# 1. Katarina

## Abstract 1

Raman spectroscopy is a physical method with a broad spectrum of applications in multiple scientific fields. Our work here presents the possibility to differentiate between 2 important grampositive species commonly found in clinical material – Staphylococcus aureus and Staphylococcus epidermidis. We tested 87 strains, 41 of S. aureus and 46 of S. epidermidis directly from colonies grown on a Mueller-Hinton agar plate. The method showed a great potential to separate these two species.

## Abstract 2

Raman spectroscopy has a broad range of applications in numerous scientific fields, including microbiology. Our work here monitors the influence of culture media on the Raman spectra of clinically important microorganisms (Escherichia coli, Staphylococcus aureus, Staphylococcus epidermidis, Candida albicans). Choosing an adequate medium may enhance the reproducibility of the method as well as simplifying the data processing and evaluation. We tested four different media per each microorganism depending on the nutritional requirements and clinical usage directly on a Petri dish. Some of the media have a significant influence on the microbial fingerprint (Roosvelt-Park Institute Medium, CHROMagar) and should not be used for the acquisition of Raman spectra. It was found that the most suitable medium for microbiological experiments regarding these organisms was Mueller-Hinton agar.

# 2. Helena

Analysis of leukemic cells and their external supporting stimuli in relation to therapy response: my possible poster

Chronic lymphocytic leukemia (CLL) is the most common form of leukemia found in adults in Europe and the United States. It is characterized by highly variable course and outcome. This malignancy is treatable but still remains incurable. Multiple studies in last few years have shown that B-cell receptor (BCR) signaling plays an essential role in the biology of CLL cells. This poster shows preliminary results from an investigation of BCR signaling in three distinct groups of patients with different prognoses.

To determine the role of BCR signaling in CLL, we used samples from 30 CLL patient (10 patients with a favorable prognosis, 10 patients with an intermediate prognosis and 10 patients with poor prognosis). B-lymphocytes were labeled with antibodies focused on BCR signaling (p-Erk, p-Akt, p-p38, p-IKKγ, p-NF- $\kappa$ B, p-BTK/ITK, p-PLC- $\gamma$ 2 and p-ZAP70/SYK) and were measured using a flow cytometer. The results showed different activity of BCR signaling in groups of patients with various prognoses. The response of BCR signaling after stimulation was highest in patients with a poor clinical prognosis and lowest in patients with the most favorable prognosis. The differences were most obvious when we focused on kinases BTK, PLC- $\gamma$ 2 and ZAP70/SYK. There was no difference observed when we focused on kinase IKK $\gamma$ . In this study we demonstrated that the BCR signaling, which is now considered as essential pathway in CLL cells, has various activity in CLL patients. The activity of this signaling correlates with patients' clinical prognosis and could be possibly used as an additional prognostic marker in clinical praxis.

# 3. Lasha

Agricultural Policy and Agricultural Cooperatives Development Perspectives in Georgia

The present study shows the relationship between the agricultural policy and the agricultural cooperatives development in Georgia from 2013 to 2015. In the methodological part in order to assess the research subject I used both quantitative and qualitative methods. In quantitative , analysis the regression model was used. The results from the regression model suggest that agricultural policy implements have a significant effect on agricultural cooperatives in Georgia. In the qualitative analysis, a special questionnaire was created and administered before interviews were conducted with 100 representatives from Georgian agricultural cooperatives. According to the completed questionnaires, there are communication and knowledge transfer problems between the Agricultural Cooperation Development Agency and agricultural cooperatives in the country. In addition, results show that there are some significant differences and commonalities between the sectors. For example, cooperatives in the wine sector carry out a diversity of functions, however cooperatives in the hazelnut sector are mostly strong in processing. Therefore, this paper will be the step forward as in scientific also in the policy making perspectives which will help decision makers to address the problems faced in agricultural cooperation in Georgia.

# 4. Jana S.

Fruits and vegetables constitute an important source of energy and nutrients for humans. According to the World Health Organisation, intake of at least five servings of fruit and vegetables (approximately 400g per day) is recommended for the prevention of noncommunicable diseases and micronutrient deficiencies. The objective of this study was to specify the eating habits, especially fruit and vegetable consumption, in the Roma population. Also, it aimed to examine the attitudes towards fruit and vegetable consumption and knowledge about recommended daily amounts and nutrient content. The study was conducted across the Southern Moravian region of the Czech Republic, with a sample size of 102 individuals using the questionnaire method. Fruit and vegetable consumption among the participants was shown to be insufficient. Only 16% of individuals consumed fruit and vegetable at least once a day. The recommended daily intake of fruit and vegetables didn't know 80 % of individuals. According to 50% of individuals, fruit and vegetable was a good source of protein. The results of this study might help to promote more focused action education in the Roma population about healthy eating habits with the health impact of sufficient fruit and vegetable daily consumption.

Keywords: nutrition, fruit, vegetable, Roma population

## 5. Pastor

#### Abstract

This paper examines Brazil's fight against poverty from 2004 to 2013. The research's aim is to test whether Conditional Cash Transfer Programs (CCTP) have had some impact in elementary education. Through CCTP, especially the "Bolsa Familia", the government has tried to reduce poverty in two ways: short term and long term through monetary transfers and human capital investment, respectively. The strategy against poverty in Brazil has adopted a multidimensional approach, focusing on education, health and standard level of life. The methodology used in this paper was Pearson X2. with a significance level of 0,05, CL=95, and the statistical programme SPSS 22. We focused mainly on testing significant statistical relationships between the dependent variable "school dropout" and the independent variable "stratum", namely, if being a Bolsa Family's beneficiary is related to the dropout rate of elementary students. However, the article shows that in families with a \$R 50 income, there is a correlation between the dependent variable "dropout" and independent variables such as age, region, years of study and working child, but not with gender, race, or the mother lives at home and stratum. Regarding families with a \$R 100 income, we find only a correlation with age, years of study and region. According to BID, these programs are very effective in short-term poverty reduction. However, there is no proof that demonstrates the effectiveness of Conditional Cash Transfer Programs' against intergenerational transmission poverty.

## 6. Jana F.

Mutations in the ATM gene are associated with inferior prognosis in CLL patients. We noted a positive response of ATM mutated CLL samples to anti-CD20 antibodies rituximab and ofatumumab in our recent study [1]. Therefore, we analyzed the "in vivo destiny" of ATM mutations during the course of CLL. We selected 25 patients covering the whole spectrum of ATM defects involving mutations: missense and other types of mutations, with and without 11q-. In these patients, the presence of particular mutations was analyzed in subsequent time points during the disease course. Nineteen cases were monitored in a relapse (with a median follow-up of 42 months: therapy involved anti-CD20 antibodies in 15 cases), while there was no therapeutic intervention in the period between samplings in six patients (with a median follow-up 18 months). Among the new samples, we detected ATM mutations in 24/25 patients. In the remaining patients (treated with ofatumumab and chemotherapy) we observed a loss of a partially selected (original proportion about 15%) missense mutation. In the other two patients, we noted the disappearance of 1 out of 2 mutations, in one case after rituximab with chemotherapy and in one case spontaneously. We also confirmed the stability of ATM mutations in the other two patients analyzed retrospectively at diagnosis (median between samplings 55 months). The same mutations were again present at both the analyzed time points. Altogether, our results clearly documented that ATM mutations are stable during the course of CLL. Their elimination by therapy is rare, despite using most potent regimens involving monoclonal antibodies.

## 7. Barbora

In clinical practice, the KRAS mutant tumours of the colon are considered to be a homogeneous group of tumours. However, currently there is direct and indirect evidence of heterogeneity not only in response to treatment, but also in survival rates. Understanding the cellular processes responsible for these differences is important for the research of other methods of treatment. The study of gene expression in combination with the clinical parameters is used for the detection of new biomarkers and is one of the techniques used to detect specific biological processes for individual tumours. The main objective of this study was to characterize the gene expression heterogeneity of KRAS mutant colorectal cancer in an unsupervised manner and to connect these findings with known clinical, histopathological and mutational markers of colorectal cancer and prognosis. A unique data set from PETACC-3 clinical study was used for this purpose; this contains information of the gene expression of 313 patients with KRAS mutations in the colon accompanied by clinical variables, molecular markers and survival. Unsupervised analysis was used to unveil gene expression heterogeneity. By the means of clustering analysis, I defined 3 subtypes of patients with KRAS mutant colorectal tumours. However, a significant associations between these subtypes and clinical variables were not determined. Furthermore, there were no differences in prognosis. The heterogeneity of colon tumours with KRAS mutation was not clarified by their study on a molecular level. It seems that unsupervised analysis is not able to identify clinically and biologically meaningful subtypes.

#### 8. Ivo

Amphiphilic peptides can interact with biological membranes and severely affect their barrier and signaling functions. These peptides, including antimicrobial peptides, can self-assemble into transmembrane pores that cause cell death. Despite their medical importance, the conditions required for pore formation remain elusive. Monte Carlo simulations with coarse-grained models enabled us to calculate the free energies of pore opening under various conditions. In agreement with oriented circular dichroism experiments, a high peptide-to-lipid ratio was found to be necessary for spontaneous pore assembly. The peptide length has a non-monotonic impact on pore formation, and the optimal length matches with the membrane thickness. Furthermore, the hydrophobicity of the peptide ends and the mutual positions of peptides on the membrane play a role.

#### 9. Marketa

Interleukin 17 (IL-17) is known as a pro-inflammatory cytokine which participate in immune responses to infection by various pathogens in many animal species. However, there is still very little information about its role in the anti-infectious immunity of pigs.

In our study, six piglets were intranasally infected with Actinobacillus pleuropneumoniae. Samples from their lungs, tracheobronchial lymph nodes, BAL and blood were collected after one and three days, post infection (2 x 3 pigs). The IL-17 level was measured both in protein (western blot) and mRNA (quantitative real-time PCR) level and compared with samples from six control pigs. Flowcytometry was used to detect cell populations responsible for IL-17 production in the lungs of infected pigs.

The expression of IL-17 mRNA was found to have increased in the lungs of the infected pigs in both times post infection. The increase was detectable also in the protein levels. Both gammadelta TCR and CD4+ T-cells isolated from pig lungs was able to produce IL-17 after in vitro stimulation with PMA. However, no significant differences in the amount of IL-17 positive cells was detected in the in vitro model from control and infected pigs. Moreover, the percentage of CD4+ and gammadelta TCR+ cells in the lungs of control and infected pigs did not differ. Therefore, interleukin 17 seems to be the player of the immune response to Actinobacillus pleuropneumoniae infection although further experiments to elucidate the main source and key mechanism of responses are needed.

The work was supported by the Ministry of Agriculture of the Czech Republic (project QJ1210120).

## 10. Hana

EMT-related miRNAs as diagnostic and prognostic markers in renal cell carcinoma

**Background**: Renal cell carcinoma (RCC) is among the most common kidney cancer in the adult population. Because of the lack of early warning signals, a high percentage of patients with metastatic RCC occurs. Nowadays, epithelial-mesenchymal transition (EMT) is considered as a crucial event of the tumor progression resulting in metastasis. However, till to date, EMT, especially in renal cell carcinoma (RCC), remains enigmatic.

**Methods**: Immunofluorescence analysis of EMT status of formalin-fixed paraffin-embedded (FFPE) sections of 29 patients was performed using specific antibodies (E-cadherin, CK-18, CK-19, vimentin, S100A4). According to this analysis, patients were divided into two groups, EMT(+) and EMT(-). In order to obtain global EMT related miRNA expression profiles TaqMan Low Density Arrays (TLDA) were performed on the same group of patients. For the first validation phase, 27 miRNAs were selected. Expression miRNAs profiles were measured using TaqMan Real Time Expression Assay (Aplied Biosystem) and compared using Mann-Whitney U test and Kruskal-Wallis test. All data were normalized to RNU48.

Results: Using TLDA, 27 EMT related miRNAs (miR-200a, miR-200a\*, miR-200b, miR-200b\*, miR-200c, miR-429, miR-141, miR-192\*, miR-215, miR-30a-5p, miR-30a-3p, miR-30b, miR-30c, miR-30d\*, miR-30e, miR-30e-3p, miR-130\*, miR-630, miR-17\*, miR-193b, miR-26a-1\*, miR-571, miR-770-5p, miR-632) were chosen and tested as potential diagnostic and prognostic markers in RCC. Data will be presented.

Conclusions: Our primary data suggest that miRNA family, miR-200 and miR-30, may play an important role in RCC pathogenesis and metastatic cascade.

## 11. Ksenia

CHANGE IN SHORT-TERM BLOOD PRESSURE REGULATION IN PATIENTS WITH RESISTANT HYPERTENSION BEFORE AND AFTER RENAL DENERVATION: A PILOT STUDY.

## Introduction

One of the causes of hypertension is a high activity of the sympathetic nervous system, which is the reason of high secretion of the renin. This hormone has a lot of different functions which include influence of baroreflex sensitivity (BRS). That is why the main aim of this study is to detect some changes of BRS in patients with resistant hypertension before and after renal denervation.

## **Materials and Methods**

We measured 10 patients with resistant hypertension (HP) before and after renal denervation (RDN). For each patient, we measured a beat-to-beat continuous non-invasive 5-minute recording of blood pressure by photoplethysmography. Records were processed using spectral analysis and the medium frequency (MF), high frequency (HF) and very low frequency (VLF) region were calculated with the following parameters: normalized power spectrum of heart rate (nRRIMF, nRRIHF and nRRIVLF [n.u.]) and systolic and diastolic blood pressure (nSTKMF, nSTKHF, nSTKVLF, nDTKMF, nDTKHF and nDTKVLF [n.u.]), absolute power spectrum of heart rate (aRRIMF, aRRIHF and aRRIVLF [ms2/Hz]) and systolic and diastolic blood pressure (aSTKMF, aSTKHF, aSTKVLF, aDTKMF, aDTKHF and aDTKVLF [mmHg2/Hz]). The effectiveness of short-term regulation of blood pressure [ms / mmHg] was established in the medium frequency (BRS), HF (GainHF) and VLF (GainVLF) spectrum as the ratio of cross spectrum between RRI with STK (CrossMF, CrossHF or CrossVLF) and power spectrum STK.

## Result

A significant difference between the hypertonic before and after operation patients' results was found in BRSMF (4,84±2,57 vs. 6,09±2,96; p<0.05) and in nDTKVLF (0,11±0,09 vs. 0,07±0,03; p=0,07).

## Conclusion

In the MF range, we saw that BRS was improved with significant difference. That means that RDN has an influence not only on RAAS but also on the short-term regulation of blood pressure.

## 12. Matous

This article presents results of the research on NGOs educating Roma children in Brno city. The aim of the study was to a pedagogical strategies which are used in the process of non-institutional education of Roma children. The aim was to find out broad educational goals of NGOs. The aim was also to reveal the effect of funding systems on the work possibilities of NGOs. The research was based on nine semi-structured interviews with NGO representatives which is the total number of NGOs educating Roma children in Brno city. The results of the research revealed that NGOs are equally dependent on state and European Union funding, that they cannot be fully independent. The ideological background of their founders also has a strong influence to their pedagogical strategies and educational goals.

The research revealed that the broad educational goal tends not to focus on school achievement, as this is usually understood, but it is focused on the integration of Roma children into mainstream society. The research also described some of the pedagogical strategies used by NGOs. The research identified that the main problems with the education of Roma children by NGOs in Brno city the ethnocentrism of NGO employees and the misunderstandings concerning the educational goals of NGOs. This article thus reflects on the non-governmental educational efforts in Brno city.

# 13. Shahla

Accentual types of Modern English words.

This paper deals with accent types in Modern English. It has been acknowledged that second language learners and even some native speakers have difficulties in pronouncing words using correct stress patterns. So pronunciation words plays an important role in communication. This research has been investigated by different phoneticians like Torsuyev, Gimson. However, after analyzing some pronouncing dictionaries we think that the notion of accentual structure opposes the latest experiment. We tested it by phonetic –experiment. The material of the experiment consists of five hundred words which are classified according to different accentual types and recorded by two speakers with original British accent. The purpose of the experiment is to check weather British English speakers will show the accentual types of words in Modern English. According to the results of the phonetic-experiment, we defined accentual types of words in Modern English. It was found that the variability of the accentual structure of English words presents great difficulty for English learners. They should be well acquainted with the four most widely spread accentual types of words and be aware of the modification of the words accentual patterns influenced by rhythm and temps in connected speech. The practical value of this text is that the practical results can be used at the seminars on theoretical and practical phonetics.

## 14. Michaela

Chronic Lymphocytic Leukemia Cells with Mutation in NOTCH1 Gene Respond Poorly to Ofatumumab

Several recurrent mutations represent important prognostic and/or predictive factor in chronic lymphocytic leukemia (CLL). However, the response of respective patients to immunotherapy targeting CD20 is poorly understood. A recent report on the outcomes of CLL8 trial noted an association between the presence of NOTCH1 mutations in CLL patients and a lack of benefit from rituximab added to fludarabine and cyclophosphamide (Stilgenbauer et al. 2014). Similarly, another study showed a poor effect of anti-CD20 immunotherapy in NOTCH1-mutated CLL patients, specifically for rituximab-based induction and consolidation treatment (Bo et al. 2014). These two observations prompted this investigation/research to determine whether NOTCH1-mutated CLL cells exhibit higher primary resistance to an anti-CD20 antibody compared to samples without mutation. The hot-spot mutation c.7544 7545delCT in NOTCH1 gene was detected by Sanger seguencing of a part of exon 34. The CLL cells (samples from CLL patients monitored and/or treated at the University Hospital Brno) were cultivated in the presence of 15% active human serum and 20 µg/ml of ofatumumab. The viability in comparison with untreated control cells was assessed by a metabolic WST-1 assay. We analyzed the level of CD20 and CD55/CD59 using flow-cytometry detection. The impact of ofatumumab on cell viability was assessed in 45 CLL samples. The median viability after 24 hours of ofatumumab treatment in the genetic groups was the following: 70% in NOTCH1-mutated samples (n = 12), 35% in ATM-mutated samples (n = 12) and 48% in wt group (n = 21). Thus, the NOTCH1mutated samples were substantially more resistant to ofatumumab than ATM-mutated samples (P = 0,002) and wt samples (P= 0,024). In line with the response to ofatumumab, the highest CD20 level was observed in ATM-mutated samples (median 72 798 of ABC units), intermediate in wt-samples (62 633) and the lowest in NOTCH1-mutated samples (46 638) (ATM-mut vs. NOTCH1-mut P = 0,094). The distribution of the sum of CD55 and CD59 densities assessed on the same samples then emerged - also in line with theresponse to ofatumumab - in the opposite manner: median level of 38 914 of ABC units in ATM-mutated samples, 48 861 in wt group and 53 566 in NOTCH1-mutated samples (ATM-mut vs. NOTCH1-mut P = 0.095). More specifically, our results indicate that this poor response may be probably accounted also to a higher primary resistance of CLL cells to anti-CD20 MAbs. In addition, our study demonstrates the usefulness of recurrent mutation identification in studies evaluating the response of CLL patients to monoclonal antibodies. Supported by XX and YY projects.