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(b) Monographs; Matthews DE, Farewell VT: *Using and Understanding Medical Statistics*. Basel, Karger, 1985.

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# SYMPOSIUM PROGRAM

EUROPEAN STRATEGY FOR PHD PROGRAMS  
UNIVERSITY OF MEDICINE AND PHARMACY VICTOR BABES TIMISOARA,  
TIMISOARA, 20-22 SEPTEMBER 2010

## Key speakers from EU partner universities:

- Johann Wolfgang Goethe University Frankfurt
  - Prof. Josef Pfeilschifter
  - Prof. Erhard Seifried
  - Prof. Heinfried Radeke
  - Prof. Reinhard Henschler
- Szeged University
  - Prof. Ferenc Bari
- Medical University of Vienna
  - Prof. Rudolf Mallinger
  - Prof. Stefan Boehm

Monday 20.09.2010, SENATE HALL – Main University building, 2A Eftimie Murgu Square

14:00-14:10	<b>Marius Raica: Welcome address</b>
14:10-14:30	<b>Simona Dragan:</b> Short presentation of project POSDRU 63117, project partners and roles
<b>14:30-15:00</b> 14:30-14:45 14:45-15:00	<b>Doctoral Schools of EU partner universities - organization, function, stake holders</b> <b>Josef Pfeilschifter:</b> Frankfurt experience in developing PhD programs <b>Ferenc Bari:</b> Szeged experience in developing PhD programs
<b>15:00-15:30</b> 15:00-15:15 15:15-15:30	<b>Romanian Doctoral Schools - organization, function, stake holders from Romanian partner universities</b> <b>Carmen Bunu, Pompilia Dehelean:</b> Medical PhD School in Timisoara – past and present <b>Smaranda Iancu:</b> Medical PhD School in Iasi
<b>15:30-16:00</b>	<b>Coffee break</b> <b>Press Conference - Official launch of the project POSDRU/88/1.5/S/63117</b>
<b>16:00-17:00</b> 16:00-16:20 16:20-16:40 16:40-17:00	<b>EU perspectives of doctoral studies Joint doctoral programs</b> <b>Erhard Seifried:</b> Frankfurt-Timisoara ERASMUS Cooperation <b>Ferenc Bari:</b> Cross border HU-RO projects <b>Corina Duncescu:</b> Orpheus document
<b>19:30-21:00</b>	<b>Dinner</b>

**Tuesday, 21.09.2010 SENATE HALL – Main University building, 2A Eftimie Murgu Square**

10:00-11:00	<b>Bridging science - meeting with the PhD coordinators from Timisoara and Iasi Medical Universities</b>	
10:00-10:15	<b>Iancu Smaranda:</b> Microbiology as model of “interdisciplinary approach”	
10:15-10:30	<b>Danina Muntean:</b> Mitochondrial centrality in cardio-vascular research – a translational approach	
10:30-10:45	<b>Margit Serban:</b> Trends in paediatric research and their impact on doctoral study	
10:45-11:00	<b>Ioan Sporea:</b> How to drive a PhD student?	
11:00-11:30	<b>Coffee break</b>	
11:30- 13:00	<b>Poster session of PhD students: Prerequisites for science</b>	
13:00-14:00	<b>Lunch break</b>	
14:00-16:00	<b>Interdisciplinary approach to medical research</b>	
14:00-14:30	<b>Reinhard Henschler:</b> Experimental Models to study interaction of cellular therapeutics with the blood vessel wall	
14:30-15:00	<b>Dan Gaita:</b> University of Medicine and Pharmacy “Victor Babes” Timisoara – 10 centers for one goal: Excellency research	
15:00-15:30	<b>Heinfried Radeke:</b> Excellence clusters: Building a brand and optimising inter-faculty programs to attract excellent PhD students	
15:30-16:00	<b>Anca Miron:</b> Bridging ethnobotany and modern medicine	
16:00-16:30	<b>Coffee break</b>	
16:30-18:00	<b>Round table Vienna, Frankfurt, Szeged Best practice exchanges between Doctoral Schools.</b>	<b>Biotechnology and Immunophysiology Center, Emergency County Hospital Timisoara – Agilrom Scientific Workshop Hands-on training – Lab-on-a-chip technology – 2100 Bioanalyzer Agilent – Symposium for PhD students, coordinated by Florina Bojin</b>
19:00	<b>Dinner</b>	

**Wednesday, 22.09.2010 SENATE HALL – Main University building, 2A Eftimie Murgu Square**

09:00-11:00	<b>Opportunities within European graduate schools</b>	
09:00-09:30	<b>Rudolf Mallinger:</b> Doctoral Schools: The European Approach	
09:30-10:00	<b>Stefan Boehm:</b> The impact of Excellence Programme of Doctoral Schools at the Medical University of Vienna	
10:00-10:30	<b>Ferenc Bari:</b> The benefits of high quality Doctoral School for developing the research career for PhD students at Szeged University	
10:30-11:00	<b>Simona Dragan:</b> Perspectives of Doctoral programs at UMFVB	
11:00-12:00	<b>Concluding remarks for best practice exchanges between Doctoral Schools</b>	
11:00-12:00	<b>Visit to Pius Brinzeu Centre of Research – evaluation of the research potential</b>	
13:00-14:30	<b>Mihai Ionac</b>	
13:00-14:30	<b>Lunch</b>	

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## 1. RADIOPACITY OF FIBER POSTS

**B. Baldea<sup>1</sup>, G. Furtos<sup>2</sup>, D. Bratu<sup>1</sup>**

<sup>1</sup>*Faculty of Dental Medicine, Timisoara, Romania*

<sup>2</sup>*"Raluca Ripan" Institute of Research in Chemistry, Cluj-Napoca, Romania.*

**Objectives:** The aim of this study was to analyse the radiopacity of some glass/carbon fibers and metal post and to compare with the radiopacity of human enamel and dentin.

**Materials and Methods:** Four disks of each post (21 materials), mesiodistally sections of human molar ( $1\pm 0.01$  mm thickness) and aluminium step wedges were radiographed on dental X-ray films. After development, dental films were digitized by scan and radiopacity values were recorded for each sample. The radiopacity of the samples was expressed in terms of the equivalent thickness of aluminium per 1 mm unit thickness of material.

**Results:** ParaPost XP (Coltene Whaledent), FRC Postec Plus (Ivoclar Vivadent), Danville Ice Light (Danville), Light Post, DT Light Post (RTD), showed radiopacity values higher than enamel, Glassix (NORDIN S.A), UniCore Post (Ultradent), Danville Ice Post (Danville), ParaPost Fiber Lux, ParaPost TaperLux (Coltene Whaledent) showed radiopacity values significantly greater than dentin while ParaPost FiberWhite (Coltene Whaledent), RelyX™ Fiber Post (3M ESPE), Mirafit White, Mirafit Carbon (Hager & Werken), Fibrapost (PDSA), Saremco posts Non-Stop Fibre (Saremco Dental AG), Aestheti-Plus, DT White Post (RTD), materials showed radiopacity values lower than dentin. Composites from Reforpost Glass Fiber (Angelus), Core post - Glass fiber post, Core post - Carbon fiber post (DenMat) had radiopacity lower than dentin while the second component of these posts metal had greater radiopacity than enamel. The results recorded showed statistically significant differences (significance level = 0.05) when evaluated with One-Way ANOVA statistical analysis.

**Conclusions:** Future fiber posts are recommended to have higher radiopacity values than dentin and perhaps ideally similar to or higher than that of enamel for improved of clinical detection. The posts with a lower radiopacity than 1 mm Al could be considered sufficiently radiopaque if the posts would be cemented with higher radiopaque cement. Further works in this direction are needed.

## 2. PARACLINICAL DIAGNOSTIC VALUE FOR ENDOCARDITIS INVOLVING OF SEPSIS

**Begezsán (Loghin), I. I., Dorobăț, M. C.**

*University of Medicine and Pharmacy "Gr.T.Popa", Faculty of Medicine, Department of Infectious Diseases, Iasi, Romania,*

Infectious endocarditis is an infection of the endocardium with microorganisms present in lesions, indicating that it may be valvular endocardium, septal defects or persistent arterial channel. In terms of infection disease, we can say about this pathology that it's a particular form of systemic infection with unlimited evolution, where the endocardium is the primary focus of multiplying germs.

Although in the last half century occurred major changes regarding endocarditis, by modifying etiological spectrum, appearance of new antibiotics and new laboratory technique; also

the susceptible population has changed, and the proportion of immunocompromised persons and those with prosthetic valve or pacemaker increasing at the detriment of those with rheumatic valvular disease. Despite major advances in diagnosis and treatment, infective endocarditis remains a disease with high morbidity, and with a mortality of 20% to 30%. One cause of the high mortality of infective endocarditis is the long latency from the onset of symptoms to the definitive diagnosis of infectious endocarditis and the initiation of appropriate treatment.

This research project aims main objective to improve the paraclinical methods of investigation and highlighting new useful parameters, in order to establish the right diagnosis, complete and early of infectious endocarditis, and also to establish the treatment at the right time.

Project will be based on two-stage study:

I. Retrospective study targeting diagnosed patients with endocarditis in sepsis by classical procedure and an other group of suspected patients.

II. Clinical and laboratory correlations between the two groups which will be added to another group of prospective study (which could lead in the second stage to conclusions regarding a right and quick diagnosis).

## 3. CLINICAL APPLICATIONS OF THE BIOMOLECULAR INVESTIGATIONS, PERSONAL CONTRIBUTIONS IN ONCOLOGY

**Boldeanu Florina Maria<sup>1</sup>, Serban Margit<sup>2</sup>**

<sup>1</sup>*Regional Center for Transplant Immunology, Emergency Clinical County Hospital, Timisoara, Romania*

<sup>2</sup>*University of Medicine and Pharmacy "Victor Babes", Timisoara, Romania*

The aim of this project is to investigate residual minimal disease in oncology utilizing the relationship between fusion genes and oncological diseases, especially residual minimal disease in case of bone marrow transplantation, and the incidence of these fusion genes and their identification by molecular biology methods. After the biological samples collection, will establish the group of the patients for the study, will make hematological and biochemical analysis, thus will establish the general physical condition before and after the surgical intervention. Will perform complex molecular biology analysis to the group of patients in order to investigate the residual minimal disease

The methods used will be Southern blot analysis, PCR (polymerase chain reaction) analysis of the T receptor cellular genes, RT-PCR analysis or immunological methods (flow cytometry – is based on detection of the abnormal or unusual phenotype). We will use DNA or cDNA obtained from RNA after RT-PCR (reverse transcription of the polymerase chain reaction). Will extract DNA or RNA from the biological samples (peripheral blood is collected on EDTA) with extraction kits (e.g. Qiagen kit) or automatic method with MagNa Pure LC Roche. The DNA or RNA will be amplified by PCR.

The results will be correlated with chronic pathology clinical diagnosed, will establish the incidence of these fusion genes and residual minimal disease. The obtained results will be compared to the literature data. These results will contain also positive controls in order to eliminate the false positive results.

#### 4. THE VALUE OF ACOUSTIC RADIATION FORCE IMPULSE ELASTOGRAPHY (ARFI) FOR THE EVALUATION OF CHRONIC HEPATOPATHIES

**Simona Bota**

*“Victor Babes” University of Medicine and Pharmacy, Timisoara, Romania, Department of Gastroenterology and Hepatology*

ARFI is a new method used for the assessment of liver fibrosis, of thyroid gland and breast nodules, provided by Siemens and integrated into ACUSON S2000 ultrasound system. Unlike Transient Elastography (TE) – FibroScan, this new technique can be also performed in patients with ascites.

This research project will assess the liver stiffness (LS) in: 200 patients with chronic HCV hepatitis, 150 with HBV chronic hepatitis, 350 with cirrhosis, 100 subjects without liver disease and 200 with malignant and benign liver tumors. In patients with chronic hepatitis, liver biopsy (LB), TE and ARFI will be performed in the same session. In subjects without liver disease, LS will be assessed by TE and ARFI. The result of LS measurements by TE and ARFI will be calculated as the median value of 10 valid measurements.

In patients with liver cirrhosis LS will be determined by means of ARFI and TE and gastroscopy will be performed. Characterization of liver tumors will be based on histological examination, imaging techniques (CT, MRI and CEUS), tumor markers and LS by means of ARFI, which will be evaluated as compared to those in healthy liver tissue.

We will compare data from patients with chronic HBV and HCV hepatitis with LB and we will create a score for predicting liver fibrosis based on the noninvasive tests. In liver tumors we will try to predict the nature of tumor based on the ARFI measurements. In cirrhotic patients we will try to determine if the ARFI measurements can predict vascular decompensation, esophageal varices and variceal bleeding.

#### 5. FETAL DISTRESS RELATED WITH PATHOLOGY OF UMBILICAL CORD AND THE PLACENTA

**Brisan, C.<sup>1</sup>, Brisan, L.<sup>2</sup>, Bernad, E.<sup>1</sup>, Meszaros, G.<sup>3</sup>, Munteanu, I.<sup>1</sup>**

<sup>1</sup>*Department of Obstetrics and Gynecology University of Medicine and Pharmacy “Victor Babes” Timisoara*

<sup>2</sup>*Department of General Surgery University of Medicine and Pharmacy “Victor Babes” Timisoara*

<sup>3</sup>*University of Szeged, Faculty of Medicine*

The prenatal diagnosis of fetal distress is an important concern of the current obstetrics. The pathology of the umbilical cord and placenta is present in a significant number of cases with fetal distress. Therefore, this project aims to generate the following:

- Making a fundamental research related to investigating and analyzing the characteristics of the flow through the placental unit, giving a special attention to identifying specific phenomena that take place at this level in physiological conditions and in various pathological conditions.

- The study and description of flow phenomena in the fetoplacental circulatory system, in view of identifying key issues of

the existence of weak flow underlying the production of severe pathologies associated with fetal development.

More accurate description of the phenomenon of flow in human circulatory system shows a great impact both at scientific and social level with long-term benefits for the society.

The novelty is brought by measurements and simulations conducted to help understand and prevent pathological changes of the fetoplacental vascular system highlighting the best time to extract the fetus from the uterus before the onset of fetal distress with psychomotor echo on neural development.

Modern techniques of three-dimensional reconstruction of the placental circulatory system geometries, numerical simulation techniques to real phenomenon dimensional flow, measurements in vivo, is a rather specific means of achieving this project.

We follow these medical and scientific objectives:

- identifying the flow issues at umbilical cord blood
- highlighting the critical flow, blood flow analysis by non-invasive methods (Doppler cross); experimental flow measurements and views on real 3D models, correlation and quantification of results obtained by numerical simulation of blood flow to the cases investigated
- combination field parameters for hemodynamic circulation with placental pathological factors that determine the onset of pathological blood flow.

#### 6. CLINICAL AND EXPERIMENTAL EVALUATION OF SOME ALL-CERAMIC FIXED PARTIAL DENTURES

**Candea, A.<sup>1</sup>, Bratu, D.<sup>1</sup>**

*Department of Prosthodontics, University of Medicine and Pharmacy “Victor Babes”, Timisoara, Romania*

The aim of my PhD thesis is to study all-ceramic inlay-retained fixed partial dentures (IRFPDs), because the field is a relatively new one; they also represent an aesthetic, minimally invasive alternative to traditional fixed partial dentures using full crowns as retainers.

My first objective is to study all-ceramic inlay-retained fixed partial dentures using finite element analysis, in order to assess geometrically the most favourable design of inlay preparations which would have the best results in the case of replacing a posterior missing tooth using all-ceramic IRFPDs.

Another purpose is to carry out a comparative study on the fracture strength of all-ceramic three unit IRFPDs with yttria stabilized polycrystalline tetragonal zirconia framework and all-ceramic three unit IRFPDs made from a heat-pressed lithium-disilicate based glass-ceramic.

I also aim at conducting a study on the marginal fit of all-ceramic three unit IRFPDs.

I want to carry out a clinical study, which would enable me to evaluate the clinical performance of all-ceramic IRFPDs, especially the longevity of the restorations and the viability of this treatment method.

Should they be favourable, the results of the studies conducted within my PhD project could provide a conceptual and practical framework for the adoption of this minimally invasive method of replacing a posterior missing tooth, maximizing the preservation of dental tissue and diminishing the risks which might occur

during the tooth preparation for full-crowns. We hope to collect valuable data which will throw new light on the suitability of this type of treatment.

**Keywords:** all-ceramic, minimally invasive, metal-free prosthodontics, inlay-retained fixed partial denture, marginal fit

## 7. BODY WEIGHT LOSS USING MEAL REPLACEMENTS

**Chirila, I.**

*PhD student at "Gr. T Popa" University of Medicine and Pharmacy Iasi*

*National Institute of Public Health - RCoPH Iasi*

**Introduction.** Given the increasing incidence of overweight and therefore escalating associated health care costs, healthcare providers must discover how to effectively treat this complex condition. A topic discussion in international literature is use of meal replacements (MR) to help patients prevent weight gain, optimizing individual interventions for weight reduction and long-term maintenance. Study objective was to assessing effectiveness of MR in body weight management, for people of working age.

**Material and methods.** Overweight or obese adults followed a low-calorie diet using MR, at least six weeks and indicators were followed: size, weight, percentage of fat (bioimpedance) and body perimeters. Were proposed a customized diet, replacing two meals a day with MR formulas and the third meal was composed of conventional, balanced food. This strategy generally provided 5040 - 6700 kJ (1200 -1600 kcal) per day and slightly high protein (1.1 to 1.3 g / kg, 20-35% of energy), low lipid (15-25%) and low carbohydrate (35-60%).

**Results and discussion.** A group of 44 adults (age  $42.91 \pm 14.5$  years, BMI =  $34.3 \pm 5.7$  kg/m<sup>2</sup>; 32 female with  $92.3 \pm 14.1$  kg, fat =  $44.9 \pm 7.3\%$ , waist  $115.7 \pm 12.7$  cm, hip  $120.2 \pm 10.6$  cm and 12 male with  $107.4 \pm 23.8$  kg, fat =  $34.1 \pm 6.5\%$ , waist  $117.3 \pm 18.2$  cm, hip  $114.7 \pm 9.0$  cm) followed a customized weight-loss program for average 4.02 months ( $\pm 0.33$  SE) and the entire group lost on average 11.87 kg ( $1.06 \pm ES$ ) of body weight, 13.67 cm ( $\pm 1.58$  SE) in abdominal perimeter and 5.85 ( $\pm 0.63$  SE) in percentage of body fat.

**Conclusions.** High protein MR increase satiety and decrease food intake, being helpful for energy-restricted diets in improving weight loss with retention of lean mass.

## 8. A HIGHER SUCCESS RATE OF DENTAL IMPLANTS THROUGH PROPHYLAXIS

**Cristescu Caius**

*University of Medicine and Pharmacy "Victor Babes" Timisoara*

The purpose of the project is to develop a methodology that sums up specific and innovative prophylactic methods applied before and after the dental implant, aiming to increase the overall quality of the patient's life.

The projects stages will be implemented as follows:

1. Identifying dental practitioners specialized in dental implants.

2. Briefing the practitioners about the project, its purpose and their involvement in the project

3. Creating the patient database

4. Before the implant:

- Informing the patients about oral and dental threatening substances

- Informing the patients about proper dental brushing, tongue cleaning, oral rinsing and oral showers – practical demonstrations

- Identifying and visualizing bacterial plaque – practical demonstrations

- Applying local prophylactic age-specific measures: fluoride treatment, dental sealing

- Professional complex cleaning of the oral cavity and teeth

- Recommendations about nutrition and diet

5. Taking part at the surgical procedures preceding the dental implants ( sinus lift, bone graft, additive bone procedures, repositioning of the mandibular canal, etc - if they are necessary, depending on the clinical situation) and at the surgical insertion of the dental implant.

6. After the implant:

- Evaluating the degree of oral hygiene

- Oral rinsing and oral showers

- Prophylactic measures, age-specific

- Complex professional dental cleaning

7. Comparative study of the patients who accepted the prophylactic measures, as opposed to the patients who refused it

- Centralizing the data collected through the customized forms from the implantologists

- Data analysis and interpretation

- Conclusion

## 9. EVALUATION OF CLINICO- FUNCTIONAL STATUS AND LIFE QUALITY IN PATIENTS WITH AUTOIMMUNE RHEUMATIC DISEASES BASED ON A COMMON FUNCTIONAL INDEX

**Razvan Gabriel Dragoi**

*University of Medicine and Pharmacy "Victor Babes" Timisoara*

*Timisoara City University and Emergency Hospital – Medical Rehabilitation and Rheumatology Department*

State of the art and potential contributions to its development

The autoimmune inflammatory rheumatic diseases are caused by autoimmune pathological processes, leading to clinical manifestations and imaging biological specific. In the current doctoral study I try to find a common way in evaluating the autoimmune rheumatic diseases that result by joint destructions and specific conditions, lack in the mioartrokinetic chain(MAK) progressively increased, leading to decreased quality of life of these patients, including direct and indirect significant costs.

**Project objectives**

The research project has as main objective the improvement in the methodology for tracking biological and clinical prognosis in major autoimmune diseases with osteoarticular

destruction and musculoskeletal injury. The secondary objectives of the proposed research project are: Quantifying the contribution that brings quality of life assessment in the management of patients with autoimmune rheumatic disease. Optimizing recovery treatment in patients with autoimmune rheumatic diseases, by setting the timetable therapeutic interventions based on monitoring of quality of life scores. Identify whether the initiation of biological therapy, depending on clinical progress and determining functional prognosis index. Extension of interdisciplinary cooperation areas (medicine, physiotherapy) to develop new methodologies and solutions for improving the efficiency of the treatment in patients with autoimmune rheumatic diseases.

**Keywords:** life quality, functional index, Rheumatoid arthritis, Ankylosing spondylitis, Psoriatic arthritis

## 10. THE ROLE OF MITOCHONDRIA IN CARDIO-PROTECTION OF SENESCENT AND PATHOLOGICAL MYOCARDIUM

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Early myocardial reperfusion, the only effective therapy able to reduce the infarct size and to improve the clinical outcome, is also responsible for the lethal reperfusion injury that may paradoxically reduce the beneficial effects of revascularization. Mitochondria play a central role in initiating cell death induced by the postischemic reperfusion as well as in cardioprotection against it. Accordingly, modulation of mitochondrial function in the settings of ischemia/reperfusion (I/R) injury represents a major therapeutic target of pharmacological agents able to reduce the cell death at reperfusion. Several drugs were demonstrated to reduce lethal reperfusion injury when administrated as monotherapy in different experimental settings. However, multiple pharmacological associations represent a recently investigated therapeutic alternative aimed at increasing cardioprotection at reperfusion while decreasing the doses.

The aim of the present project is to assess the efficiency of cardioprotective strategies applied during the postischemic reperfusion in aged animals and with experimentally induced pathology (diabetes and hypertension).

The objectives of the project are: 1) implementation and validation of the methodology for studying permeabilized myocardial fibers isolated from senescent and pathological rat hearts subjected to different protocols of global I/R injury, 2) assessment the possibility to enhance cardioprotection at reperfusion in these settings by modulating mitochondrial function via the association of pharmacological agents known for their individual protective effect and 3) investigation of the possibility to recapitulate the protective effects in human permeabilized fibers harvested from patients undergoing heart surgery.

**Keywords:** ischemia/reperfusion injury, senescent and pathological heart, cardioprotection, permeabilized fibers

Research supported by the National Authority for Scientific Research grant 42-122/2008 and Fellowship Project 1.5/88/S/ID 6311.

## 11. THE QUALITY OF THE ENDOMETRIUM IN INFECTIOUS ETIOLOGY FEMALE UTERINE INFERTILITY. CORRELATIONS BETWEEN INFLAMMATORY FACTORS AND MICROBIAL AGENTS (*Chlamydia trachomatis*, *Mycoplasma hominis*, *Ureaplasma urealyticum*)

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*Department-Obstetrics and Gynecology*

**Methodology:** The research project will be investigated a total of 200 patients diagnosed with primary or secondary infertility of uterine question. It will make hysteroscopy which will remove the endometrial biopsy, and after anatomic-pathologic examination will determine whether inflammatory endometrial pathology is present (endometritis). There will also be collected samples to determine the presence in the uterine cervix, the following agents: (*Chlamydia trachomatis*, *Mycoplasma hominis*, *Ureaplasma Urealyticum*). For microbiological examinations will be used the following methods:

- Detection of *Mycoplasma hominis* and *Ureaplasma urealyticum* and sensitivity to antibiotics will make galleries using *Mycoplasma IST 2* (Bio-Mérieux, France) will be tested for sensitivity to the following antibiotics: doxycycline, josamycin, ofloxacin, tetracycline, erythromycin, ciprofloxacin, azithromycin, claritomicine and pristinamycin.
- *Chlamydia*-antigen detection will be realised with *Chlamydia Microplate EIA* - BioRad; also will correlate with the presence or absence of Ig A, Ig M, Ig G serum *Chlamydia* from patients with Nova Lisa Novatec GmbH Immunodiagnostica.

The main objective of the study is diagnosis and treatment of endometrial pathology of inflammatory and infectious, the cause of female infertility.

The secondary objectives: We will try to determine the presence of infection correlated with inflammatory markers in serum or at the endometrial biopsy sample (immunohistochemistry methods will be used for proper determination of cytokines: IL-1, IL-8, TNF $\alpha$ ). Will compare data from tests conducted on these patients and will try develop a therapeutic protocol in endometrial inflammation, the ability to restore fertility to these women.

**Keywords:** Female uterin infertility, Embryo implatation, Genital infections, Hysteroscopy, Endometrium derived cytokines and growth factor.

## 12. THE INFLUENCE OF AMBULATORY BLOOD PRESSURE PROFILE AND RISK FACTORS ON LEFT VENTRICULAR GEOMETRY

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The objective of the study was to determine the influence of high blood pressure values and profile determined by ABPM, on left ventricular hypertrophy and LV geometry.

**Material and methods:** Ambulatory blood pressure monitoring was applied to 65 patients, with age of 50 $\pm$ 6 years, with mild to

moderate hypertension, severity stages I-II, who had never been treated and had a standard echocardiographic evaluation and risk factors evaluation. LV hypertrophy was considered present when LV mass index (LVMI) was  $>125 \text{ g/m}^2$  in men or  $110 \text{ g/m}^2$  in women. Increased relative wall thickness (RWT) was present when the ratio septal wall thickness + posterior wall thickness/LV internal dimension in diastole was  $> 0.43$ .

**Results:** With regard to the ambulatory blood pressure profiles, patients were divided into two groups: group I, consisting of 36 (55.45%) patients, in which night blood pressure levels decreased by more than 10% compared to their daytime blood pressure levels (dipper) and group II, with 29 (44.6%) patients, whose blood pressure levels did not decrease as much (non-dipper). Mass index and relative wall thickness of the patients established the following echocardiography data: normal geometry, concentric remodeling, eccentric hypertrophy, and concentric hypertrophy. In the dipper and non-dipper groups' age, gender, systolic, and diastolic blood pressure showed no significant differences. Normal geometry, concentric remodeling, and concentric hypertrophy were nearly similar in both groups. In group I, normal LV geometry was present at 27.7%, concentric remodeling at 30%, eccentric hypertrophy at 8.3% and concentric remodeling at 33.3%. In group II, normal geometry was present at 24%, concentric remodeling at 13.7%, eccentric remodeling at 37.9% and concentric hypertrophy at 24.1%. Eccentric hypertrophy was higher in the non-dipper group, compared to the dipper group (37.9% vs 8.3%,  $p < 0.05$ ). Risk factors present at group II were: diabetes at 42%, dyslipidemia at 45%, smoking at 27%, obesity at 40.5% and multiple risk factors at 58.5%.

**Conclusions:** patients with mild and moderate hypertension, whose night blood pressure does not decrease enough, develop eccentric hypertrophy more often compared with the dipper hypertensive patients. Risk factors associated with eccentric hypertrophy were obesity, diabetes and dyslipidemia.

**Key words:** hypertension, dipper, non-dipper, ambulatory blood pressure monitoring, left ventricular geometry, risk factors

### 13. CONTRIBUTIONS TO THE ELUCIDATION OF TRITERPENIC COMPOUNDS ANTITUMORAL EFFECTS

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Increasing body of evidence suggests that several molecules derived from natural compounds are promising therapeutic agents against carcinogenesis. Among these triterpenes (eg., betulinic acid, betulin, lupeol and oleanolic acid) have shown to possess anti-proliferative properties against multiple tumoral cells lines. In particular, betulinic acid has been consistently associated with important biological antiviral, anti-inflammatory, antiseptic effects, and also with the inhibition of cancer cells growth. The antitumoral property of betulinic acid has been related to its ability to induce caspase activation in a mitochondria dependent fashion, as recently demonstrated in several tumor cell lines by a reduction in mitochondrial membrane potential and

cytochrome c release. Recent experimental evidence for other triterpenic compounds relates the antiapoptotic effect to reactive oxygen species overproduction that was directly responsible for the reduction in the mitochondrial membrane potential as well as mitochondrial dysfunction.

The aim of the present work is to gain further insights into the antitumoral effects in relation with the mitochondrial function/dysfunction of two natural triterpenic compounds (betulinic acid and betulin) after acute and chronic administration in transgenic mice C57BL/6 with experimentally induced carcinogenesis and plurivisceral metastasis.

The project objectives are: (i) implementation and validation of an experimental model for the study of liver mitochondria function after isolation by differential centrifugation technique; (ii) evaluation of acute and chronic administration of two triterpenoid compounds (betulin and betulinic acid) on liver mitochondria and (iii) assessment of acute and chronic effects of triterpenic compounds on liver mitochondrial suspension in transgenic mice with experimentally induced carcinogenesis and liver metastasis.

**Keywords:** triterpenic compounds, cancer, mitochondria, apoptosis, cytochrome c

Research partially supported by the Fellowship Project 1.5/88/S/ID 6311.

### 14. ELECTRONEUROPATHOLOGICAL PATTERN OF EEG BRAIN MAPPING IN HYPERTENSIVE DISEASE ASSOCIATED WITH PREEXISTING PREGNANCY

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The research project will be evaluated in terms of quantification and localization of brain electrical activity by topographic EEG mapping a group of patients including women with hypertensive disease associated with pregnancy -especially without obvious clinical manifestation framed in the category of pregnant women with obstetrical risk. It will consider highlighting and spatial distribution of electroneuropathological pattern of brain activity in focal/ or diffuse lesions of hypoxic encephalopathy. In consecutive maternal monitoring study group will include both a balance hypertension clinical-HTA curve, weight curve, diuresis, subjective complaints, and one paraclinical cardiovascular, kidney, liver and blood chemistries changes. Fetal monitoring surveys will be performed by repeated clinical obstetrics investigation, as well as by ultrasound assessment of fetal wellbeing, including placental maturity degree and Doppler velocimetry of umbilical or cerebral fetal artery. It will aim to establish statistical correlation between the anomalies detected by EEG MAPPING and clinical/paraclinical accusations-with or without changes in blood pressure, pathological ultrasound aspects of umbilical/cerebral artery IR, fetal IUGR without an identifiable cause, and whether these changes can be a predictive factor for maternal and fetal prognosis

**Key words:** pregnancy, EEG mapping, hypertensive disease

## 15. EVALUATION OF SUDDEN DEATH RISK IN ATHLETES, DIAGNOSIS AND ELECTROPHYSIOLOGICAL TREATMENT

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**Background and Purpose:** Sudden death in athletes is insufficiently studied in relation with its gravity, in terms of subjacent mechanisms, triggering, diagnosis, risk stratification and treatment of the various underlying arrhythmias. In our previous experience a significant number of the acquired and congenital heart diseases, were strongly associated with sudden death risk (mostly during exercise). We consider that the actual screening chart for athletes by standard and effort ECG is not enough accurate to assess those at high risk.

**Methods:** The present study proposes the inclusion of minimum 80-85 athletes who consent to electrophysiological screening in presence of any rest or effort ECG changes, any clinical history of arrhythmia (presence of at least one anamnesis or documented episode), resuscitated cardiac arrest or syncope, in which the electrophysiological study will evidence and trigger any form of tachycardia. Depending of the arrhythmogenic substrate the radiofrequency catheter ablation will be tempted in selected cases.

**Expected results:** We estimate the inclusion of 80-85 patients with proved significant rhythm disorders. A risk chart will be provided after statistical analysis of the study group, which will consider the benefits of additional investigations (ECG, echocardiography, and electrophysiological study), the mechanisms and the complex management of cases after diagnosis.

**Conclusions:** The athletes at high risk, malignant arrhythmias of all types have maximum indication of exploration, guided by a thorough algorithm and adequate treatment.

## 16. INTEGRATING MEDICAL DATA IN THE ELECTRONIC HEALTH RECORD

Leonard Mada

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**Introduction:** Existing Electronic Health Records are largely confined to a single or a limited number of providers, missing the numerous interactions between different medical specialties and health care providers within a modern health care system. In modern health care, the medical information is actively exchanged between the three levels of the system, primary health care, specialty outpatient care and inpatient care. Additional data stemming from peripheral sensors and directly from the patient will become more prevalent in the future.

**Objectives:** The primary objective is to develop the principles and mechanisms for automatic semantic and temporal integration of all available medical data in a unique electronic health record. Secondary objectives include the semantic and temporal standardization of medical data to enable secondary reuse of this data.

**Materials and methods:** Applying a formal analysis of semantic

and spatio-temporal relationships in medical data and generalization and abstractization of data types and relations. Extending medical ontologies on the generated model. Defining interactions between data objects. Developing standardized query mechanisms for secondary reuse of medical data, including epidemiologic studies, quality management, automatic evaluation of loco-regional, temporal, genetic and other risk factors; facilitating clinical trials, including enhanced electronic recruitment, automatic monitoring of trial subjects and enabling pharmacovigilance studies.

## 17. LONG TERM PROGNOSIS OF PATIENTS AFTER HEMATOPOIETIC STEM CELLS TRANSPLANTATION

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**Background:** Hematopoietic stem cell transplantation (HSCT) is a life saving procedure, for many severe diseases, without other therapeutic alternatives. In Romania it was initiated in Bone Marrow (BM) and Hematopoietic Stem Cells (HSC) Transplantation Center of the Children’s Emergency Hospital “L. Turcanu” Timisoara with autologous transplant (2001), allogeneic related donor (2003), allogeneic unrelated donor (2009), allogeneic stem cells transplant from umbilical cord (2009). The purpose of this study is to develop a protocol for screening, monitoring and diagnosis of secondary morbidity of HSCT and identify the factors that can improve the long-term prognosis of the patients and increase their quality of life.

**Objectives:** 1. Secondary pathology detection of HSCT by dynamic analysis of clinical and biological parameters following a predetermined algorithm to clearly identify the factors influencing long-term prognosis of patients with HSCT. 2. Identifying factors that cause secondary pathology of HSCT. 3. Impact on quality of life of the developed protocol as a result of identifying factors that influence secondary pathology of HSCT.

**Study Material:** The study will be retrospective and prospective: patients who underwent HSCT since 2001 and patients that will be transplanted until the end of 2012 in BM and HSC Transplantation Center from our hospital. As part of the study, I will perform a descriptive and predictive experimental, multidisciplinary model, on Sprague Dawley rats. This should determine whether brain natriuretic peptide (BNP) has predictive value for veno-occlusive disease, cardiac dysfunction and major hydroelectrolitic imbalances on rats after HSCT.

**Key words:** hematopoietic stem cells transplantation, long term prognosis, quality of life, brain natriuretic peptide

## 18. THE INVESTIGATION OF CARBON MONOXIDE INTOXICATION IN FORENSIC MEDICINE

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Carbon monoxide is a colorless and odorless gas generated in the

incomplete combustion of organic substance. It binds by the hemic protein, including hemoglobin (the affinity is 210 times bigger than for the oxygen) and myoglobin, replacing the oxygen and reducing its capacity to release oxygen, which leads to hypoxia.

The objectives of my research are to implement a technique of work to determine the carboxihemoglobin concentration by electrophoresis method (the determination of carboxihemoglobin is made, usually, by spectrophotometric method) and is to implement a technique of work to determine the concentration of carboxymyoglobin (carbon monoxide binds by hemoglobin and myoglobin). Another objective is to determine the concentration of hydrocyanic acid and carboxihemoglobin in the blood of the victims of a fire, to determine if the death occurred after carbon monoxide poisoning or due to hydrocyanic acid poisoning, or because of the combination of them. I also want to determine the lethal concentration of carboxihemoglobin taking into account various parameters (age, sex, associated diseases, ethanol intoxication or intoxication with other substances). I want to determine the concentration of carboxihemoglobin in the blood kept at different temperatures (blood stored for 15 minutes at 50°C, at 70°C and at 90°C) and see the relationship between a given storage temperature and the possibility to still determine the carboxihemoglobin.

## 19. RESTORING TOTAL EDENTULOUS PATIENTS WITH PROSTHETIC RESTORATIONS ON SPLINTED IMPLANTS

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The aim of my PhD thesis is studying the behavior of different types of prosthetic restorations (fixed or removable) supported on splinted implants in totally edentulous patients.

Usually before loading implants need a rest period of 3 or 6 months (mandible respectively maxilla), to osseointegrate. Rigid splinting reduces mechanical stress exercised on each implant, increases primary stability of the pluri-implant structure, which allows immediate loading with a provisional restoration.

One of the purposes of my research project is to register tensions that occur at the interface between Mezostructure (abutments splinted with a bar) and Superstructure (Prosthesis), when the Prosthesis is subjected to forces of insertion and desinsertion, similar to those applied by the patient.

Another goal of the project is to compare how the occlusal pressures are distributed in bone to implant interface (after splinting the abutments), being known that after loading the implants, around their necks appears a conoidal bone resorption, especially in the first year of function.

Stress at the bone - implant interface will be registered in two situations: when implants are splinted with a bar made in dental laboratory and when implants are splinted by cold welding of the titanium bar, directly intraoral. The process of cold welding is called syncrystallization, and the device used for this purpose is called SINCRISTALLIZZATRICE. This method could simplify and shorten implant prosthetic treatment, the stage for manufacturing the bar in dental laboratory, no longer being needed.

The results obtained from recordings made with these studies

will allow conclusions to be drawn about how effective is splinting implants in supporting prosthetic restorations.

## 20. IMPROVING MATERNAL RISK PREDICTION IN PATIENTS WITH HYPERTENSIVE DISORDERS OF PREGNANCY BY ASSESSING THE ROLE OF CLASSICAL AND NOVEL ULTRASOUND TECHNIQUES IN DIAGNOSTIC AND PROGNOSTIC PROTOCOLS

Dan Pascut, Rodica Avram

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Hypertensive disorders represent the first cause of medical complications during pregnancy. Our aim is to optimize the protocol for imaging diagnosis and prognosis in hypertensive disorders of pregnancy (HDP) by assessing the utility of Tissue Doppler echocardiography (TDE), which would considerably improve the outcome of these patients (p) both during pregnancy and postpartum.

This study will be conducted in close collaboration with colleagues at the Clinical Hospital of Obstetrics and Gynecology Dr. Dumitru Popescu Timisoara (CHOGDPT) over a period of 3 years. We expect to include at least 300 p. Hemodynamic and clinical parameters, lab tests, ECG, Doppler imaging of peripheral circulation and uterine artery, classical trans-thoracic echocardiography and TDE will be recorded, assessing the role of EDT in predicting the risk of mortality and morbidity in HDP. Various parameters (body mass index, excess weight during pregnancy, ultrasound parameters) will be assessed as potential risk predictors for the occurrence of complications.

The lot will be classified in p with chronic hypertension, gestational hypertension, early preeclampsia (<34 weeks gestation), late preeclampsia (> 34 weeks) and preeclampsia superimposed on chronic hypertension. P will be followed until the end of pregnancy and at 1 and 6 weeks postpartum according to CHOGDPT and present study protocols. Using this multidisciplinary approach, we also aim to find eventual correlations between utero-placental, fetal and maternal circulation.

## 21. INTERFACE STUDIES OF PARTIAL FIXED PROSTHESIS

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This research project's objectives are the improving of quality and adhesion of partial fixed prostheses interfaces made from different materials. These objectives are: testing the physical parameters of prostheses, mechanical resistance, shear bond strength, through invasive and non-invasive techniques. Investigation methods involved are PC software numerical simulations, imaging investigations involving optical microscopy, optical coherent tomography in Time Domain mode, Spectral Domain, Micro Computer Tomography, Electronic Microscopy. Partial fixed prostheses can be made

from different materials which can assure a different quality of prosthesis. The interfaces between materials involved in performing prostheses, are metal-polymeric, metal-ceramic and all ceramic. Probes are divided depending on the material from which they are made, the conditioning and testing protocol, are proposed for this project.

The special part of this paper intends to investigate the interfaces through non-invasive technologies as Optical Coherent Tomography, Micro-computer Tomography, Optic and Electronic Microscopy and chemical analysis. These investigation methods will test metal-ceramic and all ceramic partial fixed prostheses which were conditioned through different methods for improving adhesion.

The researches will target conclusions with clinical applicability and will try to obtain positive long term effects through bracing the quality and thorough research into the mentioned domain.

## 22. MODERN THERAPEUTIC APPROACHES IN ARTERIAL OCCLUSIVE DISEASES OF LOWER LIMBS

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### Background:

Transplantation of various bone marrow derived STEM cells lead to engraftment of the cells in the transplanted area, with proven evidence that indicates the possibility of generating specialized cells according to the area needs, contributing to regeneration of tissues, organs etc

### Objectives:

The goal of the study is to induce angiogenesis in affected lower limbs, aiming to modify the following medical conditions: reduce the need of limb amputation, increase vascular perfusion index, reduce distal trophic lesion, increase the distance of independent walk, as well as pain reducing.

The overall output aims to increase the quality of life of affected patients and to offer valid alternative to surgical treatment. These methods would give a long-term positive effect for these potentially invalidating diseases.

### Methods:

The study protocol will follow a classical clinical approach. We will start with the selection of subjects, subsequently dividing them randomly into two (case and control) groups. Evaluation of the detailed individual state following well established parameterization. The procedure consists of bone marrow from iliac crest extraction and immunophenotyping, followed by intramuscular injection of the aspirate near vascular lesion. After the procedure both groups will be monitorized for healing process evolution.

### Expected results:

Identification of an efficient treatment for PAOD and the type of adult STEM cells most potent in angiogenesis.

**Keywords:** Peripheral Arterial Occlusive Diseases (PAOD), neovascularization, adult stem cells, autologous bone marrow transplant

## 23. CLINICAL ASPECTS AND OUTCOME OF DEPRESSIVE EPISODES IN RECURRENT DEPRESSIVE DISORDER VERSUS DEPRESSIVE EPISODES IN BIPOLAR DISORDER

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The general objective of this study is to identify potential differences in clinical symptoms and outcome between depressive episodes in recurrent depressive disorder and depressive episodes in bipolar disorder.

The research results will enhance specific knowledge regarding the depressive episode in bipolar and recurrent depressive disorder, favoring the technological transfer in the diagnostic domain (clinical and standardized), the drug therapy domain (benefit which could be taken into consideration by the pharmaceutical industry) and the non-drug therapy domain (benefits that can be taken into account by the psycho-social practices).

Another aim of this study is to disseminate the obtained information by publishing it in articles, journals and presentations at specific scientific events (both national and international) which can lead to new collaboration possibilities in the specific research field.

Research results are expected to be significant, both clinical as well as the neurocognitive, retrieval of profiles of depressive episodes may lead to both a better understanding of the disorder and the correct diagnosis of the first episodes of disease and therefore optimize therapeutic strategies to enable a good remission and stability over time and thus improve the quality of life of the patients.

## 24. QUALITATIVE AND QUANTITATIVE ANALYSIS OF BACTERIAL AEROSOLS IN DENTAL PRACTICE, INDOOR AIR QUALITY IMPROVEMENT ALTERNATIVES

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The spread of infection has long been considered one of the main concerns in the dental community. Indeed, infectious agents may be transmitted to patients and dental staff via several vectors, including instruments and air. Bioaerosols are an important consideration for infection control and occupational health, as these particles may consist of or convey potentially hazardous microorganisms, allergens, or other toxic substances. Infective causative agents may include bacteria, viruses, fungi, and possibly even prions.

Although the existence of dental and microbiologic aerosols has been known for a long time, the scientific analysis of the role they have in dentistry has been investigated only recently. Furthermore, data concerning bacterial contamination of air in multichair dental clinics, such as those found in dental schools, hardly exists.

The main objective of this study is to investigate qualitatively and quantitatively the bacterial aerosols before and after therapeutic

procedures within dental practices.

## 25. PHARMACOLOGICAL AND CLINICAL STUDY ON ANTI-INFECTIOUS DRUGS IN PERINATAL PERIOD

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### Introduction

In paediatric pharmacology, the perinatal period is a topical field that has yet to be explored more thoroughly. The first part of the thesis, which deals with the current stage of the knowledge of the perinatal period, comprises a study of data in the literature of the field regarding the pharmacokinetics of the maternal-foetal unit, with an emphasis on the mechanisms of drug transfer across the placental barrier and the mechanisms of anti-infectious drugs excretion in the breast milk.

### Objective

The second part of the thesis – personal contributions – includes clinical studies that evaluate the placental transfer of anti-infectious drugs and their transfer in the breast milk, by determining their concentration in human serum and breast milk with HPLC (high-performance liquid chromatography). Comparisons will be made among drugs of the same class and among known drug representatives and their modern forms. The main objective of the study is to establish a correlation between the levels of anti-infectious drug concentration in maternal serum, in the umbilical cord blood and in breast milk. Therapeutic efficacy will be estimated with specific laboratory tests both in the pregnant woman and the new-born. The therapeutic safety profile will be monitored by registering the possible adverse effects in relation to the administered dose, with a view to prevent toxic effects in the new-born. These will be correlated with the levels of drug concentration.

## 26. IMMUNOGENETICS STUDIES AND THERAPEUTIC CORRELATIONS IN IDIOPATHIC JUVENILE ARTHRITIS

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**Background.** Juvenile idiopathic arthritis (JIA) is the most common chronic autoimmune disease in paediatric pathology. JIA has still no reliable biomarkers for monitoring disease progression.

**Objectives.** The aim of this project is to study the correlations between biomarkers profile of the patient and the clinical subgroup of JIA, inflammatory syndrome, radiologic modifications and the evolution under the treatment of the patient.

### Study Material

The design of the project includes a retrospective and a prospective study. The retrospective study, based on a cohort of 58 patients with diagnosed JIA (in the period 2004-2009, in the First Pediatric Clinic, Timisoara) will evaluate the prevalence of rheumatoid factor (RF), anticyclic citrullinated peptide antibodies

(anti-CCP) in comparison with a control group without articular pathology. These biomarkers will be correlated with the clinical form, the inflammatory syndrome and the diseases evolution under therapy. Treatment's efficiency will be measured by DAS28 score, according to EULAR, and ACR score, according to ARA's recommendations.

The prospective study will include the new patients with JIA. Patients will be evaluated physical, biological (inflammatory syndrome), paraclinic (bone density DEXA), hormonal (prolactin, testosterone), immunological with determination of the two isotopes of RF (IgA, IgM), the study of anti-CCP antibodies (IgA, IgM, IgG), C3 and C4 components of the complement. The results will be compared with a control lot.

Cytokines profile, pro- and anti-inflammatory, it will be performed using the multiplex immunoassay method, on two lots: in children with AJI in remission respectively in patients with active juvenile arthritis.

The major histocompatibility complex HLA (DR4, DR8, DR5, B-27) will be studied depending on AJI subgroup.

**Keywords:** juvenile idiopathic arthritis, anti-CCP antibodies, rheumatoid factor, cytokines

## 27. THE ROLE OF MITOCHONDRIA IN PATHOGENESIS OF EXPERIMENTAL ENDOTHELIAL DYSFUNCTION

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Cardiovascular disease is a leading cause of morbidity worldwide and endothelial dysfunction (ED) represents both a widely investigated underlying mechanism and a therapeutic target. Endothelial mitochondria have emerged as central regulators of the calcium-dependent balance between reactive oxygen species (ROS) and nitric oxide (NO) production but the contribution of mitochondrial damage to ED is not known. Increased production of mitochondrial reactive oxygen species (ROS) clearly contributes to ED pathogenesis, albeit the most relevant sources/sites are yet to be elucidated. Recent data have incriminated monoaminooxidases (MAO) with two isoforms, A and B, as important source of deleterious hydrogen peroxide production. Increased activity of myocardial MAO-A, the predominant isoform in the cardiovascular system, has been reported to occur with ageing and contribute to both acute ventricular dysfunction associated with postischemic reperfusion, and to chronic maladaptive evolution from myocardial hypertrophy to heart failure. However, information regarding the involvement of vascular MAO-A in cardiovascular pathology with respect to endothelial dysfunction are rather scarce.

The aims of the project is to evaluate the contribution of ROS generated by the MAO-A isoform at the level of vascular endothelium as: (i) second messengers for eNOS activation and redox signaling and (ii) putative contributors to the development and progression of experimental endothelial dysfunction in pathological conditions pathologies that share oxidative stress as a common denominator.

Project objectives are as follows: 1) Implementation and validation of an experimental model to study function/dysfunction of endothelial mitochondria (respiratory function and organ bath studies of vascular rings); 2) Assessment of MAO-A contribution

to the: (i) redox signalling, including NO cross-talking (if any) and (ii) pathogenesis of experimental endothelial dysfunction, including the possibility to modulate the phenomenon with MAO-A inhibitors.

Research partially supported by the Fellowship Project POSDRU/88/1.5/S/63117

**Key words:** endothelial dysfunction, mitochondria, monoamine oxidase, nitric oxide

## 28. AN IN VIVO EVALUATION OF SOY ISOFLAVONOID GENISTEIN, INCORPORATED IN RAMIFIED CYCLODEXTRIN COMPLEX ON EXPERIMENTAL B16 MOUSE MELANOMA

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Genistein is an isoflavonoid, phytoestrogenic compound found in soybeans, the vegetal product from the plant *Glycine max* (L.) family Fabaceae. This compound is a specific inhibitor of protein tyrosine kinase and was found to exhibit chemopreventive, cardioprotective and antiosteoporosis activities. One of the major problems of this active compound is its poor water solubility.

Genistein was acquired from Extrasynthese(France), hydroxyl-propyl-beta cyclodextrin (HPBCD) from Cyclolab Hungary, and B16F0 melanoma was acquired from ECACC and Sigma Aldrich, origin Japan stored UK. C57BL/6J mice were purchased from Charles River (Germany). In order to improve the solubility of genistein a cyclodextrin complex was prepared. Genistein was incorporated in hydroxyl-propyl-beta cyclodextrin complex in a molar ratio 1:2 by kneading method and a phase solubility evaluation was prepared to observe the increasing of water solubility for active compound. Results showed a direct relation between the amount of cyclodextrin and the solubility of genistein. Ten C57BL/6J mice, female of eight weeks were inoculated with 0.5 ml B16F0 melanoma cell suspension containing  $0.5 \times 10^5$  viable cells prepared after standard protocols on tail vein using as solvent saline solution. Genistein in complex was ultrasonicated into suspension in 0.9% saline prior to injection. Mice were divided in two groups and one group was injected with genistein complex i.p. at daily doses of 15 mg/kg b. w. for five days from the first day of experiment. Animals were inspected daily for the development of tumors, which became visible after 17 days from inoculation, when they were sacrificed. Tissue samples

(skin) were fixed in 10% formalin solution and were embedded in paraffin and cut at 4 microns. Finally after deparaffinized the samples were stained with H&E (hematoxylin-eosin) and microscopically analyzed. In case of untreated group it can be observed a tumor proliferation at the level of dermal tissue and also neighboring muscle fibers. In case of treated group analyses showed ectaziate vessels and dermis with perivascular proliferative process consisting of round cells relatively uniform and hyperchrom nucleus.

The main conclusions are that the water solubility of genistein could be increased by cyclodextrin complexation and it is an active compound in melanoma on coetaneous level.

## 29. METHOD VALIDATION FOR SIMULTANEOUS MULTIANALYT DETECTION OF DRUG RESIDUES IN HONEY USING BIOCHIP ARRAY TECHNOLOGY

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A new method for simultaneous determination of drug residues in honey samples was validated according to the criteria set by Commission Decision 2002/657/EC. Three different immunoassay based multi-analytes arrays for drug residues testing have been evaluated: Antimicrobial Array I for simultaneous screening of twelve sulphonamides (sulphamethazine, sulphadiazine, sulphadimethoxine, sulphaquinoxaline, sulphathiazole, sulphisoxazole, sulphapyridine, sulphamerazine, sulphamethoxy-pyridazine, sulphachlorpyridazine, sulphamethizole, sulphadoxine), Antimicrobial Array II for quinolones, ceftiofur, thiamphenicol, streptomycin, tylosin and tetracyclines simultaneously, Antimicrobial Array III for four nitrofurantoin metabolites and chloramphenicol simultaneously. The analytes determination was carried out using Biochip Array Technology (Randox Laboratories Ltd, Crumlin, Northern Ireland), an immunoassay testing platform for the simultaneous multi-analyte assessment of a panel of tests. All the tests on the biochip are performed simultaneously in a single procedure using a single undivided sample. A supercooled charged coupled device camera and customized image-processing software simultaneously read the results for the entire panel of tests. The method shows good linearity, sensitivity, specificity, precision (repeatability and intra-laboratory reproducibility), recovery, ruggedness and decision limits (CC<sub>95</sub>) below 50% of MRL (maximum residue limits). The results of the validation process demonstrate that the method is suitable for application, as a quantitative screening method, in European Union statutory veterinary drug residue surveillance programs.

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Agilent Genomics Platform includes everything from sample preparation, qPCR and bioreagents, microarray processing and scanning, NextGen Sequencing sample preparation, laboratory automation to data analysis, exploring the different cornerstones of the living cell: **genomics** (aCGH Arrays, ChIP on Chip Arrays, Methylation arrays), **transcriptomics** (Gene Expression Arrays, miRNA Arrays, Splicing Arrays), **proteomics** (LC/MS) and **metabolomics** (LC/MS).

Scientists around the world use a wide range of Agilent instruments, software and reagents to study the mechanisms of cancer, for example. As a leading provider of microarrays, Agilent is the source of sophisticated tools for examining the genomic aspects of cancers and other illnesses. Agilent is also a main global provider of gas chromatography (GC), liquid chromatography (LC) and mass spectrometry (MS) technology used to analyze tissue samples for proteins or metabolites in an attempt to identify markers for cancer, and detect and measure carcinogens in the environment.

Agilent's broad portfolio includes software to collect and interpret data from cancer research experiments, reagents needed to run the experiments and automation options for labs performing large volumes of work. In recent years, researchers have begun to look at cancers from multiple scientific perspectives, an approach known as "**systems biology**." Agilent supports this concept by addressing multiple scientific needs.

The **Agilent 2100 Bioanalyzer** is a microfluidics-based platform for sizing, quantification and quality control of DNA, RNA, proteins and cells on a single platform. Results are delivered within 30-40 minutes in automated, high quality digital data.

Miniaturization of analytical instrumentation has many advantages over conventional techniques. These advantages include improved data precision and reproducibility, short analysis times, minimal sample consumption, improved automation and integration of complex workflows. Launched in 1999, the Agilent 2100 Bioanalyzer is the first commercially available instrument to use microfluidics technology for the analysis of

biological samples.

Today, the Bioanalyzer offers a broad range of pre-validated analysis kits combined with an easy-to-use benchtop system.

#### Applications:

- **RNA quality check with RIN** - the industry standard for RNA analysis offering total RNA, mRNA and smallRNA's data including RIN algorithm (RNA Integrity Number)
- **DNA size and quantify** - smart, high resolution separation and quantification of DNA. Highly sensitive DNA sample QC for next generation sequencing experiments.
- **SDS-PAGE replacement for protein analysis** - the fast reliable way to determine the quantity and purity of proteins from Coomassie down to silver stain sensitivity.
- **On-Chip Flow Cytometry** - the easy way to acquire dual-color, cell-based fluorescence data.

Determining the integrity of RNA starting materials is a critical step in gene expression analysis. Using intact RNA is a key element for successful microarray or RT-PCR analyses. The Agilent 2100 bioanalyzer and associated RNA 6000 Nano and Pico LabChip kits have become the standard in RNA quality assessment and quantitation (MIQE guidelines). Using electrophoretic separation on microfabricated chips, RNA samples are separated and subsequently detected via laser induced fluorescence detection. The bioanalyzer software generates an electropherogram and gel-like image and displays results such as sample concentration and the so-called ribosomal ratio. The electropherogram provides a detailed visual assessment of the quality of an RNA sample.

In order to standardize the process of RNA integrity interpretation, Agilent Technologies has introduced a tool for RNA quality assessment. The RNA Integrity Number (RIN), was developed to remove individual interpretation in RNA quality control. It takes the entire electrophoretic trace into account. The RIN software algorithm allows for the classification of eukaryotic total RNA, based on a numbering system from 1 to 10, with 1 being the most degraded profile and 10 being the most intact. In this way, interpretation of an electropherogram is facilitated, comparison of samples is enabled and repeatability of experiments is ensured.

*"The Bioanalyzer with its RIN standard is an excellent tool to assess the integrity of RNA starting material as requested by the MIQE checklist."*

**Mikael Kubista, TATAA Biocenter**

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