

# European Journal of Anatomy

December, 2013  
Vol. 17 - Supplement 1

Official Journal  
of the Spanish  
Society of Anatomy



## XXVI CONGRESS OF THE SPANISH ANATOMICAL SOCIETY

### ABSTRACTS



**Indexed in:**

**EMBASE/Excerpta Medica**

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**Fuerteventura, 5-7 September, 2013**



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**ABSTRACTS**

FUERTEVENTURA (CANARY ISLANDS), 5-7 SEPTEMBER, 2013



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# SCIENTIFIC PROGRAM

Thursday, 5th SEPTEMBER 2013

8.30-9.00: Registration

9.00-10.30: Opening Ceremony

9.30-10.30: **FOETAL ONSET HYDROCEPHALUS IS A NEURAL STEM CELL PATHOLOGY. OPENING THE AVENUE FOR STEM CELL THERAPY**

*Esteban M. Rodriguez.* Instituto de Anatomía, Histología y Patología, Universidad Austral de Chile, Valdivia, Chile

10.30-11.00: Coffee Break

11.00-13.00: Oral Communications SESSION 1: Embryology and Teaching in Anatomy

**Chairmen:** Prof. *José Ramón Sañudo Tejero* and Prof. *Miriam González Gómez*

E-O-01

## **MOLECULAR BASIS OF CEREBELLUM DEVELOPMENT**

*Andreu, A., Mecklenburg, N., Martínez, S., Echevarría, D.\** Departamento de Histología y Anatomía (UMH), Instituto de Neurociencias de Alicante, (UMH-CSCIC), Alicante, Spain

E-O-02

## **DEVELOPMENT OF THE CILIARY BODY**

*Peces, M.D.<sup>1</sup>, De la Cuadra, C.<sup>2</sup>, de Moraes, L.O.C.<sup>3</sup>, Arráz, L.A.<sup>2</sup>, Herrera, M.E.<sup>1</sup>, Mérida, J.R.<sup>2\*</sup>*

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<sup>3</sup>Department of Orofacial Sciences and Pediatrics. University of California. San Francisco (USA)

E-O-3

## **VARIATIONS IN THE DISTRIBUTION OF AQP1 IN DIFFERENT CASES OF CONGENITAL HUMAN HYDROCEPHALUS**

*Castañeyra-Ruiz L.<sup>1,3</sup>, González-Marrero I.<sup>1</sup>, González-Toledo J.M.<sup>1</sup>, Castañeyra-Ruiz A.<sup>2</sup>, Redondo A.<sup>2</sup>, Perez-Molto J.F.<sup>4</sup>, Hernandez-Diaz E.R.<sup>1</sup>, Soto M.<sup>2</sup>, Castañeyra M.<sup>2</sup>, Curbelo S.<sup>2</sup>, Periz R.<sup>4</sup>, Castañeyra-Perdomo A.<sup>1</sup>, Carmona-Calero E.M.<sup>1,2</sup>.*

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E-O-4

## **AQUAPORIN-4 EXPRESSION IN THE PARIETAL TELEENCEPHALIC NEUROEPITHELIUM OF CASES OF CONGENITAL HUMAN HYDROCEPHALUS**

*Castañeyra-Perdomo A.<sup>1</sup>, Castañeyra-Ruiz L.<sup>1,3</sup>, González-Marrero I.<sup>1</sup>, González-Toledo J.M.<sup>1</sup>, Castañeyra-Ruiz A.<sup>2</sup>, Redondo A.<sup>2</sup>, Perez-Molto J.F.<sup>4</sup>, Curbelo S.<sup>2</sup>, Hernandez-Diaz E.R.<sup>1</sup>, Soto M.<sup>2</sup>, Castañeyra M.<sup>2</sup>, Carmona-Calero E.M.<sup>1,2</sup>.*

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DA-O-01

**USE OF AN INTERACTIVE AUDIENCE RESPONSE SYSTEM IN MEDICINE AND PHYSICAL THERAPY**

*Moreno-Cascales, M.<sup>1</sup>, González- Sequeros O.<sup>1\*</sup>, López González, L.<sup>1</sup>, López Jiménez J. J.<sup>2</sup>, Fernández Alemán, J.L.<sup>2</sup>*

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DA-O-02

**PRODUCTION AND DELIVERY OF MORPHOLOGICAL KNOWLEDGE BY DIGITAL DISSECTION OF BODILY AND EMBRYONIC STRUCTURES**

*Doñate, F., Bilbao, A., Azkue, J.J.*

Department of Neurosciences, School of Anatomy and Dentistry, University of the Basque Country.

DA-O-03

**ROLE OF EPONYMS IN CURRENT ANATOMY**

*Kacblík, D.<sup>1,2</sup>, Baca, V.<sup>1,2</sup>*

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<sup>2</sup>Department of Health Care Studies, College of Polytechnics Jihlava, Czech Republic

13.00-14.00: Posters' Discussion SESSION 1: Neuroanatomy, Embryology and Teaching in Anatomy

14.00: LUNCH

16.00: SOCIAL ACTIVITIES

## Friday, 6th SEPTEMBER 2013

9.00-11.00: Oral Communications SESSION 2: Systemic and Clinical Anatomy

Chairmen: Prof. *Damián Sánchez Quintana* and Prof. *Ibrahim González Marrero*

ACyS-O-01

**CORONA MORTIS REVISITED**

*Kacblík, D.<sup>1,2</sup>, Vymetalová, K.<sup>1</sup>, Laboš, M.<sup>3</sup>, Bába V.<sup>1,2</sup>*

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ACyS-O-02

**PRESURGICAL 3D GAIT ANALYSIS FINDINGS IN FOUR CHILDREN WITH SPASTIC CEREBRAL PALSY**

*Smith, V. M.<sup>1\*</sup>, Postigo, S.<sup>2</sup>, Postigo, M.J.<sup>3</sup>, Prado, M.<sup>2</sup>, Núñez, M. J.<sup>3</sup>, Ros, B.<sup>4</sup>, Fernández, V.<sup>3</sup>*

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ACyS-O-03

**FACIAL REINNERVATION BY MASSETERIC NERVE: A PRELIMINARY ANATOMICAL STUDY**

*Alcocer, M., Marañillo, E., Sañudo, J.R., Vázquez, T., Pascual-Font, A.\**

Department of Human Anatomy and Embryology I. School of Medicine. Complutense University of Madrid



ACyS-O-04

**THE THIEL'S EMBALMED CADAVER AS A IDEAL MODEL FOR ADVANCED ENDOSCOPIC SURGERY PROCEDURES TRAINING**

*Zubiaga, L.<sup>1</sup>, Sánchez del Campo, F.<sup>1\*</sup>, Abad, R.<sup>2</sup>, Enriquez, P.<sup>2</sup>*

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ACyS-O-05

**ALVEOLAR CHANGES AFTER THE APPLICATION OF PROTECTIVE MECHANICAL VENTILATION WITH DIFFERENT CONCENTRATIONS OF OXYGEN IN HEALTHY RATS**

*Domínguez, D.<sup>1\*</sup>, Martín-Barrasa, J.L.<sup>2</sup>, López, M.<sup>3</sup>, Villar, J.<sup>2,4</sup>, Rancel, N.<sup>3</sup>, Valladares, F.<sup>3,4</sup>*

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ACyS-O-06

**ROLE OF THE TYPE 2 ALVEOLAR EPITHELIAL CELLS IN AN EXPERIMENTAL MODEL OF ACUTE LUNG INJURY**

*López, M.<sup>1\*</sup>, Domínguez, D.<sup>2</sup>, García, S.<sup>3</sup>, Rancel, N.<sup>1</sup>, Valladares, F.<sup>1,4</sup>*

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11.00-11.30: Coffee Break

11.30-12.30: **DEVELOPMENT OF THE HUMAN CEREBRAL CORTEX**

Gundela Meyer. Departamento de Anatomía, Facultad de Medicina, Universidad de La Laguna

12.30-13.30: Posters' Discussion SESSION 2: Systemic and Clinical Anatomy

13.30-14.00: Commercial Presentation

Elsevier

14.00: LUNCH

16.00: SOCIAL ACTIVITIES

**Saturday, 7th SEPTEMBER 2013**

9.00-11.00: Oral Communications SESSION 3: Neuroanatomy

Chairmen: Prof. *Concepción Reblet López* and Prof. *Domingo Afonso Oramas*

N-O-01

**HUMAN CUTANEOUS MEISSNER CORPUSCLES DEVELOP POSTNATALLY**

*Feito, J., Viña, E., Cobo, T., López-Muñiz, A., García-Suárez, O., Vega, J.A. \**

Dpto de Morfología y Biología Celular, Universidad de Oviedo

N-O-02

**OCCURRENCE OF MECHANOPROTEINS IN CUTANEOUS MECHANORECEPTORS OF *Maccaca fasciculata***

*Cabo, R.<sup>1\*</sup>, Gálvez, A., San José, I.<sup>2</sup>, Pastor, F.<sup>2</sup>, López-Muñiz, A.<sup>1</sup>, Vega, J.A.<sup>1</sup>*

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<sup>2</sup>Departamento de Anatomía y Radiología, Universidad de Valladolid

N-O-03

**ARE THE TRPC2 POSITIVE CELLS IN THE OLFACTORY EPITHELIUM OF ADUL ZEBRAFISH THE EQUIVALENT TO THE MAMMALIAN VOMERONASAL ORGAN?**

*Viña, E.<sup>1</sup>, Parisi, V.<sup>2</sup>, Fernández, B.<sup>1</sup>, Cobo, J.L.<sup>1</sup>, Germanà, A.<sup>2</sup>, Vega, J.A.<sup>1</sup>, García-Suárez, O.<sup>1\*</sup>*

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N-O-04

**THE PROTEIN INSULIN-RECEPTOR-SUBSTRATE 2 (IRS2) IS ESSENTIAL FOR THE INTEGRITY OF THE HIPPOCAMPUS AND ITS ABSENCE COULD INDUCE HIPPOCAMPAL NEURODEGENERATION**

*Carretero, M.<sup>1</sup>, Carretero-Hernández, M.<sup>2</sup>, Blanco, E.J.<sup>2,3</sup>, García-Barrado, M.J.<sup>3,4</sup>, Iglesias-Osma, M.C.<sup>3,4</sup>, Catalano, L.<sup>3</sup>, González, M.L.<sup>3</sup>, Herrero, J.J.<sup>5</sup>, Burks, D.J.<sup>6</sup> y Carretero, J.<sup>2,3\*</sup>*

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N-O-05

**LENGTH AND DIAMETER OF AXON INITIAL SEGMENT OF PYRAMIDAL NEURONS IN LAYER II AND LAYER III OF ANTERIOR CINGULATE CORTEX OF THE RAT BESIDES NUMBER AND DISTRIBUTION OF INCOMING AXO-AXONIC SYNAPTIC BOUTONS**

*Bueno-López, J.L.\**, Chara, J.C., Reblet, C.

Department of Neurosciences, School of Medicine and Dentistry, The University of the Basque Country (UPV/EHU)

N-O-06

**IS THE ARYTENOID MUSCLE SUPPLIED BY THE RECURRENT AND THE SUPERIOR LARYNGEAL NERVES?**

*Cubillos, L.E., Pascual-Font, A., Sañudo, J.R.\**, Vázquez, T., Valderrama-Canales, F., Maranillo, E.

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11.00-11.30: Coffee Break

11.30-12.30: CHOROID PLEXUS-CSF NEUROPEPTIDE INVOLVEMENT IN INTRACRANIAL PRESSURE REGULATION: IMPLICATIONS FOR THE PUTATIVE INTRACRANIAL HYPERTENSION OF SPACEFLIGHT.

*Conrad E. Johanson*. Dept. of Neurosurgery, Alpert Medical School at Brown University, Providence, Rhode Island, USA.

12.30-13.30: 16th General Meeting of the SAE

13.30-14.00: THE ANATOMICAL DISSECTION AS FACILITATOR OF THE MEDICAL DEVELOPMENT. THE CONSOLIDATION OF THE VESALIUS' ANATOMY IN SPAIN

*Ricardo Vázquez Rodríguez*. Former Editor in Chief of European Journal of Anatomy

14.00: LUNCH

16.00: SOCIAL ACTIVITIES

21.30: CLOSING CEREMONY AND DINNER

# MAIN CONFERENCES



## C1: FOETAL ONSET HYDROCEPHALUS IS A NEURAL STEM CELL PATHOLOGY. OPENING THE AVENUE FOR STEM CELL THERAPY

Esteban M Rodríguez<sup>1</sup>, Maria M Guerra<sup>1</sup>, Karin Vío<sup>1</sup>, Maria C Jara<sup>1</sup>, Alexander Ortloff<sup>1</sup>, Nicole Lichtin<sup>1</sup>, Roberto González<sup>1</sup>, César González<sup>1</sup>, Antonio J Jiménez<sup>5</sup>, Maria D Dominguez-Pinos<sup>5</sup>, Sara Rodríguez<sup>1</sup>, Eduardo Ortega<sup>2</sup>, Jaime Jaque<sup>2</sup>, Francisco Guerra<sup>3</sup>, Deborah A Sival<sup>4</sup>, Wilfred F A den Dunnen<sup>4</sup>, José M. Pérez-Fígares<sup>5</sup>, James P McAllister<sup>6</sup>, Conrad Johanson<sup>7</sup>.

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**Background.** It is now understood that hydrocephalus is not only a disorder of CSF dynamics, but also a brain disorder, and that derivative surgery does not resolve most aspects of the disease. The common history of congenital hydrocephalus and brain maldevelopment starts early in the embryonic life with the disruption of the ventricular (VZ) and subventricular (SVZ) zones. The VZ contains neural stem cells (NSCs) while the SVZ harbors neural precursor cells (NPCs). Evidence obtained from hydrocephalic human fetuses and hydrocephalic mutant animals indicates that a cell junction pathology in NSCs and NPCs leads to the VZ/SVZ disruption. The present investigation was aimed: (1) to investigate the translocation of NSCs and NPCs into the hydrocephalic CSF (hyCSF), to harvest and culture them into neurospheres. (2) to establish how compatible the hyCSF might be to host grafted NSCs; (3) to explore strategies that might help repair the loss of NSCs, including intraventricular neural stem cell grafting.

**Materials and Methods.** For aim 1: Cells obtained from the VZ/SVZ of non-hydrocephalic HTx rats at postnatal day 1 (PN1), and from the CSF of hydrocephalic HTx rats at PN1 were cultured for six days (6DIV) to test their capacity to form neurospheres. The last DIV neurospheres were exposed to BrdU. They were processed for immunofluorescence using antibodies against BrdU (proliferation), nestin, beta3tubulin, and GFAP (differentiation), caveolin 1 (functional plasma membrane proteins), and N-cadherin (adhesion junctions). For aim 2: Protocol 1. NSCs and NPCs from the telencephalon of PN1 and PN7 of non-hydrocephalic HTx rats were cultured to obtain neurospheres. After 7 days in vitro (DIV) they were transferred to a culture medium free of growth factors and containing normal or hydrocephalic

CSF, for two weeks. Protocol 2. CSF from hydrocephalic HTx rats PN1, plus 30% of neurosphere medium, was incubated at 37°C. Protocol 3. Pure CSF from hydrocephalic HTx rats PN1 was incubated at 37°C. for aim 3: Neurospheres from the telencephalon of non-hydrocephalic rats were grafted into the lateral ventricle of PN7 hydrocephalic rats.

**Conclusions:**

(1) The hyCSF contains NSCs and NPCs that under culture conditions proliferate to form small and large neurospheres, respectively.

(2) These neurospheres express the same pathology as the VZ cells in the living hydrocephalic animal or human.

(3) Neurospheres obtained from NSCs/NPCs present in the hyCSF may be used to investigate cell and molecular alterations underlying the disease. Thus, the inability to obtain human brain biopsies for diagnostic and research reasons may be overcome. This might open a new avenue in clinical research.

(4) The CSF of normal and hydrocephalic animals promotes the differentiation of the NSCs/NPCs of neurospheres into neurons and astrocytes.

(5) The hyCSF of hydrocephalic subjects appears as a friendly medium to host grafted neurospheres.

(6) Neurospheres from the telencephalon of non-hydrocephalic rats grafted into the lateral ventricle of PN7 hydrocephalic litter mates disassemble and NSCs populate the disrupted ventricular zone. The miracle of repairing the disruption of the ventricular zone, key phenomenon leading to hydrocephalus and abnormal neurogenesis, seems possible.

No conflict of interest to declare. Supported by Fondecyt 1111018 (EMR) and Hydrocephalus Association (EMR, PM, CJ).

## C2: DEVELOPMENT OF THE HUMAN CEREBRAL CORTEX

Gundela Meyer

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The human cerebral cortex is a highly complex structure which requires for its development a long gestation period and multiple coordinated steps of cell generation, migration and differentiation. We focus here on some peculiarities of human cortex development that are distinct from those described in the mouse, at present the most widely used animal model. We describe the stratification of the pallial wall, in particular its transient compartments – marginal zone, subplate, and subventricular zone. The neuronal populations of the marginal zone are Cajal-Retzius (CR) cells and the neurons of the subpial granular layer (SGL). CR cells derive from the cortical hem system at the boundaries of the telencephalon and the choroid plexus; they are the main source of reelin, a protein essential for radial migration of excitatory neurons and laminar positioning. The protracted survival of human CR cells may be related to the expres-

sion of the anti-apoptotic protein DeltaNp73. The SGL is characteristic of human brain and not present in rodents; it is formed by GABAergic interneurons that migrate tangentially below the pia from its origin in the basal forebrain to their destination in the cortex, where they populate the superficial layers.

The subplate is another compartment that is highly differentiated in human, especially in the second half of gestation, and characterized by excitatory, Tbr1+ neurons, and a variety of GABAergic interneurons. Both classes give rise to the interstitial neurons of the white matter, along with the neurons of the intermediate zone. Finally, the subventricular zone (SVZ) is a cortical compartment that acquires an extraordinary prominence during the second half of gestation, when it continues to proliferate giving rise to Tbr2+ neurons, and also to calretinin+ interneurons, in contrast to the rodent SVZ. The SVZ is highly reduced in mutations of genes such as DOUBLECORTIN and LIS1, which cause severe malformations of the lissencephaly 1 type. Disturbances of these fetal compartments – for many reasons, genetic, traumatic, infections etc- can cause severe alterations of cortical development, with often dramatic anatomical and functional consequences in the postnatal period.

### **C3: CHOROID PLEXUS-CSF NEUROPEPTIDE INVOLVEMENT IN INTRACRANIAL PRESSURE REGULATION: IMPLICATIONS FOR THE PUTATIVE INTRACRANIAL HYPERTENSION OF SPACEFLIGHT**

Conrad E. Johanson

*Dept. of Neurosurgery, Alpert Medical School at Brown University, Providence, Rhode Island, USA.*

Intracranial pressure (ICP) elevation is detrimental to brain function in many ways. To achieve better control of ICP, it is important to ascertain whether brain fluid pressure is actively regulated by a neuropeptide-mediated feedback mechanism to reduce choroid plexus (CP) production of cerebrospinal fluid (CSF) when ICP rises. Atrial natriuretic peptide (ANP) is a candidate CSF neuropeptide to mediate such a servomechanism to prevent untoward elevation in ICP. Choroid plexus epithelium has a high density of natriuretic peptide receptors (NPR) for ANP, BNP and CNP ligands. Bmax (transport capacity) and Kd (receptor affinity) have been extensively characterized for NPRs at the blood-CSF interface in the ventricular plexuses. There is great plasticity in choroidal NPR expression, with compensatory changes occurring in hydrocephalus, and in spaceflight when CSF volume and pressure apparently undergo changes in microgravity (near zero G). In mammalian species experiments (including hydrocephalus), ANP infused into the lateral ventricles curtails CSF production rate and lowers ICP. Dark, neuroendocrine-like epithelial cells are induced by ANP in vitro and in hydrocephalus; such dark cells are typically associated with inhibited fluid production in vivo, and have been observed consistently in CP by electron microscopists and CSF physiologists for decades.

Choroid plexus epithelial cells, manufacturing most of the CSF, undergo interesting morphologic changes (suggesting modified fluid movement) when exposed to ANP in vitro, and when subjected to hydrocephalus (kaolin-induced) and actual spaceflight (rodents carried by the USA space shuttle; Gabrion et al.). Moreover, in short-term space flight (days to weeks), experimental rat CP undergoes alterations in ANP receptor density; and expression in apical membrane aquaporin 1 (AQP1 water channel, a driver of CSF) as well as enzymes involved in CSF production (choroid epithelial immunostainings). Together, these findings point to down-regulated CSF formation by CP in response to the presumed augmented ICP in spaceflight.

The experimental observations by us, and other laboratories, thus fit the model that the homeostatic CP is sensitive and responsive to ICP alterations. The collective data are consistent with several clinical studies demonstrating that the titer of ANP in human CSF varies directly with ICP. It appears that all the elements of a neuropeptide-mediated servomechanism, to adjust ICP levels, are present in the CP-CSF system and nearby hypothalamus. To control CSF volume and ICP in astronauts orbiting the earth in the International Space Station or on longer space missions (e.g., Space Exploration Vehicle to Mars), there may be a need for therapeutic agents to minimize any elevation in ICP (or CSF volume shifting) occurring in space-travelers. ANP, for intranasal administration to access the CSF and thus circumvent the blood-brain barrier, appears to be a peptide fluid-regulating agent worthy of expanded experimental pursuit.

### **C4: THE ANATOMICAL DISSECTION AS FACILITATOR OF THE MEDICAL DEVELOPMENT. THE CONSOLIDATION OF THE VESALIUS' ANATOMY IN SPAIN**

Ricardo Vazquez

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This essay is divided into two parts, the first one is a short review dedicated to how the dissection was essential for the developing of the Medicine until the Renaissance and the second reviews how the work of Andreas Vesalius set up the Modern Anatomy.

Galen was the first one in considering Anatomy from a functional point of view but base on the dissection of animals. This kind of medical knowledge lived until the middle of the sixteen-century, the Renaissance. The human dissection, during that long period of time, was made only in a sporadic way and in most of cases, for legal aspects.

In the sixteen century Andrea Vesalius made the anatomical revolution. It consists in doing, not only reading, the lecture from the “cathedra” but making at the same time the dissection of the cadaver related with the text. With his experience, and with the help of excellent drawings, he wrote the “*Humani Corporis Fabrica libri septem*”. That book was quickly accepted and used for many European Universities; in this way Anatomy became the first independent discipline for teaching medicine. After

that came as independent disciplines: Surgery, Physiology and later the rest of medical specialties.

The pupils of Andreas Vesalius translated to many countries his knowledge and method, including Spain. The Spanish Universities of the sixteen-century, with more or less success, set down the Vesalius's knowledge. In this way were created different "cathedra" of Anatomy using the dissection as the method for knowing the human body and with it facilitate the development of the Medicine.

Precise rules for teaching Anatomy, mainly in relation with dissection and difficulties that supposes for the prosecutor, were established. With the aim of teaching anatomy were created the anatomical Theatres. The first anatomical Spanish theatre was named "the Anatomical house" and was built in Salamanca.

The Vesalius' anatomy, step by step, was won territory to the Galen's anatomy and was the first step for establishing the base for the development of the Modern Medicine.

**ABBREVIATIONS****Topic areas**

ACyS = Systemic and Clinical Anatomy

DA = Teaching in Anatomy

E = Embryology

N = Neuroanatomy

**Mode**

O = Oral presentation

P = Poster

**Codification**

Topic area – Mode – Order number

**Example**

DA-0-02 : Teaching in Anatomy communication, Oral presentation, number 2



# ORAL COMMUNICATIONS



**E-O-01 MOLECULAR BASIS OF CEREBELLUM DEVELOPMENT**

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We investigated the cellular and molecular mechanisms underlying the proper development of the cerebellum by experimental designs in mouse embryos. Several genes and molecules were identified to be involved in the cerebellar plate regionalization, specification, and differentiation, as well as in the establishment of cellular migratory routes and the subsequent neuronal connectivity. Indeed, pattern formation of the cerebellum requires the adequate orchestration of both, key morphogenetic signals, arising from distinct brain regions, and local expression of specific transcription factors. Thus, we present here the embryonic specification and the molecules involved of cerebellum development (topography and topology) in order to understand causal processes regulating cerebellar cytoarchitecture, its highly topographically ordered circuitry and its role in brain function.

*This work was supported by the following Grants:*

*European Consortium: EUCOMMTOOLS (Contract 261492), the Spanish Ministry of Science and Innovation: FEDER (BFU-2011-27326) and CONSOLIDER (CSD2007-00023)*

**E-O-02 DEVELOPMENT OF THE CILIARY BODY**

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The distal portion of the OC (optic cup) was studied on 36 human embryos and 29 human fetuses, belonging to the Embryology Institute of Complutense University of Madrid. The parameters used to determine post-conception age were the greatest length, external and internal criteria. During E14, the OC is made up of two layers, outer (OL) and the inner (IL) layers of the OC separated by the optic ventricle. Both layers continue into the distal part of the OC, constituted by a compact, triangular shaped cellular zone and vertex pointing to the optic ventricle. At E15, the distal portion of the OL makes contact with the IL via small, cytoplasmic expansions, except in the most distal portion where the marginal sinus is formed. The OL presents more abundant pigment granules in the apical zone where both epithelia come into contact. At E20, the distal part of the OC is related to mesenchymal condensation corresponding to the developing sclera. Between the distal part of the OC and the sclera, vascular mesenchyme, trabecular in appearance, may be observed. This is the anlage of the stroma and the ciliary

muscle. During the 9th week of the development, undulations related to the development of the periorcular mesenchyme vessels appear in the pigmented epithelium. We feel that they correspond to the anlage of the ciliary folds. During the 13-14th weeks, two zones become defined in the epithelia of the ciliary body; one with folds, the anlage of the pars plicata of the ciliary body and a smooth proximal zone, the anlage of the pars plana of the ciliary body.

Our study in human specimens at 5-14th weeks of development show the morphological changes in the distal portion of the OC that delimit the anlage of the epithelia of the ciliary body, and also the initial stages of development of the ciliary muscle.

**E-O-03 VARIATIONS IN THE DISTRIBUTION OF AQP1 IN DIFFERENT CASES OF CONGENITAL HUMAN HYDROCEPHALUS**

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Aquaporin1 (AQP1) is transmembrane protein that regulate the movement of water and small solutes through the cell membrane, contributing to water homeostasis. AQP1 is highly expressed at the ventricular surface of the choroid plexus epithelium in mice, rats, humans, and other species. AQP1 expression has also been reported at the basolateral (blood side) of the choroid plexus epithelium in rodents and humans but shows a weaker pattern. The higher ventricular expression of AQP1 is thought to play a significant role in transcellular transport of water molecules for the production of CSF. This polarized expression of AQP1 at the choroid plexus has been confirmed in humans. Since AQP1 specific distribution was found out, has been reported its role in the production of CSF, and now exist a few studies concerning AQP1 and Hydrocephalus. A few studies show a down-regulation of AQP1 expression in hydrocephalus suggesting an adaptive feedback mechanism that acts decreasing the production of cerebrospinal fluid (CRF) and intracranial pressure (ITC). (8,11) Other studies do not confirm these observations, therefore is a very controversial subject.

Congenital hydrocephalus human at 22 gestation week were used and aquaporin-1 (AQP1) was visualized by immunofluorescence. An Arnold-Chiari syndrome and a

Sylvian aqueduct obstruction were used as non-communicating hydrocephalus and two tetraventricular ventriculomegalia of unknown origin were considered communicating hydrocephalus.

The results of the present work do not show any down-regulation of AQP1, however we found spectacular changes concerning AQP1 distribution in both type of hydrocephalus. In communicating hydrocephalus was highly expressed at the ventricular surface of the choroid plexus while in noncommunicating hydrocephalus while in communicating hydrocephalus was clearly found at the basolateral membrane of the choroid plexus epithelium. Therefore, it could be thought that the mechanism regarding to an adaptive feedback, that acts to lower the production of CSF and ICP, depends on type of hydrocephalus and that even could have a change of polarization of AQP1 in non-communicating case, which could allow turnover of CSF through basolateral membrane of the choroid plexus epithelium.

#### E-O-04 AQUAPORIN-4 EXPRESSION IN THE PARIETAL TELENCEPHALIC NEUROEPITHELIUM OF CASES OF CONGENITAL HUMAN HYDROCEPHALUS

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Aquaporin-4 (AQP4) is one of the most selective water channels of all the aquaporins, it is observed in astrocytes and is scarce in CNS neurons. AQP4 is mainly located in the brain barriers and in the subependymal ventricular layer. In the present work, the expression of AQP4 in the brain parenchyma in control and congenital human hydrocephalus (communicating and obstructive) is analyzed. Three control brains at 18, 20 and 21 gestation weeks and four hydrocephalic brains at 20, 21, 21 and 22 gestation weeks were used, two were obstructive hydrocephalus and two were communicating. AQP4 in the brain parenchyma in control and congenital human hydrocephalus were studied by enzyme immunoassay (ELISA), western blot, immunohistochemistry and immunofluorescence. The ELISA showed that, AQP4 concentration, in parenchyma extracts, was higher in controls than in hydrocephalus. The western blot also showed that AQP4 labeling was significantly lower in the

hydrocephalus brain extracts. The AQP4 immunoreaction was clearly positive in the control. The AQP4 labeling was low in most of the ependymal and subependymal layer tissue in the both types of hydrocephalic brains (obstructive and communicating). The AQP4 immunoreaction was also located around blood vessels. In contrast, AQP4 immunoreaction was clearly observed in cerebral vessels, the ependymal and subependymal layer and the basolateral membrane of the ependyma in the controls by immunohistochemistry and immunofluorescence. AQP4 is a water channel that acts in CSF production and reabsorption and the ependymal junction. AQP4 also has a modulatory effect on ependyma stability. Therefore, the decrease of Aquaporin-4 (AQP4) is one of the most selective water channels of all the aquaporins, its observed decrease of AQP4 in the brain parenchyma could be due to the fact that AQP4 passes from the parenchyma to the CSF in congenital hydrocephalus, and this AQP4 movement may be a consequence of ependyma denudation.

#### DA-O-01 USE OF AN INTERACTIVE AUDIENCE RESPONSE SYSTEM IN MEDICINE AND PHYSICAL THERAPY

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The aim of this work is to provide grade students of Medicine (Neuroanatomy) and Physical Therapy (Musculoskeletal system of members) of an Audience Response System (ARS) similar to Educlik® in which students mobile devices are used as clickers with only a mobile application.

This interactive response system (SIDRA application) allows, by plotting the results for each question, a greater participation, interactivity and the possibility of positive feedback to students to help them to improve their learning strategies.

Due to the scant literature that values the application of this technology to specific matters, we compared, in each subject, the average ratings of the groups working with SIDRA versus those without, and assess performance and success indicators of test results. In addition, we used a questionnaire to measure student's satisfaction and motivation using SIDRA and their learning process (Likert - type scale 1-5).

Despite some technical difficulties of this new system, the features of these subjects (as extensive use of iconography) allowed proper use of SIDRA and increase the benefits about student learning. Moreover, it is possible to assess the possibilities of this application as a basis for more complex tools such as neural networks that allows student self-learning.

*Educational Innovation Project UMC Chancellor's Resolution (R-667/2012)*

## DA-O-02 PRODUCTION AND DELIVERY OF MORPHOLOGICAL KNOWLEDGE BY DIGITAL DISSECTION OF BODILY AND EMBRYONIC STRUCTURES

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Information and Communications Technologies (ICT) are now providing unprecedented possibilities for enhancing the teaching and learning of human anatomy and embryology. This report addresses the digital extraction of anatomical information from serial imaging data, for use in medical education.

Stacked clinical imaging data, including but not limited to computed tomography (CT), magnetic resonance imaging (MRI), or optical projection tomography (OPT), can be used as a source of anatomical data, although other resources such as histological sectioned material also can be used. The former are usually preferred because they contain spatially accurate structural information. If digitized serial histological sections are used as source data, a process named registration is required for re-aligning sectioned structures.

A major procedure for extracting three-dimensional information from stacked, two-dimensional data is the so-called segmentation, i.e. delimitation of the contours of a given structure of interest across sectional images. Segmentation can be run automatically by means of thresholding techniques or other detection algorithms. However, manual segmentation yields optimal results when the structures of interest are not readily distinguishable from adjacent structures and thus not easily delimited automatically. Manual segmentation requires expert knowledge and may be an arduous task. An example is presented showing the manual segmentation of the neural tube from stacked OPT data of a human embryo at the Carnegie Stage 12 (26th post-ovulatory day approximately) that were generated and distributed as part of the HUDSEN atlas project (<http://www.hudsen.org>).

Upon completion of the segmentation process, a three-dimensional polygon surface mesh representing the spatial structure of the desired anatomical element can be computed from the segmented contours. A polygon surface mesh consists of a collection of vertices, edges and faces that defines the shape of a polyhedral object in 3-D computer graphics. The structure of so-obtained polygon meshes conveys three-dimensional information defining the morphology of an anatomical structure and relative positions and topographical relationships among the segmented elements. After scaling, correction and optimization of polygon meshes, these can readily be used for 3-D visualization of anatomical and embryological scenes, virtual dissection, or rapid prototyping for a hands-on approach in the anatomy laboratory.

## DA-O-03 ROLE OF EPONYMS IN CURRENT ANATOMY

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Eponym in anatomy means a term, derived from the name of a real or mythical person, who can contribute to the description or discovery of the involved structure. Anatomical terminology and nomenclature is the basic construction stone for the denomination of structures. In anatomy, the nomenclature has a long tradition from 1895 (Basiliensia Nomina Anatomica - BNA) to the last revisions are the Terminologia Anatomica (TA 1998). The eponyms were excluded from the nomenclature as early as 1955 (Parisiensia Nomina Anatomica - PNA) but they still survive both among anatomist and among clinicians. TA brought a list of the most usual of them with their official Latin equivalents. Several website exist serving as databases of the eponymous personalities and the eponyms. In clinical extensions of TA several eponyms reappeared (Cockett, Giacomini, Santorini). But the situation still remains unclear especially for the teachers how to approach this question, mainly due to the local and interdisciplinary differences in usage and favour of eponyms.

The authors proposed a simple classification of eponyms into three groups:

A for every student and physician, useful and necessary in the whole study, scientific and medical life (e.g. Achilles tendon or Eustachian tube)

B for specialist in one specific field (e.g. Weitbrecht's retinacula or Onodi's cell)

C for archaic, obsolete and abundant terms (e.g. torcular Herophyli or vein of Lichaceva)

The next step is to complete all eponyms from groups A and B with their official Latin terms, if missing.

*Supported by Charles University Grant 236028/IRP/2013*

## ACYS-O-01 CORONA MORTIS REVISITED

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Corona mortis is a well-known anatomical variation of an artery and vein crossing the *ramus superior ossis pubis*. Unfortunately, it is not an official anatomical term (of Terminologia Anatomica) and its anatomical arrangement is, especially in veins, rather inconstant. The vessels should be denominated as *vasa obturatoria aberrans* and in case of existence of proper obturator vessel as *vasa obturatoria aberrans accessoria*.

*Arteria obturatoria* is usually a branch from the *arteria iliaca interna* (AII), but in 20% of cases it branches from the *arteria iliaca externa* (AIE) as *a. obturatoria aberrans*. There are two arteries fusing just at the ostium of the *canalis obturatorius* in other 5% of cases - *a. obturatoria* from AII and *a. obturatoria aberrans accessoria* from AIE. The overall incidence of arterial "corona mortis" is about 25% with vessel being more than 1 mm wide. It is located in average 56,3 mm

from the midpoint of the *symphysis pubica* (spanning from 40 to 90 mm).

*Vena obturatoria* is a tributary of the *vena iliaca interna* (VII). A branch into the *vena iliaca externa* (VIE) - *v. obturatoria aberrans* - occurs in 10% of cases, crossing *ramus superior ossis pubis* to exit the lesser pelvis, and draining either into the *vena pubica*, *vena epigastrica inferior* or directly into VIE. There are two different veins dividing at the ostium of the *canalis obturatorius - v. obturatoria* opening into VII and *v. obturatoria aberrans accessoria* opening into VIE or its tributaries in 83% of cases. The overall incidence of venous "corona mortis" is approximately 63% with vessel being more than 2 mm wide. It is located about 54,9 mm from the midpoint of the *symphysis pubica* (spanning from 20 to 90 mm). The data from a meta-analysis covering 4919 arterial specimens and 1164 venous specimens brings following incidence and including 451 our specimens: arterial corona mortis is present in 26,4% and venous one in 93.3% of cases.

The clinical relevance of the "corona mortis" consists in fractures of pubic bone and their surgical treatment, hernia repair, prostatectomy, angiographic embolization or harvesting of an arterial graft.

#### ACYS-0-02 PRESURGICAL 3D GAIT ANALYSIS FINDINGS IN FOUR CHILDREN WITH SPASTIC CEREBRAL PALSY

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**Introduction:** Children with spastic palsy present usually with an altered gait pattern. This pattern presents various superimposed alterations, due to spasticity and due to musculoskeletal deformities; some of them are principal and some of them are compensatory mechanisms developed by the patient. Due to the complexity of the clinical analysis of these patients, automatic gait analysis is a very useful tool

**Material y methods:** We have studied four children: 2 of 5, one of 4 and another of 9 years old. All of them had independent gait with walkers or cane aids.

We have performed a kinematic study of articular movement (pelvis, hip, knee, and heel); a kinetic study of power and strength used during movement of these articulations; and a dynamic surface electromyography (sEMG) study.

For the acquisition of kinematic and kinetic data we used a VICON system with 6 infrared cameras that register the movement in the space of antropometric markers attached to the patient and two dynamometric platforms which

register the ground reaction force. For the acquisition of EMG we used a Biopac system (MP100) with 4 wireless channels, integrating on real time the EMG analogic signal with the VICON system.

**Results:** We observed alterations in the kinematic of articulations that were different for each patient and we cannot define a common pattern of gait. In kinetic studies we observed an increase in the excentric and concentric involvement of several articulations. Gait parameters were also altered in the four patients.

**Conclusion:** Automatic gait study allows us to specify and quantify what articulations and muscles are involved in the altered gait. We also can register and quantify spasticity, determine whether spasticity is generalized or not and if there are associated morphological deformities or alterations.

#### ACYS-0-03 FACIAL REINNERVATION BY MASSETERIC NERVE: A PRELIMINAR ANATOMICAL STUDY

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**Introduction:** The facial palsy is a disabling condition; therefore a therapeutical challenge is necessary to be faced. Up to now several techniques have been proposed to reestablish the function of the facial nerve, with more or less success. Some of these techniques are muscular transfer or, more recently, nerve transfers as masseteric nerve connection. The studies using the masseteric nerve have assessed landmarks, branches or axonal load. The last aspect has been study with the classic Hematoxili-Eosyn method, therefore that studies only establish the number of total axons of the nerve. In the present study we aim to establish not only the number but also the nature of the axonal charge: motor and sensorial axons.

**Material and methods:** The study is based on the dissection of 6 human hemi-heads. The facial and masseteric nerves and their branches were exposed and samples of the nerves were obtained. The motor axons load of these samples was assessed by immunohistochemical labeling for the enzyme choline-acetyltransferase (ChAT), present in all motor fibers. The pictures of the samples were analyzed by means of Image J software to determine the number of motor axons in the nerves.

**Results:** The facial nerve shows a variable pattern of distribution of its branches; the masseteric nerve is more predictable. The motor load of both nerves is also variable but is much more similar than expected regarding the different size of both nerves: the number of ChAT positive axons in the facial nerve ranged from 468 to 535 and in the masseteric nerve from 245 to 494.

**Conclusions:** We can conclude that the masseteric nerve is a good candidate for being use as a candidate for reinnervating the facial muscles. Moreover, the axonal count reveals that it is a very recommendable therapeutical option.

#### ACYS-0-04 THE THIEL'S EMBALMED CADAVER AS A IDEAL MODEL FOR ADVANCED ENDOSCOPIC SURGERY PROCEDURES TRAINING

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Today, the domain of laparoscopic techniques is essential in the formation of contemporary general surgeon. To this end, are different experimental courses offerings: pelvitrainers, animals (usually pigs), fresh or embalmed cadavers and recently, virtual reality simulators. None of them is the perfect model, and each has advantages and disadvantages. While pelvitrainers and virtual simulators allow the acquisition of basic laparoscopic skills; training in anesthetized pigs offers a unique opportunity to learn the procedures and maneuvers in vivo conditions. However, this training tool is expensive, the results different from human anatomy point where cadaveric models take advantage. To provide an option to shorten the learning curve in advanced laparoscopic surgery, our group has developed a training protocol based on the model of Thiel's Embalmed Cadaver (TEC) which we present in this descriptive study.

The embalming technique of Professor Walter Thiel (Graz, Austria) is a sophisticated method of preserving the human death body, that maintains most of the features of a living body, which makes the TEC an excellent pattern for gastrointestinal endoscopic surgery practice, because it allows the pneumoperitoneum and abdominal wall distention. Despite the advantages of this model, a study in 2011 revealed that while 78% of Europeans anatomists know this technique and 57% of them have no real experience with the TEC (they have never prepared or TEC dissected a model.) Also in a recent study by our group, it became clear that very few surgeons who know this resource or have come to use, considering that it is an ideal tool for the practice of surgery laparoscopic.

#### ACYS-0-05 ALVEOLAR CHANGES AFTER THE APPLICATION OF PROTECTIVE MECHANICAL VENTILATION WITH DIFFERENT CONCENTRATIONS OF OXYGEN IN HEALTHY RATS

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**RATIONALE:** Mechanical ventilation (MV) is a key tool in the management of acute respiratory failure. As with any therapy, mechanical ventilation has side effects and may result in ventilator-induced lung injury (VILI) (Lionetti et al., 2005). The potential importance of VILI in the clinical treatment of critically ill patients was established by the recent trials performed by the Acute Respiratory Distress Syndrome Network (ARDS Network, 2000, 2004). The results of these large clinical trials showed a relative risk reduction of 22% in patients ventilated with the lower tidal volume (6ml/Kg, protective ventilation). Several experimental and clinical studies lead to the hypothesis that the deleterious effects of mechanical ventilation may be mediated by lung inflammation and the systemic release of inflammatory cytokines (biotrauma) (Uhl et al., 2004).

**METHODS:** In this study we analyzed in 4 groups of healthy rats different concentrations of oxygen (FiO<sub>2</sub>: 0,21, 0,4, 0,6 and 1) while 4 hours of protective MV. The anaesthetic induction was made with ketamine and Xylazine, and a tracheostomy was performed and connected to MV (Ugo Basile, Italy). After 4 hours the animals was sacrificed and the lungs was removed and fixed in formalin 10%. Subsequently processed by light microscopy and stained by HE and Masson-Goldner Trichrome, assessing changes which set: alveolar septa, alveolar haemorrhage, intraalveolar fibrin, intraalveolar inflammatory infiltrate and fibrosis (Matute-Bello et al., 2001; Hübner et al., 2008).

**RESULTS:** In all groups was evident that a higher concentrations of oxygen a greater grade of histological lesions was appreciated.

**CONCLUSION:** The protective mechanical ventilation with lower tidal volumen by itself don't prevent the VILI because the concentrations of oxygen participates in its development.

#### ACYS-0-06 ROLE OF THE TYPE 2 ALVEOLAR EPITHELIAL CELLS IN AN EXPERIMENTAL MODEL OF ACUTE LUNG INJURY

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**RATIONALE:** The acute lung injury (ALI) is the disruption of the gas exchange in response to deleterious environment or internal agents (Slutsky & Imai, 2003); experimental studies have shown that the best mechanical ventilatory strategy in ALI is one that was used in low volumes and PEEP (Villar et al., 2009); in this experimental study we propose to assess the participation of type 2 alveolar epithelial

lial cells after applying different models of mechanical ventilation (MV).

**METHODS:** Prospective, randomized, controlled animal study using healthy adults Sprague-Dawley rats; animals were anesthetized and randomized to spontaneous breathing and to two different mechanical ventilation strategies for 4 hours: high tidal volume ( $V_T$ ) (20 ml/kg), low  $V_T$  (6 ml/kg) and a control group without mechanical ventilation; an histological evaluation of the lungs (HE and Masson-Goldner trichrome) and immunohistochemical stains for thyroid transcription factor 1 (TTF1) and surfactant protein D (SP-D) were performed.

**RESULTS:** Lungs from animals ventilated with high  $V_T$  had acute inflammatory infiltrates and perivascular oedema whereas there were no major histological differences between animals ventilated with low  $V_T$  compared to spontaneously breathing animals; in the IHC study using TTF1 and SP-D, the lungs of low  $V_T$  not present differences with the normal disposition of type 2 alveolar epithelial cells while those ventilated at high  $V_T$  showed a marked hyperplasia of these AEC2 cells.

**CONCLUSIONS:** The mechanical ventilation “*per se*” is capable of producing lung injury; in this study we postulate the idea that hyperplastic surfactant secretory AEC2 cells that appear in animals ventilated at high volume ( $V_T$ ), are a representative mechanism by which the lung tries to avoid the injury caused by MV.

#### N-O-01 HUMAN CUTANEOUS MEISSNER CORPUSCLES DEVELOP POSTNATALLY

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The dermis of the human glabrous skin contains, in addition to free nerve endings, different types of sensory corpuscles (Ruffini's corpuscles, Meissner's corpuscles, Krause's corpuscles, and Pacini's corpuscles), and this diversity can be regarded as an indication of the differentiated ability of sensory corpuscles to detect multiple and diverse sensory stimuli. In spite of the importance of touch in providing information to the central nervous system, the temporal pattern (embryonic and postnatal) of the development of sensory corpuscles, especially in humans, is poorly known. Sensory corpuscles develop from the interactions of the growing sensory axons, the non-differentiated neural crest-derived cells (Schwann-related cells) which migrate together the axons, and the target mesenchyma. It is known that human Pacinian corpuscles begin to develop from third or the fourth month of gestation but as far as we know no information is available about the development of human Meissner corpuscles. The present study was designed to analyze the development of Meissner corpuscles in human digital skin from the embryonic period (12 weeks of estimated gestational age) to adulthood (25 years), using immunohistochemistry for several axonic (neurofilaments, neuron specific enolase) and

Schwann cell (S100 protein, vimentin, N-CAM) markers, as well as for the basal membrane (type IV collagen) or the neurotrophin complex BDNF/TrkB. Results demonstrate absence of Meissner corpuscles from human digital skin during the prenatal development. Immature Meissner corpuscles were detected around birth, were well differentiated and reached an immunohistochemical profile proper of fully developed Meissner corpuscles around 18 months of life.

#### N-O-02 OCCURRENCE OF MECHANOPROTEINS IN CUTANEOUS MECHANORECEPTORS OF *MACCACA FASCICULATA*

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Mechanotransduction is the conversion of a mechanical stimulus into an electrical signal. In mammals the cutaneous mechanosensitive structures are special sensory organs known collectively as mechanoreceptors. Despite efforts to discover molecules that initiate touch sensation only recently it has been suggested that different types of ion channels are directly activated by mechanical stimuli. Acid-sensing ion channels (ASICs) and Transient Receptor Potential Vanilloid 4 (TRPV4) mediate different sensory modalities, including mechanosensation. In the present study we investigated the distribution of ASIC2 and TRPV4 proteins in cutaneous Pacinian corpuscles and Meissner corpuscles of *Maccaca fasciculata* using immunohistochemistry for ASIC2 and TRPV4, together with neuron-specific enolase and S-100 protein, and laser confocal-scanner microscopy. ASIC2 and TRPV4 were detected in both Meissner and Pacinian corpuscles. ASIC2 was found co-localized with neuronal markers but not Schwann cell-related markers demonstrating that ASIC2 expression is restricted to axons supplying mechanoreceptors. TRPV4 immunoreactivity was found to co-localize with neuronal markers, but also with Schwann related cell markers, thus being present in all cells forming the mechanoreceptors. The present results demonstrate for the first time the occurrence of mechanoproteins in *Maccaca fasciculata* low-threshold mechanoreceptors and provide evidence for a possible role of these ion channels in mechanosensation.

#### N-O-03 ARE THE TRPC2 POSITIVE CELLS IN THE OLFACTORY EPITHELIUM OF ADULT ZEBRAFISH THE EQUIVALENT TO THE MAMMALIAN VOMERONASAL ORGAN?

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The peripheral olfactory system of vertebrates consists of bipolar olfactory receptor neurons (ORNs) localized in the olfactory epithelium. In addition mammals possess an accessory olfactory system, the vomeronasal organ, which functions as a detector for chemical signals concerning social organization and reproductive status. In teleosts the olfactory epithelium contains three types of ORNs denominated ciliated, microvillous, and crypt neurons which can be distinguished on the basis of their morphology and neurochemical features. The zebrafish is an attractive model for studying the cellular and molecular basis of behaviours, including those in which the olfaction plays a key role as feeding and reproduction. Whether or not a type of ORN in the olfactory epithelium of adult zebrafish can be assimilated to the pheromone-detecting system of mammals, is not known. TRPC2 (transient receptor potential canonical 2) an ion channel highly localized in the vomeronasal sensory neurons, and it is fundamental for detection of pheromone signals since TRPC2-deficient animals exhibit striking behavioral defects in the regulation of sexual and social behaviors. Based on these data we used immunohistochemistry and laser-confocal microscopy to localize TRPC2 in the olfactory epithelium of adult zebrafish. The results demonstrate numerous TRPC2 positive neurons whose distribution and morphology resembled that of the crypt neurons; nevertheless, the density of TRPC2 positive neurons was larger than that of the calretinin positive ones (a marker for crypt neurons) and no-colocalization of calretinin and TRPC2 was observed. The true nature of the TRPC2 olfactory neurons in zebrafish remains to be established.

#### **N-O-04 THE PROTEIN INSULIN-RECEPTOR-SUBSTRATE 2 (IRS2) IS ESSENTIAL FOR THE INTEGRITY OF THE HIPPOCAMPUS AND ITS ABSENCE COULD INDUCE HIPPOCAMPAL NEURODEGENERATION**

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Insulin receptor substrate proteins (IRS) integrate signals from receptors of insulin and IGF-1 with intracellular pathways to control growth, function and cell survival; since insulin and IGF-1 exert their physiological effects through

the phosphorylation and activation of the IRS. The actions of the IRS2 within the nervous tissue are relevant because promotes the maturation and survival of the retinal photoreceptors immediately after birth, and its deletion reduces the brain size due to abnormal cellular proliferation during embryonic development, and its absence in adults produces additional pathology in the brain of these animals: hyperphosphorylation and accumulation of Tau microtubule-associated protein. Since the alterations in brain growth appear prior to the onset of hyperglycemia, the deregulation of the cascade of signals mediated by IRS2 could be a molecular bridge between diabetes and neurodegenerative diseases. Previously, using magnetic resonance imaging, we found that the study of the brains of knockout old mice for IRS2 reveals the existence of signs of neurodegeneration similar to those described for schizophrenia or Alzheimer's disease: increases in the size of the brain ventricles and decreases in the thickness of different cortical areas, the striatum, the hippocampus and the subthalamic nucleus.

In this study, we demonstrate using the post-mortem analysis that the brains of these animals presents alterations of hippocampus and dentate gyrus and horn of ammon (CA1, CA2 and CA3), which suggest that the protein IRS2 is essential for maintaining the structural integrity of the hippocampus. There is a significant decrease in the thickness of the hippocampus in four regions analyzed, with great affection of the pyramidal layer. In addition, in all of them, there is an evident accumulation of Tau-2, presence of amyloidosis was demonstrated with congo red staining and the presence of  $\beta$ -amyloid peptide by means of immunohistochemistry. These changes are accompanied by reactive gliosis, at least for GFAP-positive astrocytes in dentate gyrus. The neural sinaptology decays considerably since there is a marked and significant decrease of the presence of SV2 synaptic vesicle protein. In these animals, a significant decrease of GABAergic neurons in the hippocampus was observed.

In sum, the findings observed suggest that IRS2 protein plays an important role in the development and neuroprotection. Since the values of glycemia and insulinemia in these mice were normal, all observed variations could be attributable to changes in intracellular signals that are triggered after the phosphorylation of IRS2, and that they are not supplemented by the phosphorylation of other IRS as occurs for glucose uptake.

*Supported by: JCyL: SA073A06; y CICYT, I+D+i: BMC2002-00872.*

#### **N-O-05 LENGTH AND DIAMETER OF AXON INITIAL SEGMENT OF PYRAMIDAL NEURONS IN LAYER II AND LAYER III OF ANTERIOR CINGULATE CORTEX OF THE RAT BESIDES NUMBER AND DISTRIBUTION OF INCOMING AXO-AXONIC SYNAPTIC BOUTONS**

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This paper presents parametric data of the axon initial segment (AIS) of principal cells in layers II-III of Vogt's area

24b of rats. Three classic spiny pyramidal neurons were studied by means of the rapid Golgi impregnation under correlated light- and electron microscopy. One cell sat in layer II and the remaining in layer III. In pooled AIS of layer III cells, the mean length was  $35.04 \pm 11.62 \mu\text{m}$ , whereas the mean diameter was  $0.69 \pm 0.11 \mu\text{m}$  (the individual diameters had been averaged from estimates of the proximal, intermediate and distal segments of each AIS). The number of axo-axonic synaptic boutons apposed to the pooled AIS was  $13 \pm 5.86$ ; density (boutons divided per AIS-length) was  $0.37 \pm 0.04$ . In turn, the AIS of layer II cell measured  $53.52 \mu\text{m}$  in length and  $0.72 \mu\text{m}$  in diameter. It received 22 incoming synaptic boutons, with a density of 0.41. Thus, the AIS of the classical spiny pyramidal neuron was longer and more innervated in the cell of layer II. Results were compared to others of occipital cortex of rats [1]. The outcome was that the AIS of classic pyramidal neurons of layers II-III was longer, thinner and less innervated—both in the absolute numbers and density—in area 24b of anterior cingulate cortex than in occipital cortex. Patterns of innervation along AIS also differed in both regions.

*Work funded by UPV/EHU Grants GIU07/14 and UFI11/41.*

#### **N-O-06 IS THE ARYTENOID MUSCLE SUPPLIED BY THE RECURRENT AND THE SUPERIOR LARYNGEAL NERVES?**

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Classically it is considered that the nature of the branches from the internal laryngeal nerve (ILN) to the arytenoid muscle is exclusively sensory. However, recent neurophysiological studies suggest that the internal laryngeal nerve provides a secondary motor innervation for this muscle. In addition, certain clinical aspects as the variability of the vocal folds position after recurrent nerve sectioning persist with no satisfactory explanation. Therefore, the aim of this study was to analyse the axon load and nature of the axons within the branches of the ILN to the arytenoid muscle.

In order to achieve this goal, five human larynges were dissected under a surgical microscope. The posterior trunk of the ILN, branches for the arytenoid muscle and ramus communicans were exposed and samples of the nerves were obtained. The motor axons load of these samples was assessed by immunohistochemical (IHQ) labeling for the enzyme choline-acetyltransferase (ChAT), present in all motor fibers. The pictures of the samples were analyzed by means of Image J software to determine the number of motor axons in the nerves.

The arytenoid muscle received from 5 to 8 branches. The IHQ stain revealed the presence of motor axons in all the samples.

The existence of motor axons in the ILN branches for the arytenoid muscle confers a secondary motor innervation pathway to this muscle, confirming the dual innervation of this muscle that could justify the complex nerve supply of laryngeal muscles and the variable position of the vocal folds after recurrent laryngeal nerve sectioning.

#### **ACYS-0-07 VARIATIONS IN THE FORMATION AND ORIGIN OF THE TRUNKS OF BRACHIAL PLEXUS AND THE TOPOGRAPHICAL LOCATION OF THE MEDIAN NERVE. A CASE REPORT**

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We presented a right axillary dissection from a female corpse, in which we can appreciate several anatomical variations of the brachial plexus, in its formation and branching pattern.

The first variation is the numerical modification of the primary trunks, instead of being a single middle trunk there are two Primary middle trunks, one of them is superficial and the other is deeper. Moreover, the Upper primary trunk and the superficial middle trunk have their origin through the anterior Scalene muscle to descend to the apex of the axilla, instead of being between the anterior scalene and middle scalene like the other primary trunks.

The second variation is the atypical topographical situation of the Median nerve respect to the Axillary artery, as it's seemed "dropped", rotated into the axilla, instead of being above the artery.

The third variation is the origin of the upper and lower Subscapular nerves. The upper Subscapular nerve originates in the Upper primary trunk (external surface) immediately before to be formed the Posterior cord or posterior secondary trunk along with other primary trunks. Otherwise, the lower Subscapular nerve starts as collateral branch of the Axillary nerve, very distal from the upper nerve aforementioned.

The fourth anatomical variation is in the Long Thoracic nerve, which origin is in the superficial middle primary trunk. Moreover, along its path descending, this nerve receives a nerve supply from the deep middle primary trunk.

The fifth variation founded concerns the formation and location of the pectorals loop, as it is constituted around Thoraco-acromial artery from collateral nerve branches of the medial and posterior cords.

In addition, we carry out study of multiplanar reconstructions (MPR) and volume rendering, besides tridimensional volumetric assessment.

# POSTERS



### N-P-01 ASIC2 AND ASIC4.2. ION CHANNELS ARE EXPRESSED IN THE RETINA OF ADULT ZEBRAFISH

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ASICs are H<sup>+</sup>-gated, voltage-insensitive cation channels that function as monitor of deviations from the physiological values of extracellular pH, but also participate in synaptic transmission, mechanosensation and nociception. Different ASICs are present in all most all the retinal layers in mammals. pH fluctuations in the retina are thought to be involved in the fine tuning of visual perception, as well as in the adaptation of the retinal responses to different light intensities. Changes in the extracellular pH are also associated to pathological conditions, and consistently ASICs are expected to participate in these processes. The zebrafish is a model to study the biology and pathophysiology of visual disorders. Nevertheless, neither the expression of ASICs in the retina of adult zebrafish nor its regulation by light has been analyzed. In this study we used PCR, in situ hybridization, Westernblot and immunohistochemistry to analyze the patterns of expression of ASIC2 and ASIC4 in the retina of adult zebrafish. ASIC2 and ASIC4.2 mRNAs were detected, localized in the inner and outer nuclear layers (ASIC2 mRNA), and in the external segment of the photoreceptors (ASIC4.2 mRNA). Westernblot detected specific proteins consistent with ASIC2 and ASIC4, and the immunohistochemical study revealed ASIC2 positive cells in the outer and inner plexiform layers, as well as in the ganglion cells layer; ASIC4 immunoreactivity was restricted to the photoreceptors. Important! These results suggest a role of ASIC4.2 in regulating photoreceptors, and of ASIC2 in the synaptic transmission of the retina.

### N-P-02 THE BDNF-TRKB SYSTEM IS PRESENT IN THE INNER CORE-AXON COMPLEX OF THE CUTANEOUS PACINIAN CORPUSCLES OF MACACA FASCICULATA

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Although it is well established that some mechanoreceptors depends totally or in part of the brain-derived neurotrophic factor (BDNF) and its receptor TrkB for development and maintenance, cells expressing these proteins at the periphery are poorly characterized, and large differences were noted among species. On the other hand, the neurotrophin dependence of Pacinian corpuscles is largely unknown. In

this study we analysed expression of BDNF and TrkB in the cutaneous Pacinian corpuscles of *Macaca fasciculata* using double immunohistochemistry and laser confocal-scanner microscopy. The results demonstrate expression of BDNF, co-localized with S100 protein, in the Schwann-related cells forming the inner core of Pacinian corpuscle. Conversely, TrkB was selectively detected in the central axon, co-localized with neuron-specific enolase. These results provide evidence for the presence and segregation of a full neurotrophin system in the inner core-axons complex of Pacinian corpuscles, and claim for the occurrence of paracrine effects within them to maintain the trophism of sensory neurons innervating Pacinian corpuscles, and presumably the Pacinian corpuscles themselves. Moreover, present data provide evidence for the large differences in the expression of BDNF-TrkB in mammalian mechanoreceptors.

### N-P-03 GENOME-WIDE ANALYSIS OF THE METHYLOME DURING NEURAL DIFFERENTIATION OF ADIPOSE STEM CELLS

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Our main objective has been to investigate the implication of DNA methylation in neural differentiation from adipose-derived stem cells (hASCs). In this study we conducted a performance analysis using methylation arrays of well characterized and defined populations of hASCs before and after induction in vitro neural differentiation allows the identification of DNA methylation of genes regulated differentiation of neurogenesis. Methods: Samples from fat obtained from liposuction will be selected and after washing in PBS, the tissue was digested with collagenase. Adipocytes were separated from stromal vascular cells after sedimentation and removed by aspiration. Cells obtained were then cultured in DMEM, and homogenous cell population was normally obtained after 3 to 5 weeks of culture. The hASCs were analyzed by flow cytometry and to induce neural differentiation, were cultured with neural differentiation medium contained EGF and FGF. Immunocytochemistry, flow cytometry and RT-PCR were used to analyze their phenotype. Characterization the genome-wide CpG profiling was performed using the Illumina Infinium HumanMethylation450 BeadChip. Results: Our results demonstrated that hASCs rapidly generated a confluent layer of elongated, fibroblastic-shaped cells. hASC were negative for CD34 and CD45 and there was a high expression of CD90, CD73 and CD105. When hASCs were cultured with EGF and bFGF, they moved slowly from a fibroblastic to a neural morphology and, exhibited highly refractive cell bodies and prominent process-like extensions. Immunocytochemical and flow cytometry analysis showed a

large increase in nestin and GFAP expression in cells induced to differentiation, and PCR studies showed that these cells expressed markers of neuronal tissue as enolase and b-tubulin III. Hybridization in methylation arrays from Illumina allowed us to identify a reduced number of differentially methylated genes: 156 CpGs hypermethylated in adipose-derived stem cells and 766 CpGs are hypermethylated in their neural derivatives. Conclusion: hASCs under specific conditions, can show neural markers normally expressed at various stages of neuronal development, support neuronal differentiation for a long time. The change in the pattern of methylation can be key to study the specific role of these genes (extracted from individual CpGs) on stem cell differentiation.

#### N-P-04 THE CYTOARCHITECTURE AND THREE-DIMENSIONAL ORGANIZATION OF THE HIPPOCAMPUS IN P73-DEFICIENT MICE

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The transcription factor p73 is a member of the p53 family and is thought to determine cell fate through a proapoptotic isoform (TAp73) and an anti apoptotic one (DeltaNp73). P73 is highly expressed by the Reelin expressing Cajal-Retzius (CR) cells of the embryonic cortex and hippocampus, and essential for their survival. The mutant mice lacking both p73 isoforms show no detectable CR cells and have a disrupted pattern of Reelin expression throughout the embryonic telencephalon. P73 *-/-* mice have a high mortality rate and a phenotype that includes cortical hypoplasia, hydrocephalus and a striking hippocampal dysgenesis. The hippocampal malformation is constant and unrelated to the severity of the other abnormalities, with its main feature being the absence of hippocampal fissure. Using cresyl violet and immunohistochemical staining against calcium binding proteins we highlight the abnormal architectonic organization of the p73 *-/-* hippocampus, and point out the differences between the dorsal and ventral hippocampus. To this end, we used SketchUp 8.0 software to construct a 3D model of the p73 *-/-* and wild type hippocampus. Considering the p73 knockout as a useful model of a fissure-lacking mouse hippocampus, our results suggest a primary role for the hippocampal fissure in determining the basic architecture of the center.

#### N-P-05 EFFECTS OF THE D3 RECEPTOR PREFERENT AGONIST PRAMIPEXOLE ON THE DOPAMINE TRANSPORTER PROTEOME

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The dopamine transporter (DAT) is a membrane glycoprotein expressed in dopaminergic neurons, which is responsible for the clearing of dopamine from the extracellular space, and is regulated by different presynaptic proteins, including DA D<sub>2</sub> and D<sub>3</sub> autoreceptors. DAT dysfunction is involved in neurological and psychiatric conditions, such as Parkinson's disease, depression and attention deficit hyperactivity disorder, in whose treatment D<sub>3</sub> receptor (D<sub>3</sub>R) ligands are increasingly being used. However, the interaction between D<sub>3</sub>R and DAT is little known. This work focuses on the effects, and mechanistic aspects, of prolonged treatment with the D<sub>3</sub>R preferential agonist pramipexole on DAT in the mouse mesostriatal system.

C57BL/6J and D2<sup>-/-</sup> mice were injected with the preferential D<sub>3</sub>R agonist pramipexole (0.1mg/kg/d for 6 days) and with either the D<sub>3</sub>R antagonist NGB2904 or the D<sub>2</sub>R antagonist L741,626. Brain samples were processed for DA uptake, western-blot, immunohistochemistry, cross-linking, A11 dot-blot, immunoprecipitation and *in situ* proximity ligation assay.

The results show that Pramipexole induces: 1. D<sub>3</sub>R-mediated D<sub>2</sub>R-independent DA uptake decrease with reduced DAT affinity but no quantitative changes in DAT expression and active uptake sites, 2. Physical interaction between DAT and D<sub>3</sub>R, and 3. Formation of DAT oligomeric complexes, ranging between 150 kDa and 250 kDa, which accumulate DAT in its homomeric form interacting with D<sub>2</sub>R and  $\alpha$ -synuclein. Similar to the DA uptake decrease, modifications in DAT interactions were prevented by co-treatment with the D<sub>3</sub>R antagonist NGB2904, and disappeared after PPX withdrawal.

Conclusion: Prolonged treatment with pramipexole induces changes in DAT interactions with its proteome-interactome partners and DA uptake decrease. These changes may be involved in the neuroprotective and antidepressant effect of pramipexole.

Supported by: BFU 2010-21130 and FP7-REGPOT-2012-CT2012-316137-IMBRAIN

#### N-P-06 ANATOMICAL CHANGES OF THE VOMERONASAL AND OLFATORY PATHWAYS, WITH PRESERVATION OF THE GNRH MIGRATORY ROUTE IN P73 MOUSE MUTANTS.

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p73 is one of the members of the p53 family. It is expressed as two major isoforms, TAp73 and  $\Delta$ Np73, with opposite functions. p73 mutant mice show hippocampal dysplasia, hydrocephalus, cortical hypoplasia, and high postnatal mortality. Loss of VR2 receptors was also described in the vomeronasal organ (VNO). We studied the p73<sup>-/-</sup> mice from embryonic age from E12 to adulthood (two years) using immunohistochemical methods. In the embryonic stage, the VNO is formed without visible malformations. Its neurons are highly positive for p73, calretinin and calbindin.

The vomeronasal nerve is indistinguishable from that of controls, and guides GnRH-positive cells during their migration into the forebrain, where we find them in septum, preoptic area and organum vasculosum of the lamina terminalis. Postnatally, the accessory olfactory bulb (BOA) is anatomically altered; its layer of reelin + mitral cells is reduced, the internal granular layer is present, but glomerular layer and periglomerular cells are absent. The absence of labeling with synaptophysin and absence of a vomeronasal nerve visible at postnatal ages leads us to hypothesize that the nerve is formed correctly in embryonic life, but is unable to establish synapses in the BOA. We attribute the behavior of the mutant animals (lack of sexual interest, lack of social contacts) with a dysfunction of the pheromone pathways. We conclude that the vomeronasal system fulfills two functions: an embryonic one, serving as guide of GnRH cells, conserved, and a postnatal one as a station of the pheromone pathway, abolished in our mutant.

Surprisingly, also the main olfactory bulb (BO) shows anatomical changes which, however, are not reflected by the animals' behavior. The mitral cell layer does not seem to be affected, but the glomerular layer is greatly reduced in size. Individual glomeruli cannot be recognized, and periglomerular cells are decreased in number and diffusely distributed.

#### **N-P-07 EXPRESSION OF RAD9B IN ANIMAL MODELS OF PARKINSON'S DISEASE AND DURING BRAIN DEVELOPMENT**

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Rad9B is a member of Rad family proteins, which shares extensive amino acid homology with Rad9A, holding its capacity to form heterotrimeric 9-1-1 (Rad9, Rad1 and Hus1), and probably its functions in the cell cycle control: detection of genomic damage, repair DNA and mediation of apoptosis. Unlike Rad9A, which is abundant in all tissues, Rad9B is expressed primarily in testis and muscle. Recent studies reveal that Rad9B is also found in the brain of mouse fetuses, although its functional meaning remains unknown. Here we studied the expression of Rad9B in the rat brain during development and in two animal models of Parkinson's disease (intracerebroventricular injection of 6OHDA in rats and intraperitoneal injection of MPTP in rhesus monkeys) using RT-PCR and in situ hybridization techniques. Our results show that Rad9B is strongly expressed in the rat brain during the prenatal life. Thereafter its expression decreases becoming absent in the adult brain. Interestingly, Rad9B expression is induced in the midbrain of 6-OHDA injected rats and MPTP treated monkeys. These data suggest that Rad9B may be involved in the pathogenesis of neurological disorders, and particularly in Parkinson's disease.

#### **N-P-08 DOPAMINE D3 RECEPTOR REGULATES LOCAL TRANSDUCTION OF THE DOPAMINE TRANSPORTER IN STRIATAL TERMINALS**

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The dopamine transporter (DAT) is a membrane protein exclusively expressed in DA-cells. DAT is responsible for the reuptake of DA from the extracellular space, so regulating DA transmission. DAT dysfunction is involved in different neurological and psychiatric diseases, such as Parkinson's disease and depression. DAT activity is regulated by different presynaptic proteins, including D2 (D2R) and D3 (D3R) receptors. The D2R/DAT interaction has been extensively studied. However, besides the increasing therapeutic use of D3R-ligands in diseases in which DAT is involved, the D3R-DAT interaction remains poorly known. The aim of this work is to investigate the effect of prolonged treatment with the preferential D3R agonist pramipexole (PPX) on DAT expression in the mesostriatal system, by using morphological, biochemical and molecular techniques. Our results show: 1. A D3R-dependent D2R-independent decrease of DAT levels in the striatum of treated animals, without changes in tyrosine hydroxylase (TH, the limiting enzyme in DA synthesis) levels. 2. No changes in DAT mRNA levels were detected in the

substantia nigra. 3. DAT and TH mRNAs are expressed in the striatum. 4. PPX induces a decrease in DAT mRNA, but not in TH mRNA. 5. The use of nigrostriatal lesions in rats and primary cell cultures shows that striatal DAT and TH mRNAs come from the substantia nigra. In conclusion, we can say that: 1. DAT and TH mRNAs are transported from the somata of nigral DA-cells to their terminals in the striatum, and 2. Besides post-translational mechanisms, DAT mRNA transport and local translation in the striatum are regulated by PPX. This mechanism may be involved in the neuroprotective and antidepressant effect of PPX.

#### **N-P-09 DEEP INTERSTITIAL CELLS OF FRONTO-PARIETAL PERIVENTRICULAR WHITE MATTER HAVE AXON PROJECTION TO ANTERIOR CINGULATE AND DORSOMEDIAL PREFRONTAL CORTEX IN ADULT RABBITS**

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In this study we observed interstitial cells of deep white matter below subplate (IdWM cells) labeled following CTb injections in areas 24b and dorsal mPFC of adult rabbits. CTb-labeled IdWM cells lay between lateral subventricular zone (SVZ) and the claustrum in frontal-parietal region. Some CTb-labeled IdWM cells were immunopositive for GABA. We also observed CTb-labelled cells in the antero-lateral SVZ; however, the morphology of these CTb-labelled SVZ cells was different from that of CTb-labelled IdWM cells. IdWM cells resulted labelled after injections in the anterior dorsal claustrum too. In prenatal and postnatal development, labeled cells after DiI injections in the claustrum and cells immunopositive for GABA, Calbindin and Calretinin lie in the ventrolateral migratory stream, from lateral SVZ towards external capsule [1, 2]. We consider that CTb-labeled IdWM cells observed in adult animals belong to the same cell class than that of cells in the ventrolateral migratory stream during development. Neurons in the white matter are actively involved in coordinating interareal and subcortical connectivity and regulation of blood flow; they have been associated with a variety of neurological and psychiatric disorders [3]. Our present results add knowledge on the nature and projections of neurons in deep white matter.

*Work funded by UPV/EHU Grants GIU07/14 and UF11/41.*

{1} Reblet C et al. (2002). *Brain Res. Bull.* 57, 495

{2} Reblet C et al. (2005). *Brain Res. Bull.* 66, 461

{3} Judas et al. (2010). *J. Anat.* 217, 381

#### **N-P-10 DEPRESSIVE SYMPTOMS ARE PRESENT IN FIBROMYALGIA INDUCED BY RESERPINE**

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Fibromyalgia is a disease of unknown etiology characterized by musculoskeletal pain, which normally appears associated with many symptoms, standing out the sleeping and mood disorders. Depressive disorders are the most common psychiatric comorbidity in patients with fibromyalgia, with prevalence rates ranging from 20% to 80%. This high coexistence suggests common pathogenic pathways, such as disturbances in the hypothalamic-pituitary-adrenal axis or decreased central monoamines. In 2009 a rat model of fibromyalgia based on central monoamine depletion by reserpine administration was proposed by Nagakura and coworkers, which produced a decrease in perceptual thresholds of pain and depressive symptoms. However, depressive symptoms results of this model were criticized for the methodology used. The aim of our study is to provide evidence of depressive symptoms in fibromyalgia model induced by reserpine using a methodology that provides more data than those described in the original protocol. For this purpose, we used the forced swimming test modified using the parameters proposed by Lucki and his team in 1996. To provide greater support for the results of this test and discard the effect of fatigue as a cause of differences in the immobility time, we employed a test traditionally used to study anxiety responses, the open field, introducing a food deprivation prior to test. Our results show a greater presence of depressive symptoms in rats with fibromyalgia in comparison to control rats. These results reinforce the validity of the model of reserpine-induced fibromyalgia, the symptoms show comorbidity is also present in humans with fibromyalgia. On the other hand, it shows the involvement of the monoaminergic system in both diseases, suggesting a common pathophysiological origin and also yields data on etiology of fibromyalgia. Also, they suggest that the use of drugs to correct the depletion of monoamines, such as duloxetine, would be of great interest in the treatment of fibromyalgia, opening the door to future studies to confirm this hypothesis.

#### **N-P-11 ALTERATION OF THE HIPPOCAMPAL OSCILLATIONS IN A RESERPINE-INDUCED MODEL OF FIBROMYALGIA IN THE RAT**

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The aim of this study was to test whether the rat model of fibromyalgia based on central monoamine depletion by subcutaneous administration of reserpine, proposed by Nagakura and coworkers in 2009, causes changes in the



oscillatory pattern of the sleep/wake cycle similar to those described in patients with fibromyalgia. The study was conducted in Sprague-Dawley male rats. Recording electrodes were chronically implanted in the hippocampal CA1, the primary somatosensory cortex and the dorsal raphe nucleus, and an EMG electrode on the neck muscles. All the electrodes were concentrated on a multichannel plastic pedestal. After a period of recovery and habituation, reserpine was administered (1mg/kg daily sc for 3 consecutive days). Control recordings were performed in the same animals during the days prior to the reserpine administration. The results showed a reduction of slow waves (0.3-3 Hz) and an increase in theta (3-8 Hz) and alpha (8-12 Hz) hippocampal oscillations in the treated group, in contrast to control conditions. After the reserpine administration, the animals also showed an increase in total sleep time, fragmented by a greater proportion of awakenings, with less alternation between slow-wave states and states with predominance of theta activity in sleep patterns. These results show a destruction of sleep architecture, which combined with the state of hypersomnia in the rat during wakefulness, suggesting the presence of restless sleep, a symptom that has recently been included as diagnostic criteria for fibromyalgia. Our results reinforce the validity of the model of reserpine fibromyalgia proposed by Nagakura and coworkers in 2009 and suggest an alteration of monoamines in the pathogenesis of fibromyalgia.

#### N-P-12 ATTENTION DEFICIT HYPERACTIVITY DISORDER (ADHD): NEUROPSYCHOLOGICAL FEATURES

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ADHD is a neurobiological entity characterized by disruptive behavior in the presence of a persistent pattern of inattention and / or impulsivity. Currently this disorder is one of the most common disorders in childhood and adolescence, and continues into adulthood in 50% of cases. Etiologically not well elucidated, but it seems clear that it is a multifactorial disorder with a genetic predisposition base and interacting with environmental factors. It is known that affected individuals show widespread cortical hypo activation, contrary to what may seem for his behavior. Structurally there is a decrease in the volume of the right prefrontal cortex, the striatum, the corpus callosum and the right cerebellum, in all cases with impaired availability of dopamine and norepinephrine.

Using the WISC-IV test, the Wechsler Intelligence Scale for Children, which has been shown as an excellent test for the exploration of cognitive abilities, we studied 20 subjects, aged between 7 and 11 years. There were two groups, control and experimental, both consisting of 6 men and 4 women, who were matched by age and gender, and even by level of schooling.

The analysis of working memory (WM) showed significant differences in the arithmetic section, but not in the other sections. Verbal Comprehension (VC) showed no differences between the groups, however, as a whole, we can speak of significant differences in the overall result. In the Perceptual Reasoning (PR), we found significant differences in cubes section, and the whole entire test. In the Processing Speed (PS) we found significant differences in all tests, with the exception of the animals test. And finally, the Intelligence Quotient (IQ) showed significant differences in overall test between the two groups.

#### N-P-13 ROLE OF THE LOCUS COERULEUS IN THE ANTIHYPERALGESIC EFFECT OF PREGABALIN

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The locus coeruleus (LC), also known as A6, is one of the main nuclei of noradrenergic brain stem. It is considered the most important center in noradrenergic descending pain control. It shows high  $\alpha 2\delta$ -subunit expression; therefore it is related to the antihyperalgesic effect of pregabalin. Our hypothesis suggests that pregabalin would block the inhibitory effect of the GABAergic neurons on the noradrenergic LC neurons, resulting in "disinhibition" of the latter, which would increase the release of norepinephrine in the spinal cord resulting in a decrease of neuropathic pain. Our goal would be to study whether the administration of pregabalin increases the expression of c-fos in the LC, as a sign of increased neuronal activity. Our results showed that pregabalin induced an intense Fos-like immunoreactivity in the LC. These results would be consistent with the proposed hypothesis because the "disinhibition" of LC neurons by the effect of pregabalin in fact leads to an activation of the LC neurons that support Fos increase observed in the LC of pregabalin-treated group. It would be very interesting to confirm the noradrenergic nature of the Fos-positive neurons detected in the LC of pregabalin-treated animals using double-label immunocytochemistry techniques Fos-TH (tyrosine hydroxylase). Our results support the involvement of LC in the antihyperalgesic effect of pregabalin.

## N-P-14 MODIFIED GEOMAGNETIC FIELDS INDUCE GENOMIC EXPRESSION CHANGES IN THE VISUAL CORTEX

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The induction of c-fos early gene is widely accepted as a functional marker of activated neurons. Characterizing the neuroanatomy of magnetoreception by c-Fos expression in mole rats, Nemeč et al. (2001) found that the mesencephalon contains neurons that are responsive to geomagnetic stimuli. In this regard, previous results in our laboratory have shown that changes of a component of the low-intensity magnetic field elicit the expression of c-Fos and BDNF in rat's brain (Rienda, 2012). Besides, recently, it has been known that the microRNA (miRNA) system could be related with the BDNF expression in the central nervous system.

The aim of this study was to evaluate the effect of changes in the geomagnetic field on the expression of miRNAs in control visual cortex in comparison with amblyopic cortex. Experimental animals were underwent to modified magnetic field and microarray analysis revealed a lower expression of miRNAs which targets plays active roles in the promotion of trophic processes in the visual cortex of amblyopic animals in comparison with controls. Among these miRNA, we found Rno-let-7b and Rno-miR-376c which diminished the expression of BDNF and  $\beta$ -synuclein, respectively. In order to confirm the active participation of these miRNAs in our experimental model, antagomirs against them were injected in the visual cortex. Preliminary results showed an overexpression of BDNF or  $\beta$ -synuclein in the cortex after the injection of antagomirs.

In the future, we will explore the effects described here to study the application of modified geomagnetic fields on processes involved in neurodegenerative diseases.

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Grants: Gobierno Vasco Saiotek (SAI12/255, SAI11/156) y Osakidetza 2012.

## N-P-15 DYSFUNCTION OF CHOROID PLEXUS IN A TRIPLE TRANSGENIC MOUSE MODEL OF ALZHEIMER'S DISEASE

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Adult 3xTg-AD mice, harboring PS1/M146V, APPSwe and tauP301L transgenes, is an animal model that mimics many critical hallmarks of Alzheimer's disease (AD). Showing cognitive deficits and other behavioral alterations at ages when overt neuropathology is not yet observed, but when intraneuronal amyloid A $\beta$ -peptide, synaptic and cholinergic deficits can already be described. This model progressively develops  $\beta$ -amyloid plaques and neurofibrillary tangles with a temporal- and regional-specific profile that closely imitates their development in the human AD brain. It has been suggested that choroid plexuses (CP) are involved in the clearance of  $\beta$ -amyloid from brain, and a malfunction of these structures could contribute to the accumulation of  $\beta$ -amyloid in brain, causing an acceleration of the pathology. In this work, we studied the role of the choroid plexus in this animal model of Alzheimer's disease.

In order to study the CP we have used confocal microscopy. The results show both an increase in low density lipoprotein receptor-related protein 1 (LRP-1) as in the  $\beta$ -amyloid 1-42, as well as a decrease in aquaporin 1 (AQP1) and transthyretin (TTR) in CP of 3xTg AD mice, compared to Non-Tg animals. These results suggest that there is a dysfunction in CP resulting in a reduced production of cerebrospinal fluid and of secretion of TTR. We have also found an accumulation of  $\beta$ -amyloid 1-42, which together with increased LRP-1, could mean that the mechanism of clearance and degradation of  $\beta$ -amyloid in the CP of 3xTg AD mice may be impaired.

## E-P-01 STUDY OF THE RETROSTYLOID SPACE IN HUMAN FETUS

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The retrostyloid space has attracted the attention of numerous researchers, anatomists and surgeons, which have studied and described the location and the structures that compound it. It has been denominated with several names as posterior parapharyngeal space (Lang, 1995), posterior latero-pharyngeal space (Testut, 1983) and posterior subparotid space of Sébilleau (Paturet, 1951). The limits of this

space have not been studied during the fetal period, thus we have analyzed 23 human fetus of 12-17 weeks of gestational age, to determine which are the structures that contribute to limit this space and to know if the elements of this area are surrounded by a specific fascia.

According to our observations, in the upper part of the retrostyloid space, it is significance to note how does not exist a clear limit between the retrostyloid and prestyloid spaces. The upper part of the retrostyloid space corresponds to a post otic and post tympanic topographical area without relationship with the pharynx. From 76 mm CRL a connective tissue, which arranged in front of the prevertebral muscles and the cervical segment of the column, limited a fascia. This fascia began to surround, in its upper part, (much better observed at 93mm CRL) not only vessels: internal carotid artery and yugular vein, but also the other elements of the retrostyloid space. The superior cervical ganglion is not included into the connective tissue, located dorsally to the fascia referred before.

We consider that the stylopharyngeus muscle is the satellite muscle of the glosopharyngeal nerve which, in its course ahead, related to its inferior aspect. Neither we observe that the glosopharyngeal nerve cross the stylopharyngeus muscle, nor had a relationship with the styloglossus muscle. In its origin, the superior laryngeal nerve constantly presented a very precise location between the internal carotid artery, the superior cervical ganglion and the vagus nerve.

#### E-P-02 EARLY DEVELOPMENT OF THE INTERCLAVICULAR BLASTEMA IN HUMAN EMBRYOS

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The aim of this research is to clarify the first stages of the development of the interclavicular blastema and its relationship with the manubrium. We examined 15 human embryos of 12.5-26.5 mm Crown-Rump Length. Different theories have been proposed to explain the development of the manubrium of sternum:

-Sternal bands only which contact each other in their cephalic ends and form an odd rudiment in the seventh weeks of development (Hamilton and Mossman, 1975);

-Two formations or rudiments, denominated suprasteral structures, which join to the sternal bands; and a middle odd structure called ventral unpaired structure (Klima,1968).

In the formation of the manubrium of sternum are involved an interclavicular blastema, located between the clavicles and is continued with them, and two intercostoclavicular blastemas which joined the interclavicular blastema to the medial ends of the first ribs, on both sides.

The sternal bands were already presents when the interclavicular mesenchyme was formed and in a first stage does not exist a connection between the first ribs and the cranial ends of the sternal bands. In a second stage, very close to the latter, a mesenchymal bonding bridge (intercostoclavicular mesenchyme) established between the lateral parts of the interclavicular mesenchymal condensation and the first rib. These intercostoclavicular formations (Rodríguez-Vázquez et al.,2013) joined very quickly in the midline. The interclavicular mesenchyme of both sides formed ventrally to the sternothyroid and sternohyoid muscles, while the sternal bands attached to the sternocostal portion of the pectoralis major muscle. The structures which form the manubrium of the sternum were related to the corresponding homologous from the sternal apparatus of the lowest class of mammals. The interclavicular mesenchyme was very similar to that in rodents and probably represents the vestige of the interclavicle (Gladstone and Wakeley, 1932). Otherwise, the topographical anatomy of the human intercostoclavicular mesenchyme seems to be an analogy of the bilateral epicoracoid structures of the sternal apparatus of the lowest mammals (Gladstone and Wakeley, 1932; Cobb, 1968).

#### E-P-03 THE BEND OF MECKEL'S CARTILAGE IN HUMAN FETUS

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The objective of our research was to establish a relationship between the bend of the Meckel's cartilage and its relationship with the torus mandibularis in human. We observed 20 mid-term human fetuses at 11-13 weeks of gestation. This stage corresponded to a critical period in which Meckel's cartilage with endochondral ossification took a bending, "intra-mandibular course" in the base of the mandible in the area of the deciduous canine and lateral incisor tooth germs.

The stages examined in the present study seemed to correspond to a critical period for the fetal development of the mandible because the morphology, especially the anterior part, displayed striking changes: the bend and the intramandibular course of Meckel's cartilage with endochondral ossification and the surrounding of the cartilage by the protruding medial lamina of the mandible under intramembranous ossification. The protuberance formed by the medial lamina, which corresponded to the posterior area where Meckel's cartilage bent and began its endochondral ossification, corresponded to the anlage of the torus mandibularis because of its morphology, topographically location and relationships.

This area was topographically located in the oral cavity, thus it was located cranially to the attachment of the mylohyoid muscle and therefore did not correspond to the mylohyoid line. Also, another aspect for its topographical arrangement was that corresponded with the location of the mental foramen in the lateral lamina of the mandible,

between the deciduous canine and the first deciduous molar germs. Location, opposite to the mental foramen, that coincided with the mandibular tori in the adult.

#### DA-P-01 DEVELOPMENT OF FORMATIVE PROGRAM AIMED TO BEGINNER TEACHERS IN THE FACULTY OF MEDICINE

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This formative project departs from an initiative of the Vicerrectorado for the Guarantee of the Quality, University of Granada that promotes the formation of the beginner professorship. The impact area of the program is the professorship belonging to the Faculty of Medicine. The experienced professorship (more than 10 years of experience) belongs to the department of Human Anatomy and Embryology (7 teachers) and the teachers beginners (less than 5 years) belong to three departments of Medicine School: Human Anatomy and Embryology (2 novels), Pharmacology (4 novels) and Biochemistry, Molecular Biology the III and Immunology (7 novels). The aim is the improvement of the educational quality, offering to the beginner professorship a basic pedagogical formation, eminently practical, which allows the incorporation of knowledge, skills and competencies that will help them grow as professionals. The realized actions have been: 1. Formative actions of general character by *Cycles of Improvement* (initial interview, identification of needs, observation by means of video-recording of the class and later analysis with the suitable instruments). We use the mentoring as system of pedagogic accompaniment of the beginner professorship that favors the critical reflection on teaching practice. 2. Formative specific activities, type seminar-workshop (Production of Educational Guides, Training tutorial, Emotional Intelligence, The TICs, Knowledge transfer, etc).

Results and conclusions. We continue detecting the usefulness of the video recording of the classes as one of the most important element to improve teaching. Some formative courses have favored teamwork among the professorship, as much between the professorship beginner, like the experienced one. We have stated that this type of work stimulates the collective commitment with the quality of the teaching. By the other hand, the mentoring shortens the curve of learning of beginner professorship. Experiences exchange and good practices between the professorship we state it as a training tool in itself. The professorship wanted to reveal three facts: 1° The lack of formation in some of the treated fields, so much of the new and experienced professorship, as the student body. 2° They requested the continuity of these courses for next years. 3° Its importance for improving the teaching-learning process.

#### DA-P-02 QUALITATIVE AND QUANTITATIVE ANALYSES OF ANATOMISTS' RESEARCH: EVALUATION OF MULTIDISCIPLINARITY AND TRENDS IN SCIENTIFIC PRODUCTION

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Our aim was to evaluate the impact of anatomy as a multidisciplinary area and to identify trends in research by anatomists over time. Data from three main sources were analyzed: SCImago Journal & Country Rank (SJR), using the number of total documents as indicator; MEDLINE (PubMed) database (1898 through October 2012), using the keyword "anatomy" in the "affiliation" field; and the Journal Citation Report (JCR), gathering impact factor and quartile data. The number of publications by anatomists increased between 1898 and 1941, followed by a reduction until 1961 and then by a marked rise to reach 36,686 between 2002 and 2012. After 1941, anatomists began to publish in journals from JCR categories other than "Anatomy & Morphology", especially after 1962. Between 2007 and 2012, only 22.23% of articles by anatomists in JCR-indexed journals were in the "Anatomy & Morphology" area and 77.77% in journals from other categories; 58% of their articles were in journals in the first and second quartiles. The contribution of anatomists to scientific knowledge is high quality and considerably greater than indicated by the SJR database. This input is especially relevant in the Neurosciences, Cell Biology, and Biology categories. In addition, more than two-thirds of manuscripts by anatomists appear in JCR-ranked publications, and more than half in the top two quartiles of the impact factor ranking. Our results show that the scientific production of anatomists has improved the quantity and quality of multidisciplinary scientific activity in different knowledge areas.

#### DA-P-03 TUTORIAL ACTION PLAN APPLIED TO THE DEGREE IN NURSING AND PHYSIOTHERAPY STUDIES

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The Tutorial Action Plan (TAP) of the Faculty of Health Sciences (FHS) has been designed in line with the guidelines provided by the University of Jaen (UJA) and with the directives marked by the European Higher Education Area

(EAHA), being mainly orientated towards incoming students in order to facilitate their integration.

However, not every newcomer participates in the TAP, due to lack of interest or awareness, and in higher grades there are other aspects, not contemplated in the TAP, brought up by the pupils which require information. Facing this scenario, it is necessary a higher involvement on the part of teachers to solve the concerns of the students throughout the different courses being taught over the years.

In this context, the Department of Human Anatomy and Embryology of the FHS of the UJA has developed a teaching innovation project orientated to the Degrees in Nursing and Physiotherapy with the following courses of action inside the TAP:

Information Day about mobility programmes and Universities with agreement reached with the UJA.

Information Day about obtaining the English Certificate B1.

Information Day, aimed to third and fourth year students, about research initiation and support grants and collaboration grants with the various departments of the FHS of the UJA.

Information Day, aimed to fourth year students, alluding to the lines of work of the final year dissertation.

Information Day, aimed to fourth year students, about career prospects and professional integration into the labour market.

Information Day, aimed to fourth year students, about the postgraduate degrees offered by the UJA.

To date, during the development of the present project, we have been able to confirm that the students matriculated in the last two school years, of both degrees, are the ones who have attended the most to these information days, and in general, the majority of the pupils attending were those studying the Degree in Nursing.

#### DA-P-04 RAPID PROTOTYPING OF EMBRYONIC STRUCTURES FROM THREE-DIMENSIONAL MODELS: A HANDS-ON LEARNING TOOL FOR ANATOMY STUDENTS

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Laboratory experiences should be based on observational and kinesthetic embodying of the three-dimensional structure of the body, the interconnections between organs and organ systems, and the structural uniqueness of the human organism. For the time being, there's no replacement for the hands-on cadaver experience of the anatomy laboratory. However, some bodily structures may be difficult to demonstrate and study by dissection, due to their small size, location, or intricateness. The developing embryo may well be seen as exemplifying this difficulty. Human embryos are not easily available for teaching purposes and, if so, they are hardly amenable to dissection. As a consequence, the available teaching resources are commonly limited to artist renditions, schematic drawings, or histological sections if available.

Here, we propose the use of rapid prototyping of digital 3D models of embryonic structures for generating physical models that can be used for the teaching and learning of Embryology. Rapid prototyping is a group of techniques used to quickly fabricate, usually by 3D printing technology, a scale model of a physical part or assembly from three-dimensional computer-aided design (CAD) data. Physical models are important resources that can be utilized for providing a better and clear insight to the students about the otherwise complex anatomy of the developing embryo. Visual and sensory learners benefit from the physical embryo models as they are able to touch, feel, and look closely at the model. Unlike commercially available ones, which are commonly roughly approximate and devoid of anatomical accuracy and detail, models generated by rapid prototyping can be designed *ad hoc* by expert learners to fulfill specific learning objectives, and digitally dissected from appropriate imaging or histological data. Models can be designed in series to illustrate stage-by-stage organogenesis, or as assemblies to be disassembled and explored by the learner.

Here, examples of so-generated models are presented, including the heart of a transversely sectioned, 33-day-old human embryo (Carnegie stage 15), as well as an embryo at early stages of neurulation (Carnegie stage 9) which was digitally dissected to demonstrate the primitive endocardial cavity, endocardial tubes, major blood vessels, notochordal plate, and somites.

#### DA-P-05 SELF-LEARNING AND SELF-EVALUATION OF HUMAN ANATOMY THROUGH THREE-DIMENSIONAL REPRESENTATIONS ASSOCIATED TOMULTIMEDIA LINKS

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Over the past decade, anatomical teaching has undergone considerable transformation and it is growing in a very narrow way together with technology and communication sciences. The aim of this study was to promote self-learning and self-evaluation of the student through a three-dimensional educational tool. All the bones that have been scanned were property of the Anatomy and Embryology Area from the University of Jaén. The 3D profiles of the bone surfaces were obtained by a 3D laser scanner (Polhemus Fastcan), and the point cloud processing was carried out with a software tool (MeshLab). Afterwards, all the different anatomical structures were marked and labeled, text and multimedia information was linked to these structures and finally, self-evaluation tool was designed. The results of this work has derived in the development of a tool which provides a 3D view of all the bone structures of lower extremity, with three-axes movement to study every detail and identification of these structures, with complementary text and multimedia information linked to these structures. The other aspect of this application, was to offer a self-evaluation tool

to the students, with several questions associated with both 3D images and complementary information. In conclusion, we have developed an information technology application focused on helping students with the acquisition of anatomical knowledge and spatial ability of the lower limb bones.

*This work has been supported by the teaching innovation project PID29B from the University of Jaén.*

#### DA-P-06 NEW PROTOCOL LATEX-BASED FOR ARTERIOVENOUS INTRAVASCULAR REPLETION IN ISOLATED HEADS OF EMBALMED CADAVERS

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The use of solidifiable masses for intravascular repletion is formally suitable in the academic world (or educational field) despite the scarce of departments of anatomy that uses them. In his methodology of complete cadavers' fixation, Thiel included the intravascular repletion with colored latex after embalming. In the light of the deficiencies found when using Thiel arterial mass, we considered to develop a new procedure of intravascular arterial repletion based on colored latex. Shortly, the procedure, called CALIVIS, was appreciated as much in graduate teaching as in different postgraduate courses. It resulted especially useful in training courses on isolated heads for neurosurgeons, whom stated the relevance of achieve the same result in the venous circuit. By technical and anatomical reasons, this new protocol was only viable in isolated heads of previously embalmed cadavers. Considering these premises, the new protocol we propose is based on: 1) cut off the head in such way that the six great cervical vessels are visible, to validate the arterial filling and to have enough length for cannulating the internal jugular veins; 2) remove the clotted blood from the venous circuit washing it with hot tap water at controlled pressure, below 0.5 bar; 3) inject the colored latex with blue for the venous circuit below 0.5 bar, and 4) remove the water excess from the interstitial space, first draining and after dehydrating the specimen with 66% ethyl alcohol, in order to reticulate the latex until its complete curing three days later. Considering the privative anatomic variability of each specimen, our results show an excellent repletion at arterial level, whereas at venous level the repletion degree is directly related with the previous extraction of the clotted blood retained inside the veins. After testing the previous use of thrombolytic substances, we obtained the best results washing the venous circuit with hot tap water at controlled pressure. As a conclusion, the proposed protocol offers highly satisfactory results for colored latex repletion of cephalic arterial and venous circuits, by means of a methodology scarcely sensitive to the technique, highly reproducible and barely vulnerable to the precedent fixation formula.

#### DA-P-07 ANTIMICROBIAL STUDY OF A NEW EMULSION FOR THE SUBMERGED OF COMPLETE CADAVERS FIXED WITH FORMALDEHYDE AND ITS REPERCUSSION IN ENVIRONMENTAL CONCENTRATION OF FORMALDEHYDE

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The most used procedure for the embalmed cadavers' conservation in the Departments of Anatomy of our surroundings is the immersion of the cadaver in a solution contained in stainless steel chests. The preservative solution mainly used is that of 10% formaldehyde, main source of formaldehyde vapors emission to the environment in the dissection rooms. The formaldehyde is also the responsible of the penetrating smell and the mucous membrane irritation of the exposed people, while causing an alteration of the morphology, consistency and color of cadaveric tissues. Additionally, the formaldehyde evaporation determines the formaldehyde concentration loss in the preservative solution, which favors bacteria, yeasts and fungi growth in those cadaver surfaces that aren't covered by the preservative solution. Our goal was to study the possibility of reducing the formaldehyde concentration of the submerged solution in order to minimize its side effects. To this aim, we elaborated a retaining solution based on a synergic emulsion of antiseptics, moistenings and aromatizing agents that were subjected to different tests. First, we made an antimicrobial inhibition test (*Mycobacterium phlei*, *Staphylococcus aureus*, *Candida albicans* y *Pseudomonas aeruginosa*) comparing our submerged solution with other preservative solutions. Secondly, some embalmed cadavers were submerged in chests filled with our experimental emulsion and some others were submerged with the habitual solution of 10% formaldehyde, in order to validate its moistening and organoleptic properties. Finally, the study was completed with the measurement of the environmental impact of our submerged emulsion and of the 10% formaldehyde solution. Our results showed that our experimental submerged emulsion had the most biocide power of the tested emulsions, moreover providing moistening and aromatizing properties, absent in the 10% formaldehyde solution. Additionally, the repercussion in the environmental formaldehyde concentration was significantly smaller when using our emulsion. From these results it is concluded that our experimental submerged emulsion, despite reducing formaldehyde concentration to less than 1%, shows more antimicrobial activity and less tissue dehydration, improves the organoleptic features and drastically reduces the environmental formaldehyde concentration.

#### DA-P-08 RESOURCES FOR ASSESSMENT AND LEARNING IN ANATOMY. PLAYING AND AVOIDING BOREDOM.

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In the higher education system, learning assessment involves monitoring of knowledge and skills. Since Human Anatomy is easily forgotten and the new education plans have less teaching hours, we need to reinforce the learning abilities of students. In order to facilitate the acquisition of knowledge and evaluate it, we have been tutoring students in small groups to present different types of work. Teamwork is necessary for the dynamics of multidisciplinary clinical teams and we want to promote collaborative work in small groups, as well as interpersonal communication skills, during Human Anatomy teaching.

To achieve all our goals we have developed different tasks that are proposed to students:

Task 1: Anatomical models and posters.

Task 2: Tutorial and explanatory videos.

Task 3: Case studies with clinical application

These activities have been done in the last 5 academic years both as independent group work and in the seminar or dissecting room with the teacher.

Results:

- We have ensured that the student does not forget the knowledge acquired.
- Collaborative work has been encouraged.
- Acquisition of manual dexterities through design of anatomical models.
- Especially encourage student motivation
- Respectful for others' work and value teamwork
- We are permitted to discuss clinical cases, which involve their professional role.
- Motivation is enhanced since the students on their own carry out these activities.

The results have been remarkable, and we are surprised because groups have competed among them and they have reach great originality and design in their works.

#### DA-P-09 CADAVER DONATION IN THE DEPARTMENT OF MORPHOLOGY OF THE UPGC

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The aim of the study was to know the evolution of the cadaver donation for teaching purposes in the Faculty of Health Sciences of the UPGC and if the donor profile has suffered modifications along the time.

We carried out an analysis of the data available since 1980 in the Department of Morphology. It were studied: a) number of donations, b) number of entries per year, c) average time between the donation and entry into the department, d) number of rejected cadavers and the most frequent cause of rejection, and e) donor profile.

It was observed an increase in the number of donors around the 2000, being the years with the highest number

of donations 2008 and 2012. The mean time between donation and reception of the body was between 4 and 6 years and the principal reason of cadaver rejection was metastatic carcinoma.

In relation to the donor profile, the nationality of the majority of donors was Spanish, followed by English and German. A lower proportion of donors were from American, African and Asian countries.

It has been a variation in the proportion of Spanish and foreign donors since 2004. There were no differences in relation with the sex, but it was observed a decrease in the age at the time of the donation in the last years.

In summary, it has been a change in the number of donations since the 2000 and in the profile of the donor since the 2004, being the currently donor profile younger and predominantly Spanish.

#### ACYS-P-01 DOPAMINE REGULATES PANCREATIC $\beta$ -CELLS SURVIVAL AT THE SAME TIME THAT MODULATES THE SECRETION OF INSULIN.

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\*MJG-B y MC-H participate in a similar way to this study.

Dopamine is a neurotransmitter that plays a critical role in neurological and psychiatric disorders and it is implicated in endocrine modulation. Insulin secretion evoked by glucose metabolism can be modulated by parasympathetic and sympathetic neurotransmitters. Dopamine precursor L-dopa in humans suffering from Parkinson disease reduces insulin secretion upon oral glucose tolerance test; moreover the antagonism of receptors participating in islet dopamine signaling generally drives increased glucose-stimulated insulin secretion. In rodents, a single injection with L-dopa results in the accumulation of dopamine in  $\beta$ -cells and inhibition of the insulin secretory responses. In isolated islets, analogues of dopamine inhibit glucose-stimulated insulin release. These findings suggest that  $\beta$ -cells might be directly responsive to dopamine. Because the presence into  $\beta$ -cells of the enzymes responsible on synthesis or metabolization of dopamine has been reported, can be accepted that dopamine exerts an auto-paracrine regulation of insulin secretion from pancreatic  $\beta$ -cells.

Because a lack of dopaminergic inhibition in the *Drd2*<sup>-/-</sup> mice induces the diminution of pancreatic beta cells mass and decreased beta cell replication in 2-month-old mice,

could be speculated that dopaminergic modulation of pancreatic  $\beta$ -cells can modulate cellular proliferation and/or apoptosis of these cells. The aim of this study is to analyze whether dopamine is a regulator of proliferation and apoptosis of rat pancreatic  $\beta$ -cells in pancreatic isolated islets after glucose induced secretion of insulin, for these propose different doses and time points of dopaminergic inhibition were assayed.

Dopamine treatment caused a significant decrease ( $p < 0.01$ ) of the percentage of  $\beta$ -cells at all times and doses studied, in a similar way, it induces a decrease in the cellular and nuclear areas of  $\beta$ -cells. In a similar way, dopamine decreased significantly the percentage of PCNA-positive cells from the total cells, and it decreased significantly the percentage of proliferating insulin-positive cells.

After double-immunolabelling for insulin and active Caspase 3 and insulin it was observed that the islets treated with dopamine showed a positive reaction to caspase, which was not the case of the control islets. The apoptotic index determined by means of ISEL method show that dopamine induced the appearance of BrdU-apoptotic boDopamine is a neurotransmitter that plays a critical role in neurological and psychiatric disorders and it is implicated in endocrine modulation. Insulin secretion evoked by glucose metabolism can be modulated by parasympathetic and sympathetic neurotransmitters. Dopamine precursor L-dopa in humans suffering from Parkinson disease reduces insulin secretion upon oral glucose tolerance test; moreover the antagonism of receptors participating in islet dopamine signaling generalizes on islet cells, but not in control islets.

*Supported by: JCyL, Gerencia Regional de Salud, BIO103/SA18/11.*

#### ACYS-P-02 PARACRINE REGULATION BY ESTRADIOL OF THE LH PITUITARY CELLS. OBSERVATIONS FROM FEMALES MICE KNOCK OUT FOR AROMATASE.

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\* EJB and FL participate in a similar way to this study.

Testosterone is transformed intracellularly into 17  $\beta$ -estradiol by action of the aromatase p450, an enzyme that is

part of the cytochrome p450 (CYP19). In our laboratory, in humans and rodents, we have described the expression of aromatase in the pituitary gland. Subsequently, these findings have been confirmed by other authors in different animal species. The role that this transformation could play in relation to the secretion of the gland remains to be elucidated, but there is evidence suggesting a physiological and pathophysiological role for pituitary aromatase. Previously, we have demonstrated the aromatase-expression in the pituitary of rats from day 17 of the prenatal development and postnatally. After puberty, around 21 days of age, begin to be marked differences between males and females. However, in old rats, the non-tumor pituitary, do not express the enzyme. Treatment with aromatase inhibitors induces morphometric changes and variations of the cellular proliferation in prolactin- or LH-positive cells. The previous findings suggest that an auto-paracrine effect is developed in the pituitary, and they suggest a defined role for the pituitary aromatase. To determine the relevance of aromatase in the modulation of pituitary function, mainly in the pituitary gland associated with reproduction, a study of LH-positive cells in the pituitary of female mice knockout (ko) to aromatase was carried out.

A selective location of positive-LH cells into the gland is described and the changes observed in every location were analyzed and compared to wild type (wt) mice. When all LH-positive cells were considered, significant increases of the cellular, nuclear and cytoplasmic areas were founded. Moreover, LH-positive cells were more abundant in ko than in wt mice. LH-positive cells in ko mice were characterized by an intensity of cytoplasmic reaction higher than in wt mice, in all regions of the gland. Although, as it is usual for this type of cells, there are two different intensities of reaction; however, for both patterns, in ko mice the intensity was stronger than in wt mice. In general, LH-positive cells were more polygonal and presented more extensions cytoplasmic prolongations, short and thick, in ko mice that the same type of cells in the pituitary of wt mice.

All the findings observed are signs of morphological cell hyperactivity, associated with an increase in the size of the cell and nuclear areas, and an increase in the percentage of positive LH cells, they suggest that local production of estradiol mediated by pituitary aromatase is necessary for the regulation of LH gonadotropic cells, exerting an inhibitory auto-paracrine regulation. This regulation is independent of the rate of circulating estrogens, since aromatase-ko female mice produce ovarian estrogens by aromatase-independent metabolic pathways.

*Supported by: FIS: PIO21803.*

#### ACYS-P-03 OPTIMIZATION OF THE EXPRESSION OF MRNA OF THE CARDIAC ISOFORMS OF TROPONIN I IN PATIENTS WITH ACUTE MYOCARDIAL INFARCTION

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The detection of cardiac troponin I (cTnI) mRNA in a bodily fluid is advantageous for the detection, diagnosis, monitoring, prognostic, or providing a predictive indicator of non-neoplastic diseases and pathological conditions of the heart, preferably acute myocardial infarction (AMI) or sub-clinical injury, and / or coronary insufficiency, including that associated with angina cTnI is considered by some authors as an excellent marker for the diagnosis of AMI, this is due to its high sensitivity and specificity of tests for detection, allowing the confirmation and exclusion of acute coronary syndrome (ACS).

A variety of methods used to detect cTnI previously as mass spectrometry, and methods of hybridization with probes labeled. have been used. The aim of the present work is to optimize a method for detection the mRNA the cTnI gene with reverse transcription polymerase chain reaction (RT-PCR) technique in plasma of patients with IAM, to identify new biomarkers which may help to the evaluation of the treatment response The population studied were 24 patients diagnosed with AMI and of 6 healthy subjects as control negative .As positive controls, we used samples the heart muscle.

Blood sample 20 millilitres were collected in one tubes containing eidetic acid (EDTA), was centrifuged and serum a was discarded of the plasma For RNA extraction for plasma we have used variation one kit in which the modification is that we add an additional organic extraction step isopropanol y acetate de amonio. We designed specific oligonucleotide primers: for nested RT-PCR.: cTnI: Sense GAG-CAGCGATGCGGCTAGG. Antisense: CGCAGGTGGGCCGCTTAAA amplify a 50 bp. GAPDH to check the integrity of the mRNA of heart muscle. 18S to check the integrity of the RNA of plasma.

We used RT-PCR assay to detect cTnI mRNA in the patients with AMI, checking that the expression of mRNA of the gen cTnI The detection of cardiac troponin I (cTnI) mRNA in a bodily fluid is advantageous for the detection, diagnosis, monitoring, prognostic, or providing a predictive indicator of non-neoplastic diseases and pathological conditions of the heart, preferably acute myocardial infarction (AMI) or subclinical injury, and / or coronary insufficiency, inclunI gave strong signals in all cases in patients with IAM, and heart muscle. No signal expressing in healthy controls. This leads to the conclusion that in patients who have suffered AMI observed mRNA of the cTnI elevator level in peripheral blood precedes to the appearance of cTnI gene in patients with AMI. Detection of circulating cTnI in mRNA by RT-PCR, can be a sensitive method for detection in myocardial injury and may have a prognostic value.

#### ACYS-P-043D ANATOMY IN SELLAR AND PARASELLAR REGION: CORRELATION BETWEEN ENDONASAL AND INTRACRANIAL VIEW.

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**INTRODUCTION:** The introduction of endoscopy in the last decades for surgical approach in the sellar and parasellar pathologies have changed the classical intracranial view. Nowadays we mixed with endonasal view.

**AIM:** To correlation intracranial and endonasal view in the same anatomical specimen for the most important structures and references in the sellar and parasellar region.

**METHOD:** We prepare an anatomical specimen with formol and latex injected (blue and red latex). Then we make coronal slices in the maxilar sinus- lamina cribosa.

We dissection sellar and parasellar region from endonasal and intracranial view. Then we take photographs at the same time from both perspectives. We show you this photographs in a 3D format.

**RESULTS:** We show you the most important structures for the surgery from endonasal and intracranial perspectives: tubérculum sella, planum esfenoidealís, sella turcica, reesus optocarotídeos, sinus cavernosus, regió retroselar, fossa interpeduncularis, cisterna prepontina , fossa pterygopalatina e infratemporalis.

**CONCLUSION:** The endonasal and intracranial dissection of the sellar and pasellar region in the same anatomical specimen and their visualization in 3D allow obtain an optimal integration and orientation of the most important anatomical references.

#### ACYS-P-05 ANATOMICAL CLASSIFICATION OF EPIDIDIMAL-TESTICULAR RELATIONSHIP IN CHILDREN WITH HERNIA OR CRIP-TORCHIDISM.

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The aim is to present a simple classification of epididymal-testicular relationship, and test and evaluate in a prospective study its practical application.

**Material and methods.** Dual study design: prospective of 358 patients from 2 months to 12 years with hernia or cryptorchidism, and retrospective to review recording of 27 patients with intra-abdominal cryptorchidism . We classify the findings into three Groups: Group 0, normal. Group I,

partial separation between epididymis and testis and Group II, absence of epididymis and/or vas.

Results: We evaluated 371 testes, and classified 184 in Group 0, 175 testes in Group I, and 12 in Group II.

The test  $\chi^2=107,295$  ( $p<0,000$ ), there are statistically significant differences between the Groups and the type of problem presented by the testis is related to their location. We analyzed the degree and type of association between the two variables using the Gamma coefficient,  $\gamma= 0,594$  ( $p<0,00$ ), statistically significant, the more proximal interruption of the descent of the testis, most likely to have morphological change.

Conclusion: This classification focuses on the relationship of the head of the epididymis and the testis. The abnormalities of relationship between epididymis and testis is bigger and more important the farther from the scrotum is the the testis, also the position of the testis affects the likelihood of having an epididymis of one or another Group.

#### ACYS-P-06 NEUROANATOMICAL STUDY OF LIMBIC CIRCUITS. WHITE FIBERS' DISSECTION IN 3D.

González López, P.<sup>1</sup> Abarca Olivas, J.<sup>1</sup>, Verdú-Martínez, I.<sup>1</sup>; Moreno López, P.<sup>1</sup>; Lloret García, J.<sup>2</sup>; Sánchez del Campo, F.<sup>2</sup>

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INTRODUCTION: Thomas Willis was the first person who described anatomy of the cortical region in medial side of the cerebral hemispheres, located between thalamus and cerebral cortex. This location was the reason because this area was renamed as limbus, a latin word which means "the limit". In phylogenetic terms, the structures of limbic system are very old. So the development of the CNS has changed the location of these structures in the human brain. At present it has a belt form in the mesial area of the cerebral hemispheres.

OBJECTIVE: Meticulous dissection of all components, white matter tracts such as gray matter nucleus, which form limbic circuit. It was made by white fibers' dissection technique.

MATERIAL AND METHODS: Five human brains were obtained fresh (within 12h postmortem). They were washed with running water and were introduced for 2-3 months in a solution of 10% formaldehyde. Afterwards they were washed for 4-6 h in warm water. Then arachnoid and contiguous vascular structures were removed. Finally the brains were frozen for 10-12 days at -15°C. Once brains were thawed, limbic region was dissected using two wooden spatulas, a suction system and micro-forceps. Every step of dissection was made with microscopic view. Each structure was imaged using 3D technique, we obtain a pair of images (right and left).

RESULTS: High accuracy was achieved in dissections, showing 3D relationship between structures of limbic sys-

tem: temporal uncus, amygdala, fimbria, fornix, stria medullaris, mammillary body, mammillothalamic tract, anterior thalamic nucleus, cingulate radiation, hippocampus, dentate gyrus, fasciola cinerea, septal area, indusium griseum, and lateral and medial longitudinal striae.

CONCLUSION: White fibers' dissection technique allows a detailed study of Central Nervous System's structures and the limbic system. Dissection proposed in this paper allows study of a much more comprehensive way than using histological sections and understand three-dimensional relationship between structures that make up limbic system.

#### ACYS-P-07 DESCRIPTIVE STUDY OF FALSE TENDONS AND MODERATOR BAND IN RIGHT VENTRICLE.

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Ventricular false tendons (FT) are fibromuscular structures considered as anatomical variations little significant. While has been associated to arrhythmias or innocent murmurs, this relationships have not been demonstrated. Most false tendons are located in the left ventricle (40-90%). In right ventricle we must differentiate from tricuspid apparatus and moderator band, a thicker structure with systolic contraction, some qualities absent in false tendons.

Of the fifty-six adult hearts that we had, only forty-one were valid for the descriptive study. The prevalence of false tendons in right ventricle is 41% (27% single FT- 14% multiple). The average length was 1.40 cm, the median 1.2 cm and mode 1.1 cm. In the case of thickness, the average was 0.13 cm, while the median and mode 0.5 cm. On the other hand, moderator band was present in 83% of hearts analyzed, the average was 1.48 cm, the median 1.5 cm and mode 1.9 cm. the thickness was 0.46 cm, the median and mode 0.4 cm. Finally, the average gradient of moderator band was 0.87 and the inclination angle was 37.7°.

To summarize, the morphology of false tendons varies in length and thickness, correlating the last one to the predominant histology. The distribution is totally random in the ventricular cavity and the prevalence is different according the ventricle studied (highest in left ventricle). Our data establish its presence in right ventricle of 41% higher than those described in other studies. Although false tendons are associated to cardiac pathologies, a great percentage of healthy people present false tendons, so we must consider them as variants of normality. From classics, the moderator band has been described as a conduction structure that contributes to the heart contraction. In this study we have shown the great prevalence as anatomic normal element describing their features.

#### ACYS-P-08 THIEL'S EMBALMED CADAVERS, AN ULTRASOUND PHANTOM TO DESCRIBE THE PECTORAL NERVES BLOCKADE.

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**Objective:** The pectoral nerves block is an easy and reliable superficial block that targets the lateral and median pectoral nerves at an interfascial plane between the pectoralis major and minor. Once the pectoralis muscles are ultrasound located under the clavicle, the space between the two muscles is dissected with local anesthetic to reach the lateral pectoral and the medial pectoral nerves. The pectoral nerves are major nerves arising from the brachial plexus innervating the pectoral muscles.

It has been described, as a suitable analgesia after breast surgery. The main indications are breast expanders and sub-pectoral prosthesis where the distension of this muscles is extremely painful. This block has been used for tumorectomies, wide excisions an axillary clearances, traumatic chest injuries, pacemakers, port-a-caths and chest drains too.

The purpose of this report is to describe how Thiel's embalmed cadavers will help to the anesthetists to know the sonographic anatomy and to learn technical skills to realize this block.

**Methods:** To study Thiel's embalmed cadavers as an useful ultrasound phantom to describe the interpectoral blockade we will use:

Thiel's embalmed cadavers. Ultrasound: Sonosite Titan®. Catheter Contiplex C 50 mm (B Braun).

**Conclusions:** Thiel's embalmed cadavers - which color, consistency and transparency of the tissue are very well preserved- can be use as a phantom to familiarize with sonoanatomy of the pectoral nerves block, in addition to learning the technique, needle placement and reliable method to detect the injectate spread of local anesthetic.

#### ACYS-P-09 COMPARATIVE ANALYSIS OF BODY COMPOSITION ASSESSMENT METHODS: KINANTHROPOMETRY, BIOIMPEDANCE ANALYSIS AND ULTRASOUND SCANNING.

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**Introduction:** The measurement of subcutaneous tissue using ultrasound can be a quick alternative to obtain the

percentage of body fat (% FM), the kilograms of fat mass (kg FM) and the kilograms of muscle mass (kg MM).

**Aims:** 1) Compare three methods of assessing body fat as they are, the kinanthropometry, ultrasonic measurement and bioelectrical impedance analysis (BIA) 2) Get a series of equations that allow to know the body composition of young and healthy adults in a fast, easy and safe way using the ultrasound technique.

**Material and Method:** Controlled, cross-sectional and analytical. We have developed two types of sample: 5 cadavers and 221 healthy young and old subjects with an age range of 15-80 years. Initially, an assessment was made with such methods in cadavers to determine the correlation between anthropometric methods (Yuhasz) and ultrasound measurements. We used the Pearson correlation for determining the relationship between fat thickness of each of the 8 zones and measurement methods used. In the sample with youth and adults were conducted multiple comparisons obtained through the Bonferroni test. To obtain the formulas to assess the FM (% and kg) and MM (Kg) with the ultrasound measurements, we performed a stepwise regression. The accuracy of the prediction equations was assessed with a validation test.

**Results:** In cadavers, there is a correlation between incisional measurement and ultrasound performed in each of the 8 anatomical zones. It is high correspondence between the incision and the skinfold areas of the biceps and leg.. In the sample of young adults, there is a statistically significant correlation (at the 0.01 level bilateral) between FM (%) obtained by anthropometric measurements, BIA, simple regression equations, multiple and the equation developed in this research (ECO). The BIA has lower values of Pearson correlation compared with other methods used for both genders and for the whole study sample.

**Conclusion:** The comparison of the methods shows that many techniques used as prediction equations FM (%) show a significant correlation in this population. In turn, these new regression equations allow use of ultrasound as a method to assess body composition in all individuals, regardless of their body mass index or physical activity level.

#### ACYS-P-10 NOVEL MARINE SPONGE-RELATED MEROS-ESQUITERPENES EXERT A POTENT ANTITUMOR EFFECT IN VITRO E IN VIVO MEDIATED BY CELL CYCLE BLOCKAGE, OXIDATIVE STRESS AND APOPTOSIS.

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The advance of drug discovery and development in oncology is mainly focused on the identification of novel

drugs able to be more selective against malignant cells, less toxic to normal cells, and effective against resistant cells. Marine organisms are considered as an important source of novel molecules with a variety of biological effects. Among them, we highlight Puupehenone and its derivatives, which belong to a family of secondary metabolites isolated from sponges of the order Verongida and genus *Hyrtios*, collected around the world. They have attracted significant attention due to their cytotoxic, antimicrobial and immunomodulatory activities. In this communication we present the antitumor properties of a new family of synthetic compounds that possess a puupehenone-related merosesquiterpene skeleton, whose synthesis procedure and antitumor effects are protected under our international patents WO2009112622 and WO2010076358, respectively. Compounds have shown a potent cytotoxic activity *in vitro* against breast, colon and lung human cancer cell lines, with inhibitory concentration 50 (IC<sub>50</sub>) in the nM and μM ranges. The most active molecule, compound 6 (C6), has an average IC<sub>50</sub> of 0.48 μM and 1.04 μM in breast and colon human cancer cell lines, respectively. Those values reflect an activity that surpass the effect of the commercial chemotherapeutic agent 5-Fluorouracil, under the same experimental conditions (10.51 μM and 3.44 μM, respectively). The values of therapeutic index showed more effectiveness of C6 in breast than in colon cancer cell lines (6.95 vs 0.44). The best effect was achieved against the MCF-7 cell line, which comprises estrogen receptor positive and p53 wild type cells. The cytotoxicity in MCF-7 was mediated by the induction of the cell cycle blockage at G<sub>0</sub>/G<sub>1</sub> phases, the production of oxidative stress and the induction of apoptosis. In addition, C6 produced a significant reduction of tumor volume in an *in vivo* model of cancer established in C57BL/6 mice after injection of E-0771 murine breast cancer cells. The results obtained suggest that C6 is a promising chemotherapeutic agent, and further studies are required to clarify whether C6 can be useful and safe for the treatment of breast cancer.

#### ACYS-P-11 IMPORTANCE OF METHYLATION PATTERNS IN THE OSTEOGENIC AND MYOGENIC DIFFERENTIATION OF ADULT STEM CELL FROM ADIPOSE TISSUE IN REGENERATIVE BIOMEDICINE.

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**OBJETIVES:** The high potential of Human adipose-derived stem cells (hASCs) to generate different adults cell lineages (muscle, bone, cartilage, nerve tissue, etc), has turned them in the last years in a mainstay in the field of tis-

sue regeneration. The ability to regenerate injured muscle and osseous structures by inducing osteogenic and myogenic differentiation allows fight off diseases. Because of that for the use of stem cells in clinical applications is very important to define the appropriate degree of differentiation of these cells and their biosafety. Recently it has been determined the significance of methylation patterns in the process of differentiation of stem cells into different tissues, and how the study of this metiloma may allow us to determine whether a cell is well differentiated.

**MATERIALS AND METHODS:** The study was performed comparing the methylation patterns of stem cells, differentiated tissues and tumor cell lines from these tissues. This is done by analyzing 27,578 CpG sites in the genome of human adipose-derived stem cells, generated by myogenic and osteogenic lineages from hASCs as well as normal and tumor cells from these tissue types, such as rhabdomyosarcoma and osteosarcoma. The modification in gene expression has been confirmed by RNA arrays and PCR techniques.

**RESULTS:** Global results obtained from methylation signals have been grouped into cancer derived lines, normal differentiated cells and mesenchymal stem cells. *In vitro* differentiation of hASCs involves methylation changes in gene clusters related to cell interaction growth and differentiation processes. The most interesting fact is that some of these genes are modified regardless of the type of cell line in which occur the differentiations. We selected three genes that represented a committed osteogenic pathway (SAA1) an established neuromuscular route (PTPRS) and a gene marker of stemness (PIWIL2).

**CONCLUSIONS:** The identification of the set of genes which change their methylation status in hASCs in relation to adult cells, tumor and during the differentiation process, can be crucial to understanding their potential in regenerative medicine applications and in critical aspects of certain diseases as tumor.

#### ACYS-P-12 NANOMEDICIN BASED IN MAGNETIC NANOPARTICLES: A NEW STRATEGY TO THE TREATMENT OF TUMORAL CELLS.

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Magnetoliposoma nanoparticles are a new strategy in biomedicine that may be used to modify cellular properties, introduce therapeutic agents inside the cell or modulate gene expression. In addition, magnetoliposomas may be used to visualize drug distribution using MRI or as a new therapeutic strategy named hyperthermia. We have developed new magnetic nanoparticles associated with liposomes

carrying 5-fluorouracil and we have assayed these particles in both normal and tumoral intestinal cells. Our magnetoliposomes nanoparticles were analyzed by X-ray diffractometry, electrophoresis, surface thermodynamics, electron microscopy, dynamic light scattering and infrared spectrometry verifying the successful association between multilamellar lipid vesicle and magnetite. Moreover, we demonstrated nule toxicity in normal (CCD-18) and tumor (T-84 from colon cancer) cells lines. On the other hand, we investigated 5-fluorouracil entrapment and release in the matrix of nanoparticles. Our results showed that 5-FU has a sustained release from the nanoparticle and that release has a significant increase after heating indicating its usefulness in hyperthermia therapy. Finally, poly( $\epsilon$ -caprolactone) (PCL)-particle associated to magnetic cores showed their ability to be included in the tumor cells in very short time suggesting their ability to improve the transport of therapeutic agents into the cell. In conclusion, our results showed a new magnetic nanoparticles that may be used as a new tool in the field of nanomedicine, especially in antitumor therapy of colon cancer.

#### ACYS-P-13A NOVEL DOUBLE-ENHANCED ANTITUMORAL THERAPY MEDIATED BY GEF AND APOPTIN GENES FOR COLON CARCINOMA.

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Colorectal cancer is a major cause of mortality, with a 5-year survival of approximately 50%. Colon carcinoma cells often prove resistant to conventional therapies, and there is an urgent need for novel alternative treatments. Double suicide gene therapy is a promising strategy for the treatment of advanced cancer. It has become an important research line in the development of gene therapy to overcome the drawbacks of single gene therapy. The aim of this study is to investigate the usefulness of a double suicide gene therapy with the two suicide genes *gef* and *apoptin* in colon carcinoma. *gef* and *apoptin* genes were cloned into a doxycycline (Dox)-regulated retrovirus-mediated gene expression system. Expression of both genes in DLD-1 cell line was confirmed by RT-PCR. Cell viability was determined with the sulforhodamine B colorimetric assay, and the cell cycle was studied by propidium iodide staining. Annexin V-FITC and PI assays were used to evaluate apoptosis and the results were confirmed by electron microscopy. The mitochondrial membrane potential was measured by JC-1 assay. Our results showed that the combined expression of *gef* and *apoptin* genes was strikingly more effective in comparison to the expression of either gene alone. Co-expression of *gef* and *apoptin* synergistically enhanced the decrease in cell viability, increasing necrosis and inducing apoptosis in colon can-

cer cells *via* the mitochondrial pathway, which can be deficient in advanced or metastatic colon cancer. Double suicide gene therapy based on *gef* and *apoptin* genes may be a candidate for the development of new colon cancer therapy.

#### ACYS-P-14 MODIFICATION OF PERIPHERAL BLOOD MONONUCLEAR CELLS IN PANCREATIC DUCTAL ADENOCARCINOMA PATIENTS TO IDENTIFY POTENTIAL DIAGNOSTIC BIOMARKERS.

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Pancreatic ductal adenocarcinoma (PDAC) is a lethal malignancy associated with extremely poor survival rates, because most cases are detected at a very advanced stage. Early detection of PDAC appears to be the most relevant and effective strategy to improve the long-term survival of these patients. The objective of this study was to study if any modification appears in peripheral blood mononuclear cells (PBMCs) in patients with PDAC. These modifications could be used to identify new markers in peripheral blood that differentiate between PDAC patients and healthy controls as a means of facilitating early detection of the disease. Peripheral blood samples from 24 PDAC patients and 24 healthy controls were analyzed by whole genome cDNA microarray hybridization. The most relevant genes were selected and validated by quantitative real-time PCR in the same set of samples. Microarray hybridization studies identified 87 genes differentially expressed in peripheral blood samples from PDAC patients, and 9 of these showed a fold change  $\geq 2$ . Out of these 9 genes, 4 were selected for analysis by quantitative real-time PCR, which confirmed the previously observed changes in median fold expression. In conclusion, PBMCs gene expression profiling can play a role in the diagnosis of PDAC. We propose a four-gene predictor set (ANKRD22, CLEC4D, VNN1, and IRAK3) for peripheral blood samples that may be useful in PDAC diagnosis.

## ACYS-P-15 ANATOMICAL ANOMALIES OF ATLANTOOC-CIPITAL JUNCTION.

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The craniovertebral junction is a transitional region of the spine that exhibits extensive structural variability. Developmental defects include a vast array of anatomical anomalies that can result in a neural compression, vascular compromise or abnormal cerebrospinal fluid dynamics.

The most frequent are:

Atlas assimilation: can result if the failure takes place during the segmentation of the region craniocervical. It can vary from an atlanto-occipital ankyloses to an internal absorption of the atlas in the occipital bone.

Anomalies of the posterior arch: Can exist different types, from total aplasia, rachischisis, hypoplasias and gaps. The most frequent are the gaps, present in the 4% of the autopsies.

Anomalies of the anterior arch: Are very rare, can be found in 0,1% of the autopsies, normally associated to a posterior rachischisis.

The most frequent variations of the atlas are: posterior ponticulus, lateralis ponticulus and posterolateralis ponticulus. They consist in bony bridges that can manage to compress the arteriae vertebrales. Another variant is the facies articularis superior atlantis bipartita, important because it must not be mistaken for a fracture of the atlas.

We review and present results to evaluate the prevalence of these anomalies as well as their implication with the symptoms of the group of patients atlas rachischisis (2 cases), anomalies of the anterior arch of C1 (1 case), bilateral absence laterales massas (1 case), platibasia (1 case) transition craneocervical abnormal (3 case) and Crowned Dens Syndrome (1 case).

The knowledge of these abnormalities is very important for a right diagnosis and surgical treatment.

## ACYS-P-16 FUNCTIONAL ASSESSMENT AND SATISFACTION SURVEY OF PARTICIPANTS COURSE OF EXPERIMENTAL LAPAROSCOPIC SURGERY IN THIEL'S EMBALMED CADAVER, BARIATRIC TECHNIQUES.

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Background: currently, the laparoscopic approach is the route of choice for a wide range of basic and advanced surgical procedures. To reduce the learning curve in advanced laparoscopic surgery, it's necessary to find new learning tools. From the Laparoscopic Institute of Alicante (ILA), we offer a practical choice as a Center of advanced laparoscopic surgical techniques using the Thiel's embalmed cadaver (TEC), as a training model.

Objective: to perform a functional assessment and measure the degree of satisfaction of the pilot course attendees laparoscopic bariatric surgery in TEC as part of professional specialization courses offered by Miguel Hernández University of Elche, through the ILA and endorsed by the Valencian Society of Surgeons.

Materials and Methods: the evaluation was completed by experts and novel surgeons for general and digestive surgery, speciality. The techniques were practiced in bariatric surgery, vertical sleeve gastrectomy and gastric bypass conversion. Data were collected through a standardized anonymous questionnaire, which used an odd rating scale according to the degree of satisfaction (1=very low, 2=low, 3=average, 4=high, 5=very high). The results are presented as percentages (%).

Results: a total of 10 participants, the general perception of TEC model was excellent. The most valuable feature was the colour of tissues. As for touch operation and the degree of coordination in the surgical management, acceptance was unanimous (10/10) because the participants felt that laparoscopic manoeuvres could be performed with the same sequence as in patients. The latest technology, equipment and laparoscopic instruments was utilised and all of them were well received, as it allowed participants to learn and try new instruments of commercial houses. In the final assessment category was evaluated with very high: 94% overall course satisfaction, with 80% for its management, the adequacy of content, materials and methods used achieved a 88% and teachers training 96%.

Conclusion: TEC appears as a suitable tool for advanced technical training in laparoscopic surgery.

## ACYS-P-17 GENETIC SEXING AND IDEAL DISCRIMINANT FUNCTIONS FOR FUTURE IDENTIFICATION OF ARCHAEOLOGICAL REMAINS ON EL HIERRO (CANARY ISLANDS)

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Sex assignment of a given bone or bone fragment is of paramount importance for the archaeologist, anthropologist and in forensic medicine, and has been performed using discriminant functions, combining several anthropometric measurements obtained from individuals with known sex. It is essential to know exactly the sex from which the measures are obtained. This is a problematic task in ancient populations, since even when the entire skeleton is available, diagnosis of sex is not 100% accurate. Sexing by genetic methods (sequencing of the first intron of the amelogenin gene) constitutes a much more accurate approach for sexing bones and may be the gold standard for further elaboration of discriminant functions, which may serve for sexing new bones dug up in future excavations. In this study we have genetically sexed 52 tibiae of prehispanic inhabitants of El Hierro, in the Canary Islands, (18 women), and then, performed discriminant functions combining several anthropometric variables. These functions show a high accuracy in sex diagnosis (94.2%, area under ROC curve=0.954 with the best of the functions), so that they allow correct sexing of tibiae or tibiae fragments (only proximal third, distal third or midshaft). Thus, genetic sexing obviates the problem of finding an accurate gold standard for the elaboration of discriminant functions for ancient bones. This method could be applied to other populations of different antiquity and different ethnicity.

#### ACYS-P-18A NON-DESCRIBED URETERAL ANORMALITY IN HUMAN

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The human ureter is a muscular tube that extends from each renal pelvis to the posterior surface of the urinary bladder. Embryonically derives from the ureteric bud, a diverticulum originated from the mesonephric duct. The ureteric bud elongates to form the ureter, and undergoes repeated branching to form the minor and major calyces.

After emerging from the adult renal hilum, running vertically down behind the parietal peritoneum that surrounds the psoas muscle, which separates it from the tips of the processes of the lumbar vertebrae. Usually enters the pelvis crossing the bifurcation of the common iliac artery, prior to the sacroiliac joint.

Ureteral alterations are many and varied, being ureteral duplication the more frequently found, but in our case we bring here a malformation that has not been described in the literature. In this case left ureter crosses to the contralateral side and flows together with the right ureter. In this raid on the opposite side of the retroperitoneal region, the anomalous ureter crosses in front of great vessels in the area. This has caused an abnormal ureter obstruction, which has led him to produce uretero-hydronephrosis with dilation of the pyelocaliceal system, giving as result a mega ureter, megacalycosis and renal atrophy, as we can see on the illustrative

pictures. However, the subject in whom we found the variant lived to be 80 years (postmortem finding), but, sure, the healthy kidney had to do the work of both kidneys, its own job and the atrophied's job. We know that this patient died because an acute heart infarction, not directly in relation with the ureter abnormality.

Knowing this abnormality is important since we can avoid renal atrophy and all clinical situations about infection and others, in terms of urine reflow, and have clinical importance as well, as aortoiliac surgery for example.

#### ACYS-P-19SCLERODERMA: ORAL IMPLICATIONS IN TWO PATIENTS

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Systemic sclerosis is a chronic disease of unknown etiology characterized for its massive collagen accumulation, fibrosis, vascular obliteration and capillary structural changes, affecting skin as well as different tissues and organs. Oral implications of this pathology are mostly detected in patients suffering progressive systemic sclerosis and most of the anatomical structures are affected.

In this paper we present two cases of patients suffering systemic sclerosis who present several oral manifestations.

##### Case 1:

Woman, 62 years old, presenting speaking difficulties and tongue induration that hinders its movement, swelling pain and dry mouth and eyes. In oral exploration we notice the dryness of the mucous membranes and teeth movement due to widening of the periodontal ligament space as shown in the radiographic study.

##### Case 2:

Woman, 75 years old with absence of skin folds, microstomia and facial telangiectasias. In oral exploration, the patient showed difficulty in mouth opening, dry, palid and bright mucosas, induration of the tongue with some depapillated areas. Dental movement and suppurating periodontal pockets could also be observed.

Other oral implications usually found in patients suffering this illness that may guide us to its diagnosis are: erosion of the mandibular angle, condyle and coronoid apophyses, xerostomia and xerophthalmia, dental caries, candidiasis, bridle shortening, fibrotic induration of the tongue and parotid enlargement.

Oral mucosa is a place where pathologies such as scleroderma may have important manifestations. Recognizing these alterations of oral anatomy may lead us to an early diagnosis of the process and this way we would be able to treat the disease as soon as possible, in both systemic and odontological aspects.

### ACYS-P-20 KINEMATICS OF THE FREE THROW IN BASKETBALL. DIFFERENCES BY GENDER

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In this study we made a kinematics analysis of basketball free throw, looking to bring a little light that may assist the physical therapist in working with players of this sport, and coach, to schedule training for improvement of this technical movement. We show gender differences as well. For that we recorded ten technical experienced players (more than ten years of practice) making a free throw, but recording and analyzed only successful shots. All of The players were measured and weighed before participating in the study in an attempt to see if there was any relationship between the cinematic parameters and physical measures. Ten nine skinfold, three bone diameters and five muscular perimeters were also taken, with the intention to show body composition, its somatotype and other indirect indexes of adiposity.

We used two HD video cameras which were placed at 90° to each other and one meter high. One of them recorded the sagittal plane and the other frontal plane motion. The video files obtained were digitized with MaxTraq program, which gave us the kinematics parameters we were interested in. So we studied the height of the ball release and lineal velocity of release, the joint range of radiocarpal joint, elbow, shoulder and knee of the dominant side, and the angular velocity of the wrist, elbow and shoulder. Mathematical analysis of the variables was made with the help of MaxMate and SPSS.

The average age of the suIn this study we made a kinematics analysis of basketball free throw, looking to bring a little ligbjects was 30.4 years ( $\pm 5.8$ ) for girls ( $\varphi$ ), and 20.2 years ( $\pm 1,3$ ) for boys ( $\theta$ ); The average height of ball release was 2.36 m ( $\varphi$ ) and 2.44 m ( $\theta$ ) and did a mean linear velocity 1.6 m/s ( $\varphi$ ) and 1,5 m/s ( $\theta$ ) with average angular velocity of the wrist of 229.8 degrees/s ( $\varphi$ ) and 223.9 degrees/s ( $\theta$ ); and different joint ranges that will be showed on full text. Anthropometrics parameters will be shown as well.

### ACYS-P-21 MYOCARDIAL EXTENSIONS OVER THE PULMONARY ARTERY SINOTUBULAR JUNCTION. IMPLICATIONS FOR THE ANATOMIC SUBSTRATE OF SUPRAVALVULAR TACHYCARDIAS.

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Purpose: Supravalvular tachycardias are a subgroup of idiopathic outflow tract tachycardias that have their origin at the level of the aortic sinuses or the pulmonary artery

(PA). The existence of ventricular myocardium in close anatomic vicinity to the ventriculo-arterial junction is a known fact. However, the presence of myocardial extensions over the level of the sino-tubular junction has not been systematically analyzed.

Methods: We examined 40 hearts of post-mortem adult humans (22 male; 56 $\pm$ 8 y/o). We performed sagittal histological sections stained with Masson's trichrome. Optical microscopy techniques were used to observe the presence of extensions of myocardium at the pulmonary valvular sinuses and over the sino-tubular junction. We measured the distance from the myocardial extensions to the basal insertion of semilunar leaflets.

Results: In all hearts, the muscle fibers around the right ventricle outflow tract joined the ventriculo-arterial junction, forming part of the muscular ring that surrounds the PA. In 8 hearts (20%), the musculature of the ventriculo-arterial junction extended in its epicardial aspect over the PA valvular sinuses. These myocardial extensions continued above the sino-tubular junction to a distance of 10 $\pm$ 2 mm from the basal insertion of the semilunar leaflets (range 7, 5-14 mm). The extensions were formed by viable myocytes surrounded by adipose tissue that reached the infundibular myocardium.

Conclusion: The presence of ventricular myocardial extensions above the PA sino-tubular junction has implications for the anatomical substrate and percutaneous treatment of supravalvular tachycardias.

### ACYS-P-22 VARIATIONS IN THE REGION OF MM. EXTENSORES CARPI RADIALIS LONGUS AND BREVIS

Carmona-Calero E.M., González-Toledo J.M., Rancel-Torres N., Hernandez-Garde I., González-Marrero I., Castañeyra-Ruiz L., Castañeyra-Perdomo, A.

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During the dissections in the Department of Anatomy was observed the presence of accessory muscle (extensor carpi radialis intermedius) in three male corpses in the right region of Mm. long and short radial extensors. The extensor carpi radialis intermedius accessorie is a muscular variant regardless born epicondyle of the humerus between the two radial muscles, and inserted into the second metacarpal. In most cases, the accessory muscles are asymptomatic and are an incidental finding during dissection, surgery or imaging techniques. In some cases, however, the accessory muscles can cause clinical symptoms, these symptoms may be related to inflammation palpable or they can be a compression mechanism of neurovascular structures. Recognition and evaluation of the accessory muscles can assist in diagnosis and treatment.



### ACYS-P-23 VARIATIONS IN THE SKULL AND THE VERTEBRAE ALONG THE SPINAL COLUMN

González-Toledo J.M., Hernandez-Garde I., Rancel-Torres N., González-Marrero I., Castañeyra-Ruiz L., Castañeyra-Perdomo A., Carmona-Calero E.M.

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**Background and Objectives:** The study of the skull and its foramina are important for anatomists, anthropologists, forensic... Also the dimensions of these holes and variations therein are clinically significant for the delicate neurovascular structures passing through them. Also, the spine is a frequent location of the normal anatomic variations, or variations in normal skeletal maturation, which may simulate pathology and result in misdiagnosis. **Material and Methods:** The study was conducted in the Department of Anatomy, Faculty of Medicine, and University of La Laguna. The materials included in the study were adults' skulls and vertebrae. All were completely macerated, degreased, cleaned, dried and photographed for later analysis

**Conclusion:** Variations in the holes of the skull and the spine are important, clinical implications and necessary knowledge for better planning of surgical treatments involving this region.

data was undertaken, establishing a confidence level of 95%,  $p < 0.005$  being the statistical significance.

Our results, with a median follow up of 24 months, show that levels of proctitis  $G > 2$ , rectal bleeding  $G > 2$ , and GU toxicity  $G > 2$  were respectively 19.56%, 15.21% and 15.2% in the first group, compared with 4.17%, 2.08% and 8.33% in the second. Statistically, significantly less total and acute proctitis, late onset rectal bleeding, anal fissure, total and acute haematuria, total and acute urinary frequency and total urinary incontinence was observed. There was no statistically significant evidence of a lowering in toxicity in terms of acute and late onset dysuria, or of a relationship to the TNM, Gleason or PSA or in function of the grade of stability according to the CBKvCT action level protocol.

Therefore, verification through CBKvCT in this series of patients is associated with a statistically significant lowering in levels of toxicity, even with use of a protocol to maintain vesical and rectal content stable in both groups. This justifies its use. Greater monitoring would be necessary in order to assess the impact of verification at the level of biochemical control.

### ACYS-P-24 CONE BEAM KV CT (CBKVCT): CONTRIBUTIONS IN THE REDUCTION OF TOXICITY IN PROSTATE CANCER TREATMENT WITH EXTERNAL 3D RADIOTHERAPY (3DRT)

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The aim is demonstrate that verification through CBKvCT in a series of patients treated with 3DRT for prostate cancer (PC) is related to a reduction in levels of toxicity.

We elaborated a non-randomized, retrospective study of two homogenous groups of patients treated between 2005 and 2008. 46 patients were verified using Electronic Portal Devices (EPID), and 48 through CBKvCT. The median age of these patients was 67 years (42-85 years old). These patients received 3DRT for localized stage PC (T1-T3bN0M0), and were prescribed the same doses, simulated and planned with the same criteria and with the same equipment. The median monitoring time was 24 months. The exclusion criteria were: treatment with oral anticoagulants, history of rectal or prostatic surgery, inflammatory intestinal disease or connective tissue disease. All patients followed the same protocol, with the objective of maintaining rectal and vesical content stable. Urinary and gastro-intestinal toxicity was determined through medical consultations according to the CTCAE.3 and the RTOG scales. Statistical analysis of



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Volume 17 · Supplement 1 · December 2013

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ISSN 2340-311X



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