls, their origin, variations, races, gender, pathologies or comments we can get from Hyrtl´s book "Vergangenheit und Gegenwart des Museums an der Wiener Universität" (1869). To this year the collection contained 1584 pieces. In 1874 part of the collection (139 skulls) Hyrtl sold to an American scientist Thomas Bache who established in Philadelphia Mütter Museum, where these skulls are still exhibited and well known.

Hyrtl failed in establishing a special museum for his skulls and so it split up during the 115 years from his death. The substantial part of the original Hyrtl´s collection is nowadays available in Narrenturm – Museum of Pathology and Anatomy in Vienna, where also we had an opportunity to study it. There is also his first skull he received from professor Czermak in 1834. Hyrtl enlarged his collection continuously during his study journeys and also through his broad contacts. He obtained many skulls of different races from several continents, of famous musicians (e. g. Mozart, Beethoven or Hayden), of patients with various diseases or even famous criminals. Every skull was documented and depicted by his own hand. The collection is also rich in teeth variations. In the lecture we will introduce you some unique specimens of this collection and their history.

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Czech breastfed children growth in the first year of life

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Introduction: Breastfeeding is recommended by World Health Organization (WHO) as the best way of alimentation in the first sixth monthes of the life and it is included amongst important preventive factors in the obesity. Basic tool for child growth evaluation are growth charts. Drawback of contemporary charts is its unsufficient specifically evaluate growth of breastfed children. WHO disclosed new growth standards in 2006, that were constructed on the base of growth data of lactated children with timespan at least 4 months.
Goal: to compare growth process of czech long-term breastfed children aggregate since birth till 1.5 year of age with growth charts used for evaluation of growth of czech children population.
Materials and methods: We have used 959 children breastfed at least for 4 months. Data collection ran over years 2009 and 2010. Czech reference charts are constructed according to data acquired during 5th and 6th Nation Wide Anthropology Research.
Results: Course of children growth in the monitored aggregate differs significantly from regularly used growth charts for the czech population. The highest discrepancies are found in the weight data. In the first 4 months breastfed children have higher weight, in the following months weight increment gets lower compared to reference data. Discrepancies in the length located on the 50th percentile are max. up to 0.5 cm, therefore, current reference data of length are sufficient even for long-term breastfed children.
Conclusion: Contemporary growth charts do not appreciate completely growth of lactated children, especially in lower age cathegories. Nevertheless, with the knowledge of specificity of breastfed child growth it is still possible to use czech growth charts.

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Analysis of expression of drug resistance proteins in non-small cell lung cancer in relation to prognosis

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We detected p53 and XRCC1 protein expression in 54 samples of NSCLC (non-small cell lung cancer). For detection of monitored proteins the immunohistochemical method was used. Tissue samples were divided according to type of tumor. Next we compared our results with basic clinicopathological parameters. Statistically significant correlation was found between type and p53 expression ($p<0.05$). Comparing the p53 expression with grade resulted in strong positive correlation, $p<0.05$ ($R^2=0.9223$).The percentage of p53 positive tumors increased from 0% in grade 1 to 75% in grade 4, respectively. No correlation in p53 expression and tumor stage was found. In case of XRCC1 the highest level was find in squamous cell type, where 71% of samples were positive. In case of large cell type it was 67% and in adenocarcinoma 52% samples showed XRCC1 immunoreactivity. We did not find significant correlations between type, grade and early stage of NSCLC and expression of XRCC1 protein profile without neoadjuvant therapy.

We found significant statistical correlation between expression of p53 and type of tumor. It is possible that stabilized p53 protein plays an important role in squamous and large cell types development. Our findings also suggest that p53 expression cumulates with dedifferentiation of cancer cells. Expression of XRCC1 is probably not fixed and could be changed by the status of cancer cells and in relation to therapy. The relevant data about pre- versus post- chemotherapy and XRCC1 expression are needed to evaluate the influence of XRCC1 to drug resistance.

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Parvalbumin and Calretinin immunoreactivity of the infralimbic cortex in the rat

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The infralimbic (IL) area is one subdivision of the medial prefrontal cortex. These subdivisions differ significantly functionally, in their connectivity and in expression of transmitters and neuropeptides. Our recent findings indicate that the IL area belongs among brain structures that are seriously damaged after status epilepticus (SE) in young immature animals.

For further exploration of inhibitory functions of the IL area, a precise description of distribution and morphology of inhibitory interneurons might be of of great value. We used calretinin (CR) and parvalbumin (PV) immunochemistry, as these calcium binding proteins are well known as markers of different interneuronal inhibitory subpopulations.

Brains of six male Wistar rats weighing 350 – 400 g were used for this study. Brains were postfixed, cryoprotected, sectioned at 50 μm and stained immunohistochemically for CR and PV**.**

The overall level of CR – immunopositivity in the IL is lower than that in the neighbouring prefrontal cortical areas. CR- ir neurons were distributed in all layers but their density was higher in supeficial layers. Additionally to CR, on PV stained sections the IL is characterized by markedly lower immunopositivity of neuropil. PV – ir neurons were more numerous in deep layers.

The pattern of CR and PV – immunorectivity could contribute to explanation of severe damage of the IL area after SE.

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Is there any correlation between results of the final exam in anatomy and results of the current assesments in practicals?

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The two-semester subject anatomy belongs to among the basic and at the same time most difficult fields of the preclinical medical study. Aquired knowledge covers extensive structural data and anatomical nomenclature. The programme of the lectures and practical training is as follows: 1st semester - locomotor apparatus, GIT and respiratory system; 2nd semestr - urogenital system, cardiovascular system, nervous system and sense organs. Primary the basic approach is systematic with special attention paid to regional anatomy as well as to orientation in X-ray pictures, CT scans, MR images and angiographic examinations. To pass the subject and the final examination succesfully it is required to have a well elaborated system of current assesment (oral examination, tests) at practicals.

The aim of our study is to compare the results of current assesments with the results of final anatomical exam. Altogether 1152 students in 8 years (2002/2003 – 2009/2010) were evaluated. In 56% of the students the results of the final exam were in good correlation with the results of the current assesment. In 32 % of the students the results of the final axamination were worse and in 12% better than those of the current assesments. Possible causes of these differencies are discussed. These results suggest that approximately in 44 % of the students the results of current assesments at practicals do not predict the results of the final examination.

Normal development of cardiac conduction system in the mouse

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Conduction system development in the mouse (Mus musculus) was described histologically in the 1970s, and recently, optical mapping provided clues about its functional deployment. However, quantitative evaluation of ventricular activation times and patterns, necessary for interpretation of changes observed in transgenic mouse models of arrhythmias, is lacking.
Applying method of optical mapping of action potential propagation, we have studied function of conduction system in mouse embryos starting at 9th embryonic day (ED). We measured total activation time of the left ventricle and evaluated way of ventricular activation by classification into activation patterns; namely activation utilizing primary ring, left bundle branch (LBB), right bundle branch (RBB), both bundle braches and transitional types.
Typical situation at any given ED is represented by a spectrum of several activation patterns, therefore, is impossible to describe normal development with analysis of less than 10 embryos per group. Early conduction at ED9-11 is characteristic by utilizing the primary ring, structure in the future interventricular septum. As development proceeds, activation through the primary ring disappears, and at ED13 no heart was activated by this structure. From ventricular conduction system compounds is first active LBB but with appearance of transitional type of activation, when electrical impulse is originating in apex with fast spread in primary ring. At ED11 becomes active RBB or both branches. At ED14 is majority of hearts activated from two centers, but occasionally originated from single one.
Spread of electrical impulse is accelerated in the developmental window ED10 – 12, as evidenced by shortening of left ventricular epicardial activation time. Thereafter, the activation time remains flat but since the heart continues to grow, the apparent speed of conduction increases. Quantification of normal development of ventricular activation provides a necessary framework for analysis and interpretation of mouse mutants with suspected developmental conduction system pathologies.

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Morphologic signs of cell-to-cell communication in hESC colonies

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Human embryonic stem cells (hESC) are undifferentiated pluripotent cells derived from the inner cell mass of blastocysts, with the capacity to both self-renew and to differentiate into all somatic cell lineages. During their *in vitro* cultivation, hESC proliferate to form colonies. Cells within colonies employ various communication mechanisms to influence their activities including possible initiation of differentiation. Here we have investigated whether communications between individual hESC can also be observed at the level of their morphology.

Cells of CCTL14 line of hESC (passage No 23 and 35) were routinely cultured in DMEM media supplemented with serum replacement and 4 ng/ml FGF2 on the layer of mouse embryonic fibroblasts serving as feeder cells. Colonies of hESC were processed for transmission and scanning electron microscopy on days 3 and/or 4 after seeding.

Neighbouring cells in colonies communicate directly via intercellular junctions. All types of intercellular connections were found: occluding, adhering and communicating (gap junctions). Interactions with distant cells may be mediated by various types of vesicles - numerous vesicles were shed from the cell surfaces, and, on the other hand, intensive formation of coated vesicles was observed. In addition, uncommon intercellular connections were found in most colonies. This special type of connection was accomplished by long processes (0.2-0.3 μm thick) extending from the surfaces of two separate cells with peculiar figures resembling rosette at the contact site.

Intercellular junctions and numerous morphologic signs of material exchange reflect intensive intercellular communication in hESC colonies. Special type of intercellular connection needs further investigation.

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Ontogenesis and phylogenesis of cardiac conduction system

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The cardiac pacemaking and conduction system is responsible for generation of intrinsic rhythm of the heart and coordinated spread of the action potential through the cardiac compartments. During ontogenesis, the first cardiac contractions start at the straight tube stage, approximately ED8.5 in the mouse and 22 days in humans. The excitation is started in the primitive pacemaker, situated at the inflow portion of the tube that shows slow speed of impulse propagation and peristaltoid activation pattern. During chamber formation, the conduction velocity remains slow in the atrioventricular (AV) canal and notably accelerates in the atrial and ventricular chamber. The AV canal thus fulfills the function of the delay generator later executed by the AV node. The primitive ventricular conduction system is formed by the trabecular network, and hallmark of its function is ventricular activation from the apex towards the base, which appears before completion of septation. This functional arrangement is conserved among vertebrates down to fish, including the apex-to-base activation of the ventricles. While there is no morphologically distinct ventricular conduction system in the lower vertebrates, the function is performed by the trabecular network. There is also no distinct AV node, but the ECG is remarkably similar to homoiotherms, as the function of delay generator is performed by the AV canal connected directly to the ventricular trabeculae. The comparative and ontogenetic approach is useful to understand the mechanisms of conduction system formation and its pathologies.

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Co-expression of extracellular matrix and cytoskelatal proteins in adult human brain tissue cultures

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The extracellular matrix (ECM) of vertebrates is composed of proteins and carbohydrates that binds cells together. Fibronectin (Fn) is one of the major glycoproteins of the ECM, produced by a variety of mesenchymal and neoplastic cell types. Glial fibrillary acidic protein (GFAP) is intermediate filament cytoskeleton specific for astroglial cell differentiation.

The aim of this study is to demonstrate influence of culture conditions on GFAP and Fn co-expression. The explant tissue cultures were prepared from 20 adult human brain bioptic samples. All cultures were examined using indirect and double immunofluorescence with antibodies against Fn and GFAP. In confluent primary cultures we have found approximately 0,1 % of GFAP+/Fn- astrocytes which disappeared by 4th passage. The remaining cells of non-glial phenotype showed strong extra- and intracellular staining for Fn. The cultures in early passages showed rapid cell proliferation and contained homogeneous population of GFAP-/Fn+ cells. During passage 8 to 15 the cell growth spontaneously slowed down, cells changed their morphology and showed positive staining for GFAP. The intensity of Fn staining slowly decresed in GFAP-positive cells. However, the majority of GFAP-positive cells retained the Fn expression at various intensity.

Currently Fn was found only in cultured embryonic and immature astrocytes. The positive staining was not found in adult astrocytes. Our result indicate that small amount of differentiated GFAP-positive astrocytes which persist in early passages are negative stained for Fn. However, during the process of spontaneous differentiation of adult human brain tissue cultures the cells co-express GFAP and Fn.

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Changes of rat pulmonary parenchyma in experimental model of early sepsis and gentamicin treatment pharmacokinetics

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We investigated the influence of covariates generated by a rat sepsis model on gentamicin pharmacokinetics. Endotoxaemia was induced by i.v. administration of 1mg/kg Pseudomonas aeruginosa lipopolysaccharide in combination with 15μg/kg recombined mice interleukin-2. Gentamicin i.v.bolus (3mg/kg**)** was followed by i.v.170-min gentamicin infusion (0.09 mg/kg.min^{-1}) started 10 min later. Experimental groups included controls infused with saline only (group 1, n=20), rats infused with saline followed by gentamicin (group 2, n=7), rats given i.v. LPSI bolus and i.v. saline in continuous infusion (group 3, n=27), and rats given LPSI followed by gentamicin (group 4, n=7). Blood sampling was done at time zero (baseline), during gentamicin treatment, and at 420min of experiment, when animals were euthanized and target organs removed for histopathologic examination.

Endotoxaemia induced microvascular leakage, decrease in glomerular filtration rate, tubular dysfunction, and lactacidemia (group1versus 3, examinated by clinical biochemistry). Histopathology proved pulmonary changes (leakage, alveolar wall oedema, inflammatory cells and erythrocytes in the alveoli). Spleen showed a higher amount of erythrocytes infiltrated to the white pulp.

Changes of gentamicin pharmacokinetics in endotoxaemia (group4versus 2) were as follows: extended volume of distribution: 0.343±0.117 versus 0.224±0.027 L/kg (*p>0.02)*, decreased gentamicin renal clearance: 2.18±1.55 versus 3.81±1.39 mL/min.kg^{-1} *(p>0.03),* while total clearance was comparable 5.95±2.04 versus 5.78±0.20 mL/min.kg^{-1}, with rising interindividual variability in endotoxaemia (34.3% versus 2.9%, respectively, CV[%]=σ/|μ|). T1/2 was prolonged: 45.43±12.15 versus 30.58±1.16 min (*p>0.007).* In endotoxaemia, gentamicin dosage should be kinetically quided.

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Epithelial-mesenchymal interaction in cancer

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Cancer microenvironment represents a very important factor significantly influencing the biological behavior of the tumor such as local aggressiveness and metastasation to regional lymph nodes and distant organs. This tumor spreading through organism was depicted in so called Seed and soil hypothesis published 120 years ago by the London surgeon Stephen Paget. It has been resurrected in the last decade by people working in cancer stem cells research. It is a very good parallel to adult tissue stem cells which also require the specific microenvironment called niche to maintain their stemness. The cancer stem cell niche seems to be presented by the tumor stroma – the complex tissue composed from fibroblasts, extracellular matrix, inflammatory cells and capillaries. The aim of this presentation is review of data about cancer-associated fibroblasts (CAF), predominantly in tumors originated from squamous cell epithelia. The presented data demonstrate that CAF are able to significantly influence the phenotype of epithelial cells, and their role in the participation of the control of tumor cell growth and migration is probable. Therapeutic manipulation of these cells can represent a new paradigm of biological anticancer therapy.

Changes in collagen volume fraction and microvasculature in human atrial wall during atrial fibrillation

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Atrial fibrillation (AF) is one of the most often encountered arrhythmic diseases in the clinical practice. In this project we focused on morphological and functional changes in endomysium of atrial myocardium with special reference to changes in the extracellular matrix, formation of blood vessels and pericytes regulatory role in remodeling of atrial myocardium. We studied the morphological changes of atrial biopsies performed at 70 patients (29 patients with AF, and 41 with the sinus rhythm) undergoing bypass or mitral valve surgery.The atrial samples were fixed with 4% paraformaldehyde and embedded into paraffin. Sections from atrium were histologically examined using routine staining (HE, Sirius Red) and immunohistochemical techniques. When the morphometrical calculation of fibrosis based on Sirius Red staining was performed, we obtained the following results, depending on the localization: left auricle SR (23.78±5.1%) and AF (28.44±6.7%); left atrium SR (24.20±3.1%) and AF (29.31±8.8%); right auricle SR (27.40±6.1%) and AF (27.16±4.45%). As a result of the immunohistochemical analysis, we found presence of protein α-SMA in pericytes and lack of protein desmin. Our findings show a slight increase in atrial myocardium fibrosis of patients with AF; however, this increase is not statistically significant. The results are of value for the further study of endomysial changes during AF.

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Why five fingers ? Genetic control of limb development.

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Digits are essential structures of terrestrial vertebrates evolved for functionally different goals like movement, feeding, communication. Specialized regions of the developing limb bud regulate its development including the patterning of digits. Different genes and gene families such as *Shh*, *Hox*, *Fgf* , *Bmp* are involved in this intricate process.
Despite the extensive studies of limb development in model organisms (mainly chick and mouse) and studies of congenital anomalies found in humans the exact mechanisms that control the number and identity of the digits remain unclear.
We review the current understanding of this problem and also results of our work with polydactylous *Lx* and hypodactylous *hd* rat strains.
Lx in SHR.*Lx* rat manifests in homozygotes as hindlimb preaxial polydactyly
A 2,964-bp deletion in *Plzf* intron 2 removes the most deeply conserved noncoding element leading to later onset and reduced pattern of *Plzf* expression in *Lx/Lx* limb compared to both +/*Lx* and WT. Anterior expansion of *Hoxd10–13* and Bmp2 genes expression, in the absence of ectopic *Shh* expression is observed in *Lx/Lx*.
Rat *hd* is an autosomal recessive mutation manifesting in homozygotes as reduction or loss of digits II and III on both hind and forelimb and impairment of spermatogenesis leading to male infertility
hd mutation is caused by an insertion of an endogenous retrovirus into intron of the *Cntrob* gene resulting into the translation of a truncated protein.
Expression of *Sox9* and*Bmpr1b* (acting as an upstream activator of *Sox9* expression) is absent from the distal part of the digit condensations II and III, in digits which are affected in *hd* rats.

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Effect of thyroid status alteration and red palm oil supplementation on motor activity and thermal sensitivity of adult rats.

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We studied effect of thyroid hormone alteration and red palm oil (RPO) supplementation on motor activity and thermal pain perception in adult female and male Wistar rats. Mobility was tested by computer-based tracking system in circular arena, latency of responsiveness to thermal stimulation was tested with radiant heat applied to the plantar surface of each hind paw. The mobility test indicated that the HY (hypothyroid) rats performed notably shorter tracks when compared to the EU (euthyroid) and TH (hyperthyroid) ones, RPO normalized mobility of the HY rats to the level of the EU ones and increased mobility of the TH rats above the EU level. Off-line track analysis showed eminent thigmotaxis, the highest in the hypothyroid rats, as all rats preferentially walked around the walls of the apparatus. RPO had no significant effect on this parameter in either thyroid group. Thermal test showed a tendency for increased latency (decreased sensitivity) in the HY rats and opposite changes in TH rats, these differences were statistically significant as shown by multiple comparison method. RPO decreased latency in HY and EU rats (although in the latter to a lesser extent), while it did not affect this parameter in TH rats. Both tests revealed minor sex differences. Our results thus show that alteration of thyroid status can to minor extent change rat mobility as well as their thermal pain perception and that RPO can partly ameliorate some differences in the mobility and thermal stimuli perception caused by alteration of the thyroid statuses.

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Revolutionising the large-scale production of high quality adult stem cells – PurStem project

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Stem cells (SC) offer a promising avenue to therapy for a wide range of complaints. However, for this potential to be realized, a consistent and plentiful supply of well-characterised SC is essential. There has been relatively little progress in the development of new culture technologies for the large-scale manufacture of mesenchymal stem cells (MSCs). There is a strong possibility that this limited ability to produce SC will result in delays to the translation of new therapies to the clinic. This will have a direct negative effect on the health of EU citizens suffering from diseases untreatable by conventional medical technology and delay EU efforts to promote "NanoMedicine - Nanotechnology for Health". PurStem will progress the state of the art in the production of MSCs in large quantities. The current state of the art has several weaknesses - there are no standards for characterisation, isolation or identification of MSCs from any tissue, nor are there standard protocols for differentiation of MSCs to various lineages. Additionally, surface markers used for MSC characterization lack specificity and cryopreservation protocols are not standardized. Critically, current production methods for MSC require the use of animal products with major contaminant implications. PurStem will:
- Identify the MSC "receptome" and; - Use this repertoire of growth factor receptors to; - Develop novel serum-free media for MSC production. PurStem will also result in novel antibody reagents for specific MSC characterization and contribute to GMP manufacturing standards to enable rapid progression to production of serum-free MSC for clinical applications. The impact on a range of therapeutic and research domains of having a reliable supply of industrial levels of categorised MSCs will be significant. PurStem represents a key enabler for SC applications in a range of therapeutic fields.

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The synovial bursae in the past and today

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The synovial bursae belong to the integral parts of the locomotor systems of the upper and lower extremity, and many of them are of great practical importance in the clinical medicine. As the first one in the history, who paid them the attention, was B. Albinus. He described 16 pairs of bursae on both extremities in year 1734. In the same century enlarged substantially the detailed description of bursae A. Monro Jr. (1788) and J. C. Rosenmüller (1799). During the first half of the 19th century many other authors have been studying the bursae, above all J. Hyrtl and W. Gruber. The top of all these efforts represents the monograph of A.S.D. Synnestvedt (1869), in which 121 pairs of bursae were described. Since the rise of the first official anatomical terminology (B.N.A., 1895) the amount of the acknowled bursae was fluently and gradually reduced up to the recent 44, as they were defined in the last version of the Terminologia Anatomica (1998). The knowledge of these structures plays recently an important role in the diagnostics and therapy of their pathologies, namely in the general surgery, orthopaedic surgery, rheumatology and in radiology, above all.

Activated satellite glial cells in the trigeminal ganglion after unilateral infraorbital nerve injury

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Peripheral nerve injury induces an activation of the satellite glial cells (SGCs) in the sensory ganglia. Activated SGCs are believed to contribute to increased excitation of the primary sensory neurons and neuropathic pain induction. The goal of our work was to investigate bilateral activation of SGCs in the trigeminal ganglion (TG) following unilateral infraorbital nerve ligation (IONL).
Wistar rats (males, 270g) were operated on unilateral IONL (n=12) and allowed to survive for 3 (n=6) and 7 (n=6) days. Naive (n=4) and sham-operated rats (n=4) were used as control. Immunofluorescence staining of TG sections for GFAP and ATF3 were used for detection of activated SGCs and injured neurons, respectively. The quantitative evaluation of GFAP staining was performed using image analysis system Lucia.
We found that activation of SGCs was significantly increased in bilateral TG after unilateral IONL. Although injured neurons of infraorbital nerve lie in the maxillary region, a moderate activation of SGCs was also detected in the mandibular region of TG. A low activation of SGCs was found in ipsilateral TG removed from sham-operated rats.
Our results revealed that unilateral IONL affects the activation of SGCs not only around injured (ATF3+) but also ATF3- neurons of TG.
Satellite glial cells are thought to play a role in neuronal activation in response to peripheral nerve injury leading to neuroinflammation and neuropathic pain.

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Mouse 3T3 fibroblasts under the influence of fibroblasts isolated from stroma of human basal cell carcinoma acquire properties of multipotent stem cells

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Many types of cancer involve cells with multipotent stem cells properties (Nanog, Oct4 positive). Their function is not realy known, but it mean poor prognosis for the patient. Similarly,.several types of tumours contain sporadic bone or cartilage tissues inside the tumour, mainly in the stroma. It refer on atypical differentiation of stem cells. Low differentiation level of this cells can be conditional by cancer microenvironment especially stroma cells. To verify pro-stem potentional cancer associated fibroblasts, we detect their influence on 3T3 mouse fibroblasts (can divide mouse and human cells). Results show that 3T3 fibroblasts under influence of cancer associated fibroblast from basal cell carcinoma were Nanog, Oct4 and Sox2 positive. Additional, 3T3 get stem cell properties alike mesenchymal stem cell and be able diferentiate in couple cell types – adipocytes, osteocytes and chondrocytes. With mRNA analysis (Illumina) we designed growth factors, which may can play a role in acquire features of multipotent 3T3 fibroblasts. Last we can claim, that fibroblasts isolated from human basal cell carcinoma are capable elevate stem potential co-cultured fibroblasts, which has an effect on biological properties of cancer.

Key words: stem cell, cancer stroma, cancer associated fibroblasts, 3T3 fibroblasts

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Comparative anatomy – microscopic study of the spleen structures.

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The spleen belongs functionally to the vascular system, but it is also the largest lymphoid organ included in the blood circulation. It represents the main point of defense against infection and harmful substances penetrating into the circulation. The spleen of our studied species (fallow deer and mouflon) represents the spleen of animals which escape is virtually the only means of defense.

The aim of the work was to observe the microscopic structure of the spleen in animals and their comparison. Animal samples were procured from a hunting society Turček in Bratislava. For purposes of comparative anatomy of the spleen, we used a samples of fallow deer (6 pieces) and mouflon (3 pieces). Animal samples were stained by hematoxylin and eosin, chloroacetate esterase and impregnated according to Gomori for reticular fibers.

The results of spleen observation in both species compared with human show a striking difference in the content of reticulin fibers and clarity of marginal sinuses. Both are sinusoidal-type of spleen as presented in man. Studied animal spleens differ in mild details, on the vascularization and innervation. Most striking difference, compared with man, the apparent lack of reticulin fibers in the walls of the sinuses of red pulp in both species. Extramedullary haematopoiesis, we clearly have not seen in both species but we can not exclude it.

We assume that in both cases, the spleen serves as a blood reservoir and plays an important role in clearing the blood of foreign particles. Described results will find application in pedagogical work in teaching histology and microscopic anatomy, not only for veterinary and agricultural schools, but also at medical schools.

Quantification of vasa vasorum in human varicose great and small saphenous vein

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Our aim was to (i) establish a technique of histological quantification of vasa vasorum in varicose human great (GSV) and small (SSV) saphenous veins and (ii) to describe the distribution of the vasa vasorum within the venous wall.

Great (n=11) and small (n=5) saphenous veins (length of 15-40 cm) were collected from patients undergoing stripping due to chronic venous insufficiency. The veins were divided into 5 cm long segments. In total, 92 tissue blocks were taken to trace the variability of density and distribution of vasa vasorum in the proximodistal direction. The endothelium was detected by immunohistochemistry (von Willebrand factor). Using stereology, we quantified the number of microvessel profiles per section area, as well as the relative distance of microvessels from the outer border of the adventitia.

Quantitative histological analysis proved that the enhanced ingrowth of vasa vasorum from the tunica adventitia into the venous wall is a universal finding along most of the length of these pathologically changed veins. Although the density of the vasa vasorum increased with decreasing thickness of the more distal segments, this vasa vasorum ingrowth was found in proximal as well as in distal segments of both GSV and SSV. Our findings suggest that unlike in healthy saphenous veins with prevailing longitudinal vasa vasorum, the vasa vasorum network in varicose veins underwent ingrowth and remodelling to loops of microvessels without distinct directionality or preferential orientation.

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The changes in jejunal mucosa cell populations during mesenteric ischemia-reperfusion injury in rats

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The progress of the changes in populations of the cells in jejunal mucosa in the course of mesenteric ischemia-reperfusion injury at different time periods was investigated. Mesenteric ischemia lasting 1 hour followed by 1 hour of reperfusion caused significant disintegration of the mucosa, reduction of the muscular layer and diminution of the wall thickness. The loss of epithelium included, except enterocytes, Paneth cells ($p<0.05$) and goblet cells ($p<0.001$), particularly. Paradoxically, increasing numbers of serotonin-producing cells ($p<0.01$) and the beginning of regenerative processes expressed by significantly higher proliferation ($p<0.001$) were recorded in the epithelium during this period. Disintegration of connective tissue and massive degranulation of serotonin–positive cells were found in the *lamina propria* ($p<0.001$). After 24 hours of reperfusion, restitution of the mucosa was observed, expressed by normal villous morphology and re-epithelialization. However, some parameters were still significantly affected, even more than in the acute phase of reperfusion. In the epithelium, decreased numbers of Paneth cells ($p<0.001$) and increased population of serotonin-producing cells ($p<0.01$) were detected. The greatest proliferation of the connective tissue cells ($p<0.001$) and intensified reduction of the muscular layer ($p<0.01$) were observed in this reperfusion period too. After 30 days of reperfusion moderate damage remained, but only the increased number of Paneth cells within the epithelial lining of the crypts ($p<0.001$) and decreased number of serotonin–producing cells ($p<0.05$) in the *lamina propria* were significant.

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Prognostic significance of LC3 autophagic protein in non-small cell lung cancer

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The microtubule associated protein 1 light chain 3 (LC3A) is an indispensable component in the autophagic machinery. Evaluation of various prognostic factors, the role of autophagy a self-degradative process involved in the turnover of cytoplasmic material, remains unexplored in lung malignancy.
Autophagic activity was studied in non-small cell lung carcinoma (NSCLC) patiens. We used the LC3A antipody and a standard immunohistochemical techniqe. Autophagic activity was correlated with clinical and pathological parameters. Immunuhistochemical examination revealed three paterns of autophagic activity (diffuse cytoplasmic, perinuclear cytoplasmic and „stone-like“ structures (SLS). A high count of SLS was associated with tumor-specific structures and had a relationship to prognosis of behaviour of tumors.

Literature:

Karpathiou G et al. (2010) LC3A autophagic aktivity and prognostic signifikance in non-small cell lung cancer. CHEST December 9, 2010 101831published ahead of print December 9, 2010

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Airway remodeling in experimentally provoked bronchial asthma

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Marks of chronic alterations known as remodeling appear in bronchial walls of patients suffering from the bronchial asthma. While the morphological changes of adult human bronchial walls have been thoroughly described, fewer papers exist about the bronchial remodeling in small children and even lesser about the changes in laboratory animals.

Rats of the Brown Norway (BN) strain, which are especially responsive to sensitization by various allergens and develop the state that resembles the human bronchial asthma, were used. In the currently presented part of the study, the adult animals were sensitized by repeated intraperitoneal injections of ovalbumin (OA). During following 2 weeks, the rats regularly inhaled the aerosolized OA in low concentrations. Two control groups were bred simultaneously. The first of them was injected and inhaled by saline (S), the second group was untreated (C). Functional respiratory parameters and specific serum IgE levels were measured. Lungs were processed for the light microscopy and morphometrically examined.

The bronchial walls of the OA group were remodeled. The total and inner (mucosal + muscular layer) wall areas were significantly increased compared to groups S and C. The OA animals had significantly higher IgE levels and respiratory rate, too. The other respiratory and morphometric parameters did not differ significantly.

The first part of the proposed study confirmed the bronchial sensitivity of BN rats and constituted a base for further experiments comparing bronchial reactivity and remodeling of adult and young animals. We expect more pronounced changes in young animals imitating our paediatric patients.

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Endoplasmic reticulum stress in ischemic small intestine

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The influence of small intestine reperfusion, after the failure of compensatory mechanisms, usually leads to cell death through apoptosis or necrosis. IR stress significantly affects the endoplasmic reticulum (ER), which dysfunction induces responses through activation of kinases which stimulate anti-apoptic mechanism through Grp78 (BIP) or pro-apoptic mechanism by activation of Gadd153 (CHOP).

We analyzed the effects of IR injury of the small intestine wall of rats after 1 hour ischemia and subsequent reperfusion times in periods 1h, 24 h and 30 days (R1 to R30) to expression of pro (Gadd153) and anti (Grp78) apoptic genes on the mRNA level. In addition we have been monitoring the levels of corresponding proteins as well. After isolation of RNA and reverse transcription into cDNA, we were measuring changes in gene expression by PCR. For the determination of the protein levels Western blot was used with consequent chemiluminiscent detection.

We found that increased mRNA level of Gadd153 in R1 group (37 ± 11 % higher than controls) was closely related with the delay in protein production, which maximum (55 ± 10,2 % higher than controls) was shifted to 24 hours reperfusion time. Opposite to this we detected maximal level of protein Grp78 in R1 group (120 ± 13,6 % higher than controls) probably because of its release from ER membrane after 24 hours of reperfusion.

There is still no effective approach to the treatment of affected ischemic intestine tissue, to stop the processes with could eventually lead to MODS. Therefore it is necessary to study intestinal ischemia-reperfusion injury at the molecular level and try to find new therapeutic routes to protect affected tissue.

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Development of the limb girdles

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The development of the limbs has been studied in a lot of detail but the development of structures that anchor the limbs to the trunk has been neglected. By using classical microsurgical manipulations, gene expression and transgenic murine and avian models we described novel developmental mechanisms of the limb girdles.
In the pelvic region we found that the sphincters and perineal muscles develop from the musculature of the pelvic limb via a novel mechanism „in-out". The myogenic cells migrate first from the somites „in"-to the pelvic limb bud and later move „out" of the base of the limb bud towards the adjacent midline orifices (cloacal/urethral, anal). Experimental limb bud ablation or genetic disruption of myogenic cell migration result in the absence of these muscles.
Analogous „in-out" mechanism is also deployed during the development of pectoral and latissimus dorsi muscles. They anchor the pectoral limb to the axial skeleton and thus their continuity with the limb is not lost as in the case of the perineal muscles and the pelvic limb. Also other girdle muscles use this developmental mechanism, however the deepest ones which attach the scapula to the axial skeleton (anterior serrate, rhomboids and levator scapulae muscles) develop directly from the somites. It is worth noting, that the medial margin of the scapula where these four muscles attach, develops also from the somites.
Defects of the pectoral limb caused by conditional ablation of *Tbx5* gene documented its intimate relationship to the development of the diaphragm and the sternum. This is the first genetic evidence supporting the theory that sternum, which in evolution served primarily as the attachment of the pectoral muscles, is a part of the limb girdle.

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Myoid cells of the human thymus – their possible development and function

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The myoid cells were first noted in 1888 by S. Mayer, who saw them in the frog thymus; they were described as long, spindle-shaped cells showing distinct striations and closely resembling skeletal muscle fibers. Their origin and biological role is not yet clear. Myoid cells hold similar characteristics as epithelial cells, therefore they were initially assumed to be of myoepithelial origin, and later on speculations about their non-thymic mesodermal origin were suggested. Today, the hypothesis of Nakamura and Ayer-Le Liére (1986) is the most accepted, where they suggest a neuroectodermal origin of the myoid cells. That is, they are derivatives of the neural crest.

Thymic myoid cells produce many proteins, which are specific for skeletal muscle fibers, e.g. myosin, troponin T, desmin, rapsyn and utrophin. Wakkach et al. (1999) demonstrate the excessive production of TNF- and IL-8 in tissue cultures of myoid cells. These factors presumably protect thymocytes from apoptosis. Myoid cells play an important role in the differentiation of T cells. The protective effect of myoid cells against apoptosis of thymocytes is decribed also by Panse and Berrih-Aknin (2005). Myoid cells have, similarly to thymic epithelial cells, surface receptors for acetylcholine. Thereby, it is presumable that both cell types can play an initial role in the auto-sensitization during the autoimmune disease myasthenia gravis.

In our study we studied the imunohistochemical chyrycteristics of thymic myoid cells in thymuses from children with congenital heart defects removed after surgical procedure (monoclonal antibodies again myosin, alfa-actin and desmin). We describe the correlation between occurrence of thymic myoid cells and type of congenital anomaly of heart (focused on heart defects derived in disrupted migration of neural crest-derived cells), as well as between number of thymic myoid cells and expression of anti-apoptotic protein Bcl2. Our preliminary results show, that thymic myoid cells can protect thymocytes from apoptosis and are derived from embryonic neural crest.

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Immunohistochemical evaluation of new possible predictive marker in breast carcinoma

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Determination of prognostic and predictive factors represents an important part of breast carcinoma diagnostic and therapeutic management. Because both traditional morphological (tumor size, carcinoma type, histological grade, lymph node status) and biological parameters (analysis of hormone receptors and HER2) do not always lead to exact prediction of prognosis and/or therapeutical response, new approaches are researched to allow more precise patient´s stratification. Expression of antioxidant proteins in tumour cells has been assessed as predictive factor of response to cytotoxic treatment. Among them, emphasis is placed on Pi class glutathione S-transferase (GSTP1).

The aim of our work was to determinate the expression of GSTP1 in 99 samples of invasive breast carcinoma and compare results versus normal breast cells. Immunohistochemical method was chosen for the detection of GSTP1.

Normal breast tissue showed strong positive GSTP1 expression in all cases. It could confirm protective function of this enzyme in healthy breast tissue. We have found that majority (66%) of breast carcinomas shows GSTP1 positivity (nuclear and cytoplasmic immunoreactivity). Overexpression of GSTP1 outlines an increase of resistance to chemotherapy in positive samples. It is expected that GSTP1 positive tumours would show a poorer prognosis than GSTP1 negative ones.

Immunohistochemistry is a useful method for investigating the expression and cellular localization of GSTP1 within tumours. It may be applied to a routine clinical test and it can serve as a potential useful marker for resistance to chemotherapy.

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VEGF expression in healthy human skin

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Expression of the vascular endothelial growth factor (VEGF) was evaluated in healthy human skin from various topographical regions. Skin samples were taken from the cadavers devoted to the anatomical dissections. Specimens were standardly processed – fixed in formalin, embedded into the paraffin. VEGF expression was detected by immunohistochemical method with monoclonal mouse anti-human VEGF clone VG1. Intensity of the immunoreactivity and the percentage of the labelled cells were evaluated by the semiquantitative method. χ^2 test was used for the statistical analysis. No statistically significant differences in VEGF expression depending on the topographical region were found neither in epidermis, nor in dermis and endothelial cells of subpapillary plexus. However in the general comparison of the VEGF expression in epidermis, dermis and endothelial cells of the subpapillary plexus revealed statistically significant differences ($p<0.01$). Strongest VEGF expression as for the intensity of the immunoreactivity and the percentage of the labelled cells (more than 10%) was seen in the endothelial cells of the subpapillary plexus, moderate intensity of the immunoreactivity and lower percentage of the labelled cells (less than 10%) in the epidermis and only mild and rare expression was observed within the fibroblasts of the dermis. Normally the VEGF expression in the skin is low compare to the organs like lungs, kidneys or heart but after the tissue injury or in psoriatic plaques it is markedly increased.

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Pre-screening of potential anti-cataractous agents: novel pyridoindole derivatives tested *in vitro*

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WHO statistics consider cataract as the most common cause of blindness worldwide. Unfortunately, there is no available treatment for cataract, the only cure is surgery. The pharmacological intervention of degenerative eye lens proteins changes would be desirable. The main goal of this work was to test the efficacy of novel pyridoindole derivatives - (-)-cis-2,3,4,4a,5,9b-hexahydro-2,8-dimethyl-1H-pyrido[4,3-b]indole (Stobadine) and (2-benzyl-2,3,4,5-tetrahydro-1h-pyrido[4,3-b]indole-8-yl)-acetic acid (ARI-BE) on selenite-induced *in vitro* model of cataractogenesis. Lens, acquired from male Wistar rats, were long-term cultivated *in vitro.* The opacity was formed by the addition of sodium selenite into the cultivation medium M-199 (Sigma-Aldrich, Germany). At particular time points of the experiment (0, 24, 48 and 72 hours) lens were examined directly on the U-96 plate by a spectrophotometer (Infinite M200, Tecan, Switzerland) with *multiple reads per well* function, mode 7x7 at 420 nm. These measurements correlated well with picture digital analysis (morphometric software Analysis, Olympus, Japan) of the lens scanned (CanoScan 8800F, Canon, Japan). Both novel pyridoindole derivatives tested showed a tendency to decrease the rate of eye lens opacification such render them to be promising molecules for further studies. This work contributed to the search for pharmacological substances beneficial in preventing and/or delaying deleterious cataractous changes of eye lens proteins.

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Mitotic activity in the 5-day chick embryonic kidney

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Embryonic kidney of the 5-day chick embryo consists of several populations of nephrons at different stages of development. We attempted to reveal laws of their growth, especially changes of the proliferative activity in both, tubules and glomeruli, in dependence of the nephron development. The nephrons originate gradually in a cranio-caudal sequence and appear in layers lined up in a ventro-dorsal order.

We studied the proliferative actvity using the colchicine method. Embryos were treated by 2.8 µg of colchicine in 10 µl Ringer solution administered intraamniotically for 120 min. Mitotic index (proportion of dividing cells), MI, was measured in controls, the stathmokinetic index, SKI, in the colchicine-treated embryos, with a system for stereological analysis C.A.S.T Grid (2000 Olympus, Denmark).

Both indexes showed higher values in the younger, dorsally located nephrons. The SKI was always higher than MI, regardless of the nephron position and tissue examined. A cranio-caudal gradient of the proliferative activity manifested in the SKI of both epithelia, tubular and glomerular, in the older, ventral nephrons. The most important was finding of much higher SKI in tubules of embryos at stage HH 25 (8.6 – 9.0 ventral and 8.7 – 12.5 dorsal nephrons) in comparison with embryos by several hours older (HH 26-27 : 3.6 - 5.2 and 4.6 – 5.3 in analogous categories). Also a duration of mitosis showed a distinct prolongation between stages HH 25 (in tubules of the middle portion of the kidney: 31 - 46 min) and HH 26-27 (59 – 86 min in the same location). Values of SKI indicated a presence of ventro-dorsal gradient in the tissues of ventral nephrons, but not in the dorsal ones.

We conclude, that the proliferative activity in mesonephric nephrons at the day 5 still reflects both gradients of the nephron induction showing gradual decrease of the cell division in older nephrons.

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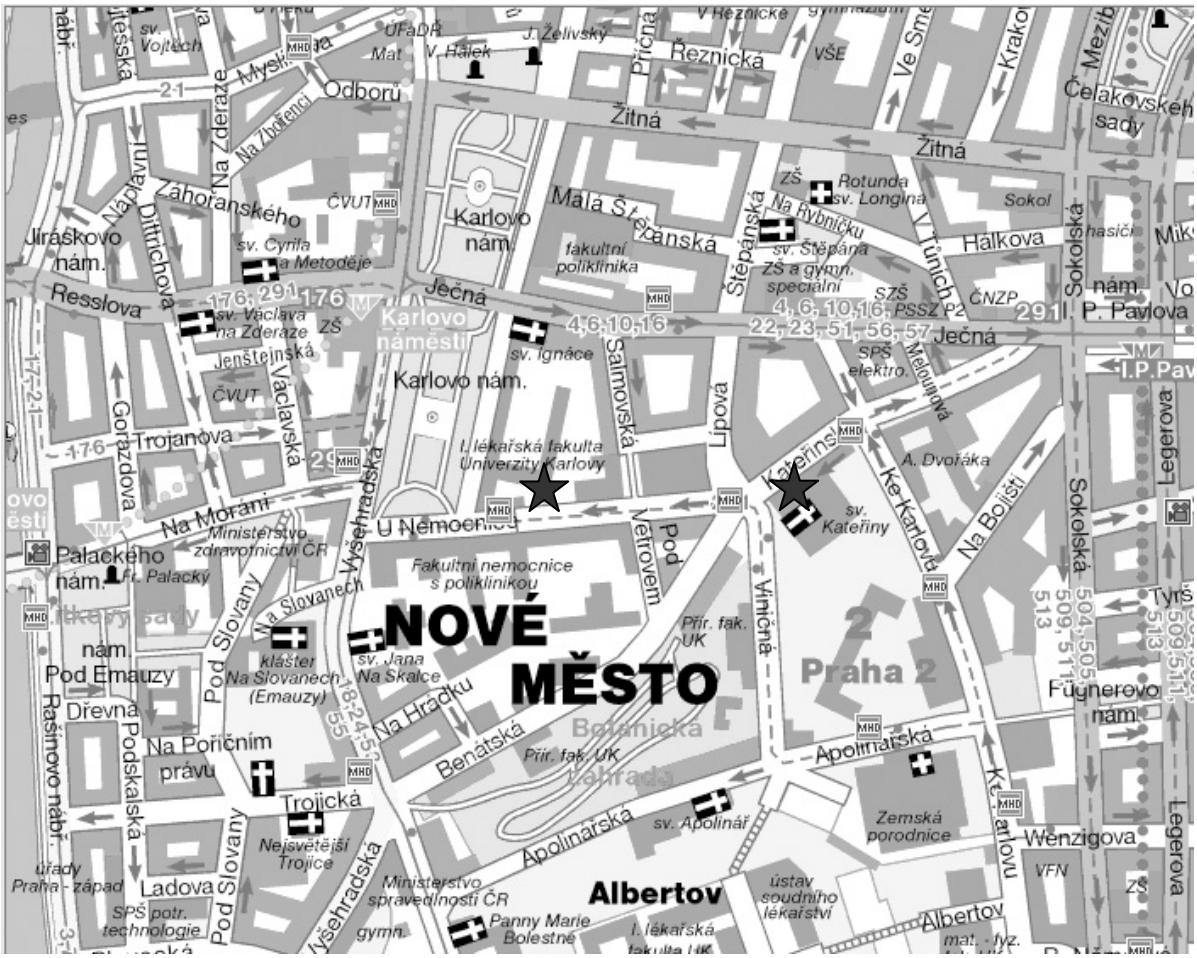
Maps:

Institute of Anatomy, U Nemocnice 3, Praha 2
By tram No. 4, 6, 10, 16, 22, 23, station Štěpánská; by tram No. 4, 10, 16, 18, 24 station Moráň or Karlovo náměstí
By Metro: line C (red) - station I.P.Pavlova; line B (yellow) - station Karlovo náměstí

The St. Catherine Church, entry in the corner of Kateřinská and Viničná st. - walking distance from Institute of Anatomy (cca 150 m)



The New Town Hall " (Novoměstská radnice), Karlovo nám. 1/23, Praha 2
By tram No. 3, 6, 16, 18, 21, 22, 24 station Karlovo náměstí or by tram No. 3,9,14,24, station Lazarská.
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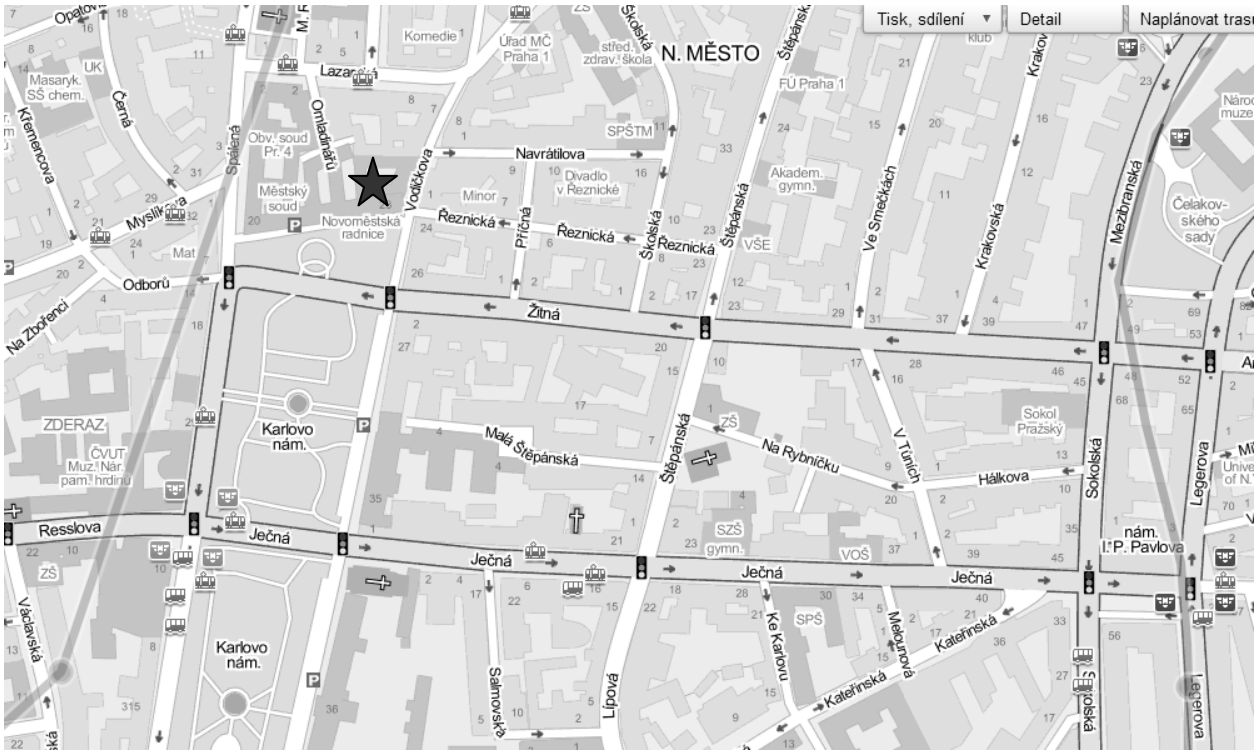
The New Town Hall is in walking distance from Institute of Anatomy (cca 400 m)



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