

Missing numbers are oral presentations without abstract

002

Lessons from the history

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Evidence of planned operations date back to the creation of the first woman. It is God himself, who performed the first thoracotomy in general anaesthesia, by creating Eve out of Adam's rib. No postoperative infection has been recorded.

After thoracic surgery, next specialty which is evident already in the neolithic age, is Neurosurgery. The rounded edges of healed neolithic skulls indicate that at least some of these patients survived from these early attempts. They must have also recovered from the wound infection that almost inevitably followed the procedure. Open package was most probably the therapy used.

The code of Hammurabi is the oldest existing medical text. It dates back to the 3rd millennium B.C. The code imposed strict outcome controls on surgical procedures and set the surgeon's fee for opening an eye abscess. It was the birth of Ophthalmology.

Several kinds of operation were performed in Egypt, including circumcision. Wound edges were apposed with adhesive drapes and sutures. The wounds were irrigated and covered with a mixture of fat and honey.

Around 1200 B.C. in Homer's "Illiad" it is described how Machaon, a Greek surgeon and son of Asklepios, attended wounds that occurred by spears, swords and arrows. Wound care consisted of removing foreign bodies and bandaging the wound. Healing herbs were also used during Antiquity and in a Pompeian fresco the physician Japix removes the head of an arrow that had wounded Aeneas the founder of Rome, after having left Troya. The wound was not healing up to the moment that Aeneas's grandmother-Goddess Venus-brought the herb called dictamon, still used in Crete, to put it on the wound.

It is known today that copper salts, such as malachite and chrysocolla, have antibacterial properties. They were first used for eye-infections in Egypt. Therefore, it is no wonder that Achilles, medical student of Centauros Cheiron, cures the wound of King Telephos by dropping scrapings from his spear-point into the wound.

Throughout "Illiad" and "Odyssee" suppuration was not recorded as a problem, except for the chronic, festering wounds of Philoctetes bitten by a snake. The wound was stinking so much that anaerobes are today highly suspected as the causative organisms. Because of the wound smelling, Philoctetes had to be left out of the boat in the island of Lemnos, where still today Fango earth is used for medical purposes. We observe here medical baths that survived 3500 year.

Hippocrates lived some 800 years after Homer, during the "golden age of Greek antiquity". He laid the foundation of modern medicine and separated disease from religion. He was teaching that disease is a natural phenomenon and not God's punishment for sinners. Hippocrates recorded symptoms and signs and emphasised on the study of diseases by careful observation.

In the chapters of the Corpus Hippocraticum devoted to surgery, the famous maxim : ubi pus, ibi evacua is found. Here is also recommended that wound management includes washing of the wound with wine, bandaging it with fresh-washed and sun-dried clean clothes, over which more wine was poured.

The antiseptic qualities of wine were proven by Nichol and collaborators, who showed that the active ingredient in wine is not only the alcohol, as we believed, but the polyphenols, which not only kill the common wound organisms but also the ones found in drinking water, such as E. coli, Salmonella typhi murium and Vibrio cholerae. This might explain why Greeks used to drink water mixed with wine in special bowls to make it safer. 1 part of wine in 9 parts of water was sufficient if left of 4 h to kill the above mentioned bacteria.

In cases of Empyema, Hippocrates used to open the pleural cavity and clean it with wine. Thereafter, he left in the cavity clean clothes soaked in olive.

We have placed the emphasis in the abstract on Antiquity, however the lecture will high light most of the major steps combating Wound and Surgical Site Infections.

004

Antifungal Resistance Testing

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Primary and secondary resistance is an essential problem in the increasing number of systemic fungal infections as well as the unknown susceptibility of emerging pathogens. The Clinical and Laboratory Standards Institute (CLSI) established standardized methods for antifungal susceptibility testing of Candida spp. and Cryptococcus neoformans and molds including interpretative break-points for a selection of various antifungals. Also, the European Committee on Antimicrobial Susceptibility Testing (EUCAST) established a standard method. However, both standards are known to be time consuming, labour intensive and difficult to use, thus preventing their application as routine techniques. Alternative approaches are still needed. Agar diffusion methods and commercial microdilution methods have also been developed and are still being evaluated.

005

New Diagnostic Methods in Medical Mycology

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The prevalence of fungal infections has increased in recent years due to an increasing number of patients at risk. This is the result of advances in medicine contributing to the increased survival of immunocompromised patients. Additionally, the widespread use of antifungal prophylaxis has led to a shift in the epidemiology towards yeasts other than Candida albicans and molds other than Aspergillus fumigatus.

The diagnosis of fungal infections remains a significant problem. The clinical presentation is difficult to interpret, and noninvasive methods (X ray and CT scans) are neither sensitive nor specific. Culture results are available at the earliest in 2 to 3 days, and cultures are frequently negative due to slow or absent growth of fungi in clinical specimens. For molds in particular, the identification is laborious, time-consuming, and requires significant technological expertise. A rapid detection and identification to the species-level, however, is essential to start adequate treatment as early as possible.

In recent years, advances in detecting antigenemia and fungal DNA have been published, which will be reviewed here. Commercial assays for detection of fungal antigens, e.g. galactomannan, mannan (and antimannan), glucan, are available and have been evaluated in clinical settings. These assays in general allow early and sensitive diagnosis. However, the sensitivity and specificity vary considerably, depending on patient selection and clinical application of the test. Numerous studies have been published evaluating in-house PCR assays for diagnosis of invasive fungal infections, mostly supporting the feasibility of this approach. Molecular methods, e.g. sequence analysis, have also proved to be helpful in rapid and accurate identification of cultivated fungi.

006

Candida in Hemato-Oncology

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An increasing incidence of invasive fungal infections is observed in immunocompromised patients, especially in leukaemia patients. *Candida* species are the fourth leading cause of bloodstream infections and candidaemia is associated with substantial mortality. While *C. albicans* is common in the ICU, a shift towards non-*albicans* *Candida* species such as *C. glabrata* and *C. krusei* with decreased susceptibility or resistance to azoles has been observed in leukaemia and cancer patients. The decrease in the number of *Candida* infections and the shift towards non-*C. albicans* strains is presumably related to the use of fluconazole for prophylaxis but remains controversial. Particularly at risk for serious *Candida* infections are neutropenic patients and patients treated with corticosteroids or cytotoxic agents. In addition, risk factors in cancer patients are central venous catheters, treatment with broad-spectrum antibiotics, parenteral nutrition and prior fungal colonisation. Inadequate and delayed antifungal treatment for candidaemia is associated with increased mortality. Fluconazole is safe and effective in candidaemia caused by *C. albicans*. Within the last decade several new options for treatment of candidaemia have been studied, including newer azoles, echinocandins and amphotericin B including the lipid formulations. With these new drugs the safety and efficacy of antifungal therapy in candidaemia was improved substantially. A summary of the new treatment options in candidaemia in leukaemia and cancer patients will be presented.

007

Candida at Intensive Care

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Patients hospitalized in the ICU who have persistent fever or unexplained hypotension, despite broad-spectrum antibacterial agents, may have candidemia, invasive candidiasis, or aspergillosis or other fungal infections.

Candida species are the fourth most common cause of bloodstream infections. The Infectious Diseases Society of America (IDSA) guidelines recommend that empirical antifungal therapy should be limited to patients who have persistent fevers despite antibacterial therapy, who have *Candida* colonization in multiple sites, and who have other risk factors for candidemia. Patients in intensive care units account for the greatest number of episodes of candidemia in most hospitals. Surgical units, especially those caring for trauma and burn patients, and neonatal units have the highest rates of *Candida* infections. Besides the risks associated with young age and trauma or burns, other factors include: Central venous catheters, parenteral nutrition, Broad-spectrum antibiotics, High APACHE II scores, Renal failure requiring hemodialysis, Abdominal surgical procedures, Gastrointestinal tract perforations and anastomotic leaks, A cost-effectiveness decision model analyzed the effect of empiric versus culture-based treatment strategies using a variety of antifungal agents (standard or liposomal amphotericin B preparations, caspofungin, or fluconazole) compared to no treatment intervention. Life expectancy and cost of care were analyzed for each treatment strategy. The study results were significant for the following: The calculated prevalence of invasive candidiasis in the ICU was approximately 12 percent, in ICUs where the likelihood of invasive candidiasis was greater than 2.5 percent, empiric fluconazole therapy was the most effective strategy in reducing mortality at an acceptable cost. However, if the prevalence of fluconazole resistance was greater than 24 percent, empirical use of caspofungin was preferred, although at a much higher cost per life saved, in ICUs with a low prevalence of invasive candidiasis, culture-based fluconazole therapy was a preferred strategy.

Aspergillus species are ubiquitous, but disease due to tissue invasion with these fungi occurs primarily in immunocompromised hosts. Neutropenia and glucocorticoid use are the most common predisposing factors. Whenever possible, consideration should be given to decreasing the degree of immunosuppression as an adjunct to medical therapy for the treatment of invasive aspergillosis. Other therapeutic maneuvers, such as antifungal therapy, are only temporizing measures while awaiting the reversal of immune suppression. We recommend voriconazole as initial therapy in patients with invasive aspergillosis. The recommended dosing regimen is 6 mg/kg intravenously (IV) twice a day on day one followed by 4 mg/kg IV twice daily. When the patient has stabilized and is able to take oral medications, one can consider switching to 200 mg orally twice daily; the dose can be increased to 300 mg orally twice daily if the response is inadequate or serum voriconazole levels are <2 mcg/mL. (We suggest monitoring of serum voriconazole trough concentrations in all patients receiving voriconazole for invasive aspergillosis, particularly those receiving oral therapy. Although there are no guidelines for target voriconazole concentrations, we suggest checking a trough concentration one week into therapy with a goal range of >1 mg/L and <5.5 mg/L. In patients who are intolerant of voriconazole, we recommend use of a lipid-based formulation of amphotericin B (either Abelcet or AmBisome) at 5 mg/kg per day. This recommendation does not apply to infections due to *A. terreus*, which is intrinsically resistant to amphotericin B.

We suggest the use of combination regimens for salvage therapy in patients who do not respond to either voriconazole or liposomal amphotericin B. We suggest adding an echinocandin, such as caspofungin, micafungin, or anidulafungin, to voriconazole or to liposomal amphotericin B. There are no clinical data to support the use of amphotericin B with triazoles for combination therapy. (In patients at high risk of zygomycosis (mucormycosis), where the diagnosis of invasive aspergillosis has not definitively been made, we recommend liposomal amphotericin B rather than voriconazole, since voriconazole has no activity against zygomycetes. We suggest not using combination antifungal therapy as initial treatment. The duration of therapy is dependent upon the location of the infection, the patient's underlying disease, and the response to therapy. We suggest continuing antifungal therapy until all signs and symptoms as well as radiographic evidence of the infection have resolved for at least two weeks. For most immunosuppressed patients, antifungal therapy will continue for months. Patients who complete antifungal therapy are still at risk for reactivation of aspergillosis if neutropenia recurs. For these patients, we suggest reinitiation of antifungal therapy if chemotherapy is reinitiated and the patient is expected to become neutropenic or immunosuppression is intensified. Debridement is an essential step in the treatment of *Aspergillus* sinusitis. The use of debridement is sometimes helpful for extensive focal, necrotic cutaneous lesions and may be used on a case by case basis. Surgical excision of pulmonary lesions, especially persistent cavitary lesions, is recommended if the patient can tolerate a thoracotomy. Debridement also offers the possibility of making alternative diagnoses when specimens are properly prepared in the pathology laboratory and cultures plated in the microbiology laboratory.

The zygomycetes are a class of fungi that can cause a variety of infections in humans, particularly in immunocompromised patients and those with diabetes mellitus. Infection with zygomycetes can cause devastating rhino-orbital-cerebral and pulmonary infection with an extremely poor prognosis. Treatment of zygomycosis involves a combination of surgical debridement of involved tissues and antifungal therapy. Elimination of predisposing factors for infection such as hyperglycemia, metabolic acidosis, deferoxamine administration, and neutropenia is also critical. Intravenous amphotericin B (lipid formulation) is the drug of choice. Oral posaconazole is used as step down therapy for patients who have responded to amphotericin B and rarely as salvage therapy for patients who don't respond to or can not tolerate amphotericin B. Therapy should continue until all signs of infection have resolved, and often extends for months.

008

Aspergillosis and Other Moulds in Intensive Care Unit

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Introduction: Invasive fungal disease (IFD) has been increased over the last two decades. The population at risk includes persons with hematological or other malignancies and patients after transplantation but also patients with prolonged hospitalization in intensive care unit (ICU).

Objective: The aim of the study was to identify the patients with IFD treated in our ICU in 5,5 years period, to find the risk factors (RF) and to evaluate the efficacy of antifungal therapy.

Material and methods: Retrospectively we screened for IFD all patients admitted at 10 beds medical ICU in period from 1st of January 2003 to 30th of June 2008. Diagnosis was made according to the EORTC/MSG Consensus Revised definitions.

Results: Among 1860 patients admitted in to the ICU, 32 patients (16 female and 16 men) were diagnosed to have IFD. Mean age was 51.4. 84.38% of patients were transferred to ICU from hematology and hemato-oncology. The reasons for admission were respiratory insufficiency or haemodynamic instability. 71.88% of admitted patients needed mechanical ventilation. The most usual underlying diseases were acute myeloblastic leukemia (25%) and other hematological malignancies (18.75%). 50% of patients were classified to be at high risk for IA. Hematological malignancies (46.88%) and prolonged therapy with corticosteroids (46.88%) were the most usual RF. 34.38% of patients were neutropenic (<500/mm³). Positive cultures were available in 81.27% of cases (*Aspergillus* spp. 34.38%, *Aspergillus fumigatus* 21.88%, *Aspergillus flavus* and *terreus* 3.13%, *Candida albicans* 6.25%, *Candida glabrata* 9.38%, *Mucor* spp. 6.25%). In 12.50% of cases direct microscopy was positive and galactomannan test was positive in 50% of all patients (cutoff >0.5) (76.2% in IA). In all cases of IC blood cultures were positive. 90.63% of patients received previous broad-spectrum antibiotics. Voriconazole was the most usual empirically prescribed antifungal agent (in 37.50%). Combination therapy was used in 12.5% of cases. Overall mortality rate was 71.88% (IA 80.90%).

Conclusion: IFD was not always associated with neutropenia. Prolonged therapy with steroids was main RF in nonneutropenic patients. High mortality is partly due to late diagnosis and therapy. Insufficient diagnostic tools are the main reason for such delay.

009

Hospital Outbreaks of Invasive Mold Infections. Controlling the Risk of Air and Water Acquisition

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Healthcare-associated infections with moulds exhibit a serious threat for immunocompromised patients. Several factors can be considered as causes of the emergence of mould infection at hospital as environmental changes, construction work, contaminated air or water and an expanding population of immunocompromised hosts.

The nosocomial origin of mould infections has been demonstrated in epidemic situations with airborne spore transmission via unfiltered air, and massive environmental contamination during construction and renovation work inside and outside the hospital. Therefore air control measures are currently the more effective way of significantly reducing the incidence of nosocomial mould infections. Patient rooms should have adequate capacity to minimize accumulation of fungal spores by use of high-efficiency particulate air (HEPA) filtration, laminar air flow systems, high rates of room-air changes, positive pressure, and well-sealed rooms.

Also hospital water can be a source of moulds, in particular of *A. fumigatus*. The contamination of hospital water with *A. fumigatus* seems to be dependent on the kind of the natural water intake reservoir. Additional measures directed at improving water quality and limiting activities leading to aerolisation of water might be required.

In conclusion, suitable arrangements in the close surrounding of patients could help to protect the patient from exposure to fungal spores and to reduce the infection risks.

014

Pandemic Preparedness in the EU – Current Status and Challenges

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After many years of relative neglect, pandemic preparedness work in Europe started in early 2005. It was given a boost by the appearance of avian influenza (influenza A/H5N1) in birds in the European Union and human cases of H5N1 infection right on the EU borders in late 2005.

The European Centre for Disease Prevention and Control, which became active in May 2005, put this work high on its priority list and devised an assessment procedure which has evolved over time from being a classical three to four day assessment by an external team to one of self-assessment by a country. Since 2005, all 27 EU Member States and the 3 EEA/EFTA countries have been visited by international teams with experts from ECDC, the European Commission and WHO Regional Office for Europe.

Europe is probably the best prepared continent regarding pandemic influenza but still much needs to be done. Although Europe is quite different regarding size and structure of its Member States, the main problems are quite similar: Additional effort is needed to make plans more operational at local level, more involvement of other sectors than health is needed in an operative way at all levels and Member States should work more closely together to align their policies as to what they will do in a pandemic. Another difficulty is that still little is known about transmission of influenza and thus about effectiveness of control measures to limit spread. Therefore research should be encouraged to target these issues. Improving use of seasonal influenza vaccine and developing pandemic vaccine capacity is another high priority.

Nevertheless, the momentum should be maintained to continue to improve preparedness activities for an influenza pandemic, as all the experts are confident that it will happen, but nobody knows when and how severe it will be.

017

Tuberculosis and HIV in East Europe

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HIV is a growing challenge for the entire continent of Europe and the Commonwealth of Independent States (CIS). Over the past few years western CIS countries and the Baltic States have recorded some of the fastest growing rates of HIV infection in the world. The epidemic has reached critical levels in the Russian Federation and Ukraine, where infection rates have topped 1 percent of the adult population. Almost one third of affected individuals are in the age group of 15 to 24. The Russian Federation and Ukraine are the most severely affected nations carrying around 90 percent of the region's burden, but HIV is also spreading in Belarus, Kazakhstan, Romania and Tajikistan. The main driver of the epidemic in this region is Intravenous drug use but lately the sexual mode of transmission is also on the rise.

Tuberculosis (TB) increased dramatically as well in many of the newly independent states. Treatment services were not sustainable during the period of economic decline and access to care was reduced with the decentralization of services to oblast and rayon dispensaries. About 30 percent of adults diagnosed with HIV/AIDS and 50 percent of adults dying from AIDS had tuberculosis. Almost one third of all TB patients in Ukraine are in prisons. Some 7 percent of prisoners in Ukraine have active TB, and forty percent of deaths among them are due to the disease.

Drug-resistant TB, which is more difficult and costly to treat, is also spreading rapidly due to inadequate treatment and shortage of drugs. The development in recent years of a multi-drug-resistant strain in the CIS is growing into a major threat to the people of the region.

021

Switching of Double-boosted Protease Inhibitor (PI) Regimen to Single-boosted PI Regimen in HIV-infected Patients with Multi-NRTI and NNRTI Resistance

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Purpose: In resource-limited setting, double-boosted protease inhibitor (PI) regimen had been used in some HIV-infected patients with multi-NRTI and NNRTI resistance in the recent years when PIs were the only susceptible and available agents. Benefits of double-boosted PI over single-boosted PI have not been established particularly among PI-naïve patients. Adverse events from PIs are also more common in double-boosted PI regimen. Single-boosted PI may be an optimal regimen for these patients.

Methods: A prospective study was conducted among HIV-1 infected patients who had a history of multi-NRTI and NNRTI resistance and received double-boosted PI regimen. The double-boosted PI regimen was substituted with lopinavir/ritonavir plus tenofovir and lamivudine (LPV/r/TDF/3TC). CD4 cell count and HIV-1 RNA were monitored at baseline, 3 and 6 months after switching to LPV/r/TDF/3TC.

Results: We studied 20 patients (mean age, 38.5 years), of whom, 70% were male. Prior to the double-boosted PI regimen, all patients failed from the first NNRTI-based regimen and were naïve to PIs. Frequencies of drug resistance mutations were as follows: M184V (60%), TAMs (50%), K65R (30%), Q151M (30%) and NNRTI resistance mutations (100%). Double-boosted PI regimens used were: LPV/r + indinavir (50%) and LPV/r + saquinavir (50%). Median duration of double-boosted PI used was 34.5 months. Median baseline CD4 cell count at switching was 383 cells/mm³. HIV-1 RNA was <50 copies/mL in 60% of patients; the others had HIV-1 RNA at 84-2350 copies/mL. Percentage of patients with HIV-1 RNA <50 copies/mL at 6 and 12 months after switching to LPV/r/TDF/3TC were 80% and 100%, respectively. Mean change of CD4 were 18 and 42 cells/mm³ at the corresponding periods. All patients reported more convenience with LPV/r/TDF/3TC regimen and 60% reported better adherence.

Conclusions: Switching of double-boosted PI regimen to LPV/r/TDF/3TC in patients with prior multi-NRTI and NNRTI resistance is safe and shows favorable efficacy. Better adherence may contribute to the higher rate of undetectable HIV-1 RNA after switching. LPV/r/TDF/3TC may be an optimal regimen for patients with multi-NRTI and NNRTI resistance in resource-limited setting.

022

Anticancer and Antiviral Activities of Structural Isoform RtH2 of Native Hemocyanin, Derived from the Bulgarian Sea Mollusk *Rapana thomasiana*

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Native *R. thomasiana* hemocyanin was dissociated to two structural subunits RtH1 and RtH2 and purified by ion-exchange chromatography. Each of the two subunits contains eight functional units (FU) of 50kDa. FU RtH2 is the fifth unit from the amino-terminus of the RtH2 polypeptide chain and was tested *in vitro* for anti-tumor activity against three cell lines MCF-7 (human breast cancer cell line), A549 (human squamous alveolar carcinoma) and RD (human rhabdomyosarcoma), and for antiviral activity against influenza A virus (Flu A) and herpes simplex virus type 1 (HSV-1) and type 2 (HSV-2) by cytopathic effect (CPE) reduction assay. RtH2 inhibited cell proliferation and growth of all cancer cell lines to various extent. RD cells were the most sensitive to the subunit whereas MCF-7 were the least. The FU RtH2 exhibited antiviral activity against HSV-1 cultured in RD and MRC-5 cells and against Flu A – in MDCK SIAT1 cells, but did not have any activity against HSV-2. The Flu A virus induced CPE, infectious virus yields and the production of hemagglutinin were all reduced at non-toxic concentrations of the subunit RtH2 of native hemocyanin. The virus-inhibitory effect was selective, dose-related and strain-specific; selectivity indices ranged 9.5-68.3. The structural subunit RtH2 exhibited a strong HSV-1 inactivating activity. The inhibition affected adsorption as well as the intracellular stages of viral replication.

023

Viral Infection and Immunosuppression in Rheumatic Diseases

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The last years are characterized by increase of frequency of opportunistic bacterial and viral infections. Their appearance or reactivation is connected with immune system suppression particularly during steroid or immunosuppressive therapy. Development of viral infection in rheumatic diseases (RD) often cause severe course of the main disease with the appearance of serious complications such as hepatitis, nephritis, pneumonia and others.

Objective: To study components of immune system (interferon status (IFS), phagocytic score (PS), interleukin (IL) 2, 4) in patients with rheumatoid arthritis (RA) and systemic lupus erythematosus (SLE) complicated by viral infection.

Material and methods: 100 patients with RA and 67 with SLE who fulfilled appropriate ACR criteria and were followed up as inpatients or outpatients in the Institute of Rheumatology of RAMS were included. General clinical examination was done according to algorithms for RA and SLE. Specific serum antiviral IgM and IgG antibodies to cytomegalovirus (CMV), 1 and 2 types of herpes simplex virus (HSV), Epstein-Barr virus (EBV): VCA IgG (capsid antigen) and EA IgG (early antigen) were evaluated by immunoenzyme assay (IEA) and PCR, IFS and PS were examined, spontaneous and induced cytokine (TNF α 945; and IL4) secretion was assessed by ELISA.

Results: History of reactivation or exacerbation of chronic comorbid infection during 6 month period of steroid or immunosuppressive therapy was revealed in 126 patients (77 with RA and 49 with SLE). 52,6% of them had frequent ARD-like diseases, 34,8% - sore throat and 37,5% - bacterial infection (28 with RA, 14 with SLE). 51 (30,5%) patients (28 with RA, 23 with SLE) had monthly recurrences of opportunistic infection which hampered administration of disease modifying therapy. Serologic examination of these patients showed predominance of latent EBV and CMV infection. Combined production of antibodies to both viruses was revealed in 64 patients with RA and 29 with SLE. PCR was less informative. It allowed to reveal EBV in only 15,1% and CMV in 10,7% of patients with RD. Interferon deficit and inverse correlation of α 947; and particularly α 945; interferon production with IgG antiviral antibodies to CMV and EBV VCA (p=0,01, r=-0,82, p=0,02, r=-0,79 respectively) was present in 89%

of cases. 56,8% of RA and SLE patients had initially low chemiluminescence response of peripheral blood phagocytes. Sharp decrease of phagocytic response correlated with high titers of antibodies to Herpesviridae family and hepatitis B/C (p=0,01, r=0,80). Cytokine (IL4 and TNF α 945;) examination confirmed their hyperproduction in RD which was more prominent in SLE (p=0,02, r=0,68) and correlated with high titers of IgM/IgG antibodies to CMV and EBV IgG VCA and IgG EA (p=0,005, r=0,82). Mitogen activation enhanced synthesis of interferon α 947; TNF α 945; but not IL4. These serologic and immunologic features (interferon system hypofunction, TNF α 945; IL4 and antibodies to CMV and EBV hyperproduction) clinically manifested by peripheral vasculitis development and renal damage accompanied by moderate elevation of rheumatoid factor, anti-DNA and ANF as well as absence of effect of steroid and immunosuppressive therapy.

Conclusion: The results of the study allow discussing possibility of differentiated approach to antiviral therapy as well as expediency of steroids and immunosuppressive agents high doses administration in RD patients with latent viral infection.

025

How Breakpoints Arise – the Pharmacological Point of View

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Clinical trials of efficacy are primarily designed to satisfy requirements for the approval of new antibiotics. In contrast, trials which seek to optimize the dose, dosing interval and duration of treatment to improve antimicrobial effect while reducing development of resistance are rarely done. This might explain why antimicrobial therapy sometimes fails despite use of the chosen antibiotic according to its approved indication and “susceptible” pathogens.

Clinical breakpoints are supposed to indicate at which MIC the chance of effective bacterial killing prevails over failure, given the standard dosing schedule of a certain antimicrobial. Bacteria are categorized as susceptible if concentrations of the non-protein bound fraction of an antibiotic *in vivo* are above the MIC of a microorganism for a sufficient period of time in order to eradicate the pathogen. Thus, the breakpoint depends not exclusively on the antimicrobial activity of the antimicrobial but also on the pharmacokinetic properties of the drug. More recently three major pharmacokinetic (PK)/pharmacodynamic (PD) indices, the peak concentration (C_{max}) and the area under the concentration–time curve (AUC) over MIC as well as the time the concentration remains above the MIC of the micro-organism (T>MIC) have been increasingly used to determine breakpoints. Differences of PK/PD indices for Gram-positive and -negative bacteria and differences in pharmacodynamic action of so called concentration- or time-dependent antibiotics must be considered.

Various *in-vitro* models like killing curves and *in-vitro* kinetic models, but also mathematical models like Monte Carlo simulations are available for setting pharmacological breakpoints. Appropriate PK data are a prerequisite for every PK/PD model. More specifically, regulatory authorities recommend the use of PK data obtained from representative tissues for PK/PD considerations. Although PK/PD considerations should be used to set dosing regimes and preliminary breakpoints for clinical studies, these breakpoints should be validated in confirmatory clinical endpoint studies.



027
Is MRSA More Pathogenic than MSSA? Differences in the Frequency and Combinations of Virulence Genes between MRSA and MSSA Strains

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Purpose: The purpose of this study was to examine the presence and combinations of genes responsible for adhesin, haemolytic cytotoxin, classical enterotoxin, enterotoxin-like toxin genes and different accessory gene regulator (agr) types in *Staphylococcus aureus* strains isolated in Budapest, Vienna and Skopje and compare according to methicillin resistance.

Materials and Methods: Classical microbiological methods were used for the phenotypical identification of the strains. The genetic confirmation of the species of the strains was done by detecting the genes encoding thermostable endonuclease (nucA) and 23S rRNA. According to the presence of the *mecA* gene 48 Austrian methicillin-resistant *S. aureus* (MRSA) and 128 methicillin-sensitive *S. aureus* (MSSA), 110 Hungarian MRSA and 94 MSSA, 73 Macedonian MRSA and 29 MSSA strains were studied. The genes were detected by simplex or multiplex polymerase chain reaction.

Results: Altogether 231 MRSA and 251 MSSA strains of heterogeneity were examined. No considerable difference was obtained in the high frequency of protein-A (*spa*), collagen- (*cna*), fibronectin-A (*fnbA*) binding protein and intercellular adhesin-A (*icaA*) genes between MRSA and MSSA strains. The combination of *spa-fnbA-cna-ica* genes was 50% in MRSA and 30% in MSSA strains. Haemolysin gamma (*hlg*) and gamma variant (*hlgv*) genes were significantly more frequent in MRSA than in MSSA strains (86% v 74% and 100% v 72%, respectively). The combination of *hla-hlb-hlg-hlgv-hld* genes was 44% in MRSA and 33% in MSSA. Extremely high incidence of enterotoxin-A (86%) and enterotoxin-D (39%) genes was characteristic for the MRSA strains. The majority of enterotoxin-like toxin genes was present in the MRSA strains significantly more frequently than in MSSA ones except for *seh*, *sek*, *sel* and *seu* genes. The simultaneous presence of eight or more enterotoxin genes was 32% in MRSA while 8.4% in MSSA strains. Nearly half of the MRSA and MSSA strains harboured the *agr* type I gene while the prevalence of *agr* type II gene was significant for the MRSA isolates.

Conclusions: MRSA strains possess adhesin, cytotoxin and enterotoxin genes in a significantly higher frequency than MSSA ones and may have alterations in the regulation of virulence factor genes. Supported by ÖAD and T&T grant No.: A-19/02 and the Hungarian National Scientific Research Fund (OTKA) T 46186 and M 36764

030
Bacteremia with ESBLs-Producing Enterobacteriaceae an Emerging Challenge

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Bacteremia, sepsis and septic shock still present a considerable challenge to the contemporary medicine with mortality rate from 12% to about 50%. Microorganisms of Fam. Enterobacteriaceae are among the most common etiologic agents. Extended-spectrum beta-lactamase (ESBL)-producing Enterobacteriaceae continue to increase world-wide, incl. in blood-stream infections. Investigators urged to study the variety of newly emerging enzymes (more than 300 ESBLs: TEM-, SHV-, and CTX-M-type), and to characterize the new biologic events and their epidemiology. Severe patient condition/ underlying disease, accelerated age, previous antibiotic treatment (e.g. cephalosporins, fluoroquinolons) and previous hospitalization were identified as independent risk-factors. In recent years attempts have been made to better understand the significance of clinical and economic burden caused by ESBL-producing Enterobacteriaceae, and particularly in blood-stream infections. It was found that such infections are more difficult to treat, more costly, and associated with longer hospital stay. Most importantly, it was understood, that they lead to higher mortality, for example, in bacteremic spontaneous peritonitis with ESBL-producers!, a failure of 73% and 30-day mortality rate of 60% were registered vs. 17% and 23% respectively in the control group. Majority of studies has found that empiric therapy with cephalosporins or fluoroquinolons were associated with higher mortality rate, compared to the carbapenems, and to some beta-lactam/beta-lactamase inhibitor combinations. Nowadays the doctor is challenged by the extremely limited therapeutic options and the expansion of ESBL-producing *Escherichia coli*, *Klebsiella pneumoniae*, *Serratia marcescens*, *Enterobacter* spp, *Proteus* spp, etc, both in hospital- and ambulatory settings, the last one especially concerning CTX-M producing *E. coli*. In conclusion, as ESBL-producing Enterobacteriaceae represent a significant cause of blood-stream infection, there is a need of a strict infection control, and new antibiotic treatment attitude (restricted antibiotic policies and new clinical guidelines, e.g. for empiric therapy).

031
Antibiotic Resistant Escherichia Coli Strains are Inherited from Parents and Persist in the Infants' Intestines in the Absence of Selective Pressure

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Purpose: *Escherichia coli* (*E. coli*) is the most common facultative aerobic bacterium within the intestinal microbiota of humans. Some strains, termed resident, may persist for many months in the microbiota of the individual, while so-called transient strains have low colonizing capacities and disappear within some weeks. The emergence of resistance among commensal bacteria is a serious side effect of antibiotic usage in human and veterinary medicine because commensals may - at a later stage - cause extraintestinal infections such as urinary tract infections.

Thus, the aim of our study was to assess whether healthy breast-fed neonates and infants harbour resistant *E. coli* in their feces and whether these are transmitted from the parents to their child.

Materials and Methods: Fecal samples of 21 neonates were collected within the first 48 hours after delivery (first sample), at week 1, 2, 4 and 8, and 6 months. In addition, one stool sample was gained from their parents. All stool samples were plated on MacConkey agar plates, supplemented with defined concentrations of tetracycline, ampicillin, trimethoprim and cefazolin. Molecular comparison of the resistant *E. coli* strains was done by pulsed-field gel-electrophoresis (PFGE). Exclusion criteria for infants were poor fetal adaptation, any signs of infection, postnatally administered antibiotics or admission to a neonatal intensive care unit. Exclusion criteria for mothers were preterm rupture of membranes, signs of maternal infection, preeclampsia and antimicrobial treatment during pregnancy.

Results: Among the 21 infants 66.7% (n = 14) did not yield any resistant *E. coli* in their feces in the first 8 weeks. Three infants had resistant *E. coli* identical to those of their mothers' from the first week of life on (in one case even in the very first sample), which persisted over at least 8 weeks. In another four infants resistant *E. coli* were found from week 2 on, which persisted over several weeks. In two of these four infants the isolated *E. coli* was identical to those of their mothers'.

Conclusion: In infants resistant *E. coli* strains, probably inherited from their mothers, tend to persist over months. Therefore, the persistence of resistant *E. coli* and their possible capacity to cause asymptomatic infection or transfer its resistance genes to other bacteria deserves more attention.

033
SIRS or SEPSIS in Surgical Patients

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Despite considerable advances in the intensive clinical care, Sepsis and Septic Shock still remains the most common cause of death in non coronary Intensive Care Units. The relative lack of effective pharmacological interventions highlights the complex patho-physiologic events involved in sepsis.

To develop a more uniform set of definitions a consensus conference decided that **several infections or non infections diseases** and similar disorders be called **Systemic Inflammatory Response Syndrome (SIRS)**.

SIRS frequently leads to the development of acute lung injury, renal failure, hepatic insufficiency, haematological disorders and other like Multiple Organ Dysfunction Syndrome (MODS) and Multiple Organ Failure (MOF).

Although not completely understood, septic shock is believed to result from a widespread endothelial damage caused by persistent inflammation:

Inflammation is a natural defence mechanism, however when the release of inflammatory mediators exceed a certain level the symptoms of SIRS become apparent.

The consensus conference made another "Key" decision : to restrict the use of the word **Sepsis** in cases in which infection is documented, because SIRS can be caused not only by bacterial, viral, fungal or other infections but also by surgical trauma, injuries, haematomas, haemorrhagic shock, transfusions, medicaments and several others.

However by this consensus more disadvantages than benefit were caused, specially to the surgeons. The reason is that all classical signs of infection have been given as a present to inflammation and to the new entity (SIRS), and the potentially curable entity infection was stripped of its signs.

Conditions with increased cytokine production, like trauma or burn injury, auto-immunity illnesses like, rheumatoid arthritis, systemic Lupus erythematosus, vasculitis, drugs as Amphotericin B and Antibiotics, cryptogenic inflammation diseases, like Sarcoidosis, Kawasaki Syndrome, inflammatory bowel disease, cardiac diseases, like myocardial infarction, or congestive heart failure and many solid or haematologic malignancies have taken away all the symptoms and signs of infection. They have deprived sepsis from its clear entity. Sepsis became a bacteriological diagnosis with all disadvantages of a delayed therapy.

This lecture will show on the example of Extra Corporeal Circulation the impossibility of differentiating SIRS from Sepsis and the practical ways that we have to follow to prevent our patients from infection, without covering all patients with antibiotics for a longer period than necessary.

035

Endemic Typhus Investigation of the Epidemiological Chain in Crete Island (150 Cases 1993-2004)

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SUMMARY

- Proof of disease presence and endemic character in Chania Prefecture.
- Description of clinical and laboratory features of disease.
- Study and comparison of treatment options.
- Isolation and Culture of Rickettsia typhi.
- Re-certified classical epidemiological cycle
- We have completed the study of epidemiological chain of endemic typhus in Chania Prefecture since R.typhi was isolated in rats, fleas and humans.
- All 150 patients that were diagnosed and treated had a good outcome.
- Clinical suspicion that leads to early and targeted antibiotic treatment is of great importance for avoidance and effective management of complications.

Our study is the first clinical study that involves newer quinolones (ciprofloxacin) in the treatment of endemic typhus. In our study we concluded that patients treated with ciprofloxacin responded and were afebrile in 4,23 days without recurrence of disease. Alternative treatment with quinolones needs to be studied with further clinical trials.

The current study has emerged and proved the presence and endemic character of disease in Chania Prefecture; while registering clinical and laboratory features of disease our study offered to medical society of Chania the option of considering rickettsial infection when treating a patient with fever, headache and rash.

037

National Immunization Programs with the 7-valent Pneumococcal Conjugate Vaccine in Europe

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Streptococcus pneumoniae is a major source of morbidity and mortality worldwide. It is estimated by the World Health Organization that at least one million children die of pneumococcal disease every year, mostly in developing countries. Pneumococcal infections are the leading cause of death from a vaccine preventable illness in children under 5 years of age. Invasive diseases caused by pneumococci include meningitis, bacteraemia, and pneumonia with bacteraemia and/or empyema. Risk factors for invasive pneumococcal disease (IPD) include age, with incidence being highest in young children (<2 years) and the elderly, ethnicity, geographic location, concomitant chronic illnesses, and attendance in day care centers. The overall burden of invasive pneumococcal disease (IPD) varies worldwide and reported incidences of IPD in Europe are about 35 cases per 100,000 children in the age group <2 years. Mean incidence of pneumococcal meningitis is 9.2 cases/100,000 and 5.1 cases/100,000 for children aged <2 years and <5 years, respectively. The mean case fatality rate of IPD in Europe is 3.4% and relatively higher in pneumococcal meningitis with 7–8%. Mean serotype coverage of the currently licensed heptavalent pneumococcal conjugate vaccine (PCV7) for children aged <2 years is about 70% with regional differences based on surveillance data reported before PCV7 was introduced into National Immunization Programs (NIPs) in Europe. The most common serotypes/groups causing IPD in Europe are 14, 6B, 19 and 23. Serotypes 1 and 5 are seen more frequently in older children. Although PCV7 was officially licensed in Europe in 2001, it was only used in some countries and under specific conditions, such as in high risk patients, at that time. Since 2003, PCV7 has been increasingly included in European NIPs and is now included in more than 10 European countries. First studies on the impact of PCV7 NIPs on IPD in children <2 years are available from France, UK, Germany and Norway demonstrating significant declines in bacteraemia and meningitis. Some studies in the post-PCV7 era, particularly from France and Spain, demonstrate a change from predominately vaccine serotypes to an increase in non-vaccine serotypes, e.g. 19A, 1, 3, 6A, and 7E, thus highlighting the need for inclusion of these serotypes into future vaccine formulations. Based on currently available effectiveness data, there is no 19F/19A cross-protection, and cross protection is only seen in part for serotypes 6A/6B. Therefore, serotypes 19A or 6A need to be included in future vaccine formulations.

039

Quantitation of Human Antibodies to 23 F Pneumococcal Polysaccharide by Enzyme-Linked Immunosorbent Assay in Romania

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Purpose: To study for the first time in Romania the pneumococcal vaccine responses in elderly adults.

Material and Methods: 49 outpatients, older than 65 years, were immunized with 23-valent pneumococcal vaccine (Pneumo-23) and then investigated at 30, 90, 360 and 390 days for antipneumococcal IgG response to 23 F vaccine serotype (the most frequently encountered in invasive pneumococcal illnesses in Romania) according to WHO- 3rd generation ELISA protocol.

Results: Amongst 49 subjects investigated, 32 presented at 360 and 390 days postimmunization, antipneumococcal IgG titers of at least two fold greater than IgG investigated after 30 days. Seventeen subjects didn't show immune protection after vaccination. Seven of them, didn't have increased titers of 2 fold after 30 days and 90 days postvaccination and three of subjects, even though revealed an increase of IgG titers of at least 2 fold at 30 days and 90 days after vaccination, at 360 and 390 days presented decreased titers of IgG.

Conclusions: This study revealed that in general Pneumo-23 vaccination caused two- to fourfold rise in antibody levels in Romanian elderly adults. A subset of tested subjects was identified as poor responders to 23 F serotype, probably because of their immunological status. So, more investigations are needed to evaluate immune status in the elderly and immunocompromised patients after vaccination.

040

Immunoblot Analysis of the Immune Response to Various Antigens of *Borrelia burgdorferi* during the Post-Treatment Course of Erythema Migrans Disease

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Purpose: To examine the value of immunoblot (IB) analyses of the immune response to different proteins of *Borrelia burgdorferi* sensu lato (Bb) in the post-treatment assessment of patients with erythema migrans (EM), the hallmark of early Lyme disease.

Methods: In 50 adult EM patients, anti-Bb IgG and IgM antibodies were analyzed in a median of 5 serum samples consecutively obtained before and during a median of 513 days (range, 414-1185d) after antibiotic therapy by recombinant IB. The antigens used in the IgG IB were decorin-binding protein A, internal fragment of p41 (p41i), outer surface protein (Osp) C, variable major protein-like sequence expressed (VlsE), p39, p58, p100 and in the IgM IB p41i, OspC, p39, p100. Immune responses were correlated with clinical and treatment-related parameters.

Results: Positive IB results were found in 50% before, in 57% directly after therapy and in 44% by the end of the follow-up for the IgG class, and in 36, 43 and 12% for the IgM class. In acute and convalescence phase sera, VlsE was most immunogenic for the IgG class (60 and 70%), and p41i (46 and 57%) and OspC (40 and 57%) for the IgM class. By the end of the follow-up, only the anti-p41i IgM response was significantly decreased to 24% (p=.36). No correlation was found between IB results and type or duration of therapy or clinical outcome.

Conclusion: During long-term follow up, only the IgM immune response to p41i decreased significantly. No correlations could be found between antibody kinetics and therapy or disease outcome. Thus, IB does not add to the clinical assessment of EM patients after treatment.

P01

Enteropathogenic Bacteria and Viruses in Acute Gastroenteritis During the Year 2007

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Aim: To evaluate the prevalence of enteropathogenic bacteria and viruses in children and adults' stool samples with acute gastroenteritis.

CultureMedia/Methods: 1160 samples were examined. For the isolation of bacteria, special culture media were used. We used API-20E and Vitek2 (BIO-MERIEUX) for the identification, Kirby-Bauer method and Vitek2 for susceptibility and MIC evaluation. Rotavirus and adenovirus was detected by ImmuneChromatographyTechnique (VIKIA Rota-Adeno, BioMerieux).

Total of enteropathogenic species		
135		
Campylobacter spp	70	51.8%
Salmonella spp	53	39.2%
Shigella spp	8	5.92%
Aeromonas hydrophila	2	1.48%
Yersinia enterocolytica	2	1.48%
Results:		
Samples of children <3y old		
507		
Rotavirus-Ag	38	7.5%
Adenovirus-Ag	17	3.4%

- Toxin A of Clostridium difficile was detected in 15 samples.
- All Salmonella spp were sensitive to Quinolones, 3rd generation cephalosporins, Ampicillin, Trimethoprim-Sulfamethoxazole.

Conclusions: Viral gastroenteritis in children is mainly due to Rotavirus. Among Salmonella spp the most frequent is Salmonella enteritidis followed by Salmonella typhimurium.

Among Shigella spp, Shigella flexnerii is the most common. Campylobacter spp is detected in a large percentage.

P02

Direct Detection of Streptococcus Pneumoniae Antigen in Urine During the Year 2007

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Aim: The evaluation of direct detection of Streptococcus pneumoniae antigen (SPA_g) in urine samples for the diagnosis of pneumococcal pneumonia.

CultureMedia/Methods: In 2007, 392 urine samples of patients with symptoms of lower respiratory tract infection were sent to the laboratory. Streptococcus pneumoniae antigen in urine was detected by Immune Chromatography (Now Streptococcus pneumoniae Antigen test kit, BINAX).

Results: Streptococcus pneumoniae Ag was found positive in 38 patients (9.7%). Seasonal distribution revealed increased prevalence during winter and spring. Sputum cultures of 6 patients with positive SPA_g were sent to the laboratory. Pneumococcus was not isolated in any sputum culture. Streptococcus pneumoniae was isolated from the nasopharynx of 6 patients.

Conclusion: The direct detection of streptococcus pneumoniae antigen in urine of patients with symptoms of lower respiratory tract infection contributes significantly to the rapid diagnosis of pneumococcal pneumonia despite the fact that sputum culture did not provide positive results, in most cases.

P03

Detection of Pseudomonas Aeruginosa Producing Extended Spectrum Beta Lactamases (ESBLs) and their Prevalence in Clinical Samples by Susceptibility Testing and PCR

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Introduction: Recent studies have been revealed the Pseudomonas aeruginosa isolated from clinical samples like Enterobacteriaceae may include the various types of ESBLs with distinct epidemiological frequencies. But their prevalence is not equal to Enterobacteriaceae, however, some genes encoding specific ESBLs are typical for Pseudomonas aeruginosa such OXA, VEB, PER.

Objectives: Determination of Pseudomonas aeruginosa producing ESBLs prevalence isolated from clinical samples and their gene grouping.

Method: 200 pseudomonas aeruginosa have been isolated from clinical laboratories of Karaj hospitals. Identification tests and susceptibility testing such disc diffusion and MIC have been performed for them. The used antibiotics were Cefotaxim, Ceftriaxon, Cefepime, Cefepidoxim, Aztreonam, and Clavulanate added to Cefotaxim and Cefepidoxim. The bacteria with ESBL have been preserved in -70°C freezer. PCR has been performed on them for ESBL gene groups consist: TEM, SHV, OXA, CTX, VEB and PER.

Results: 96 (48%) were positive for ESBL. 77 (38%) of bacteria were sensitive and 27 (14%) were intermediate susceptible. Gene groups for ESBL by PCR were: TEM (37), OXA (36), VEB (85), CTX (21) and PER (0).

Conclusion: It could be seen from results that spreading of ESBL producing strains of Pseudomonas aeruginosa in community and clinical environments have increased and it could be a threat for treatment.

P04

Prevalence of Vancomycin Resistant Enterococci in Patients who Referred to Mofid Pediatric Hospital

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Introduction: Vancomycin is one of the most effective antibiotics against the Gram positive cocci. Prevalence of resistance between Enterococci and its simple spread way to other Gram positive cocci such as Staphylococci and Streptococci have lead to serious problems for in-bed patients. It could be suggested that frequency of resistant Enterococci indicate a reliable pattern of antibiotic susceptibility. Result in saving time and cost.

Objectives:

1. Prevalence of Vancomycin resistant Enterococci in Mofeed children hospital and Ali Asghar hospital
2. Antibiotic resistance patterns in Enterococci
3. How is the Vancomycin resistance between Enterococci isolated from patients' stool samples
4. Pattern of antibiotic resistance of Enterococci against other antibiotics
5. Determination of Vancomycin resistance and its relation to effective antibiotics for anaerobic bacteria
6. Prevalence of Vancomycin resistance in Enterococci isolated from stool samples from distinct wards in hospitals.

Method: Stool Sampling was performed on in-bed patients in distinct wards in hospital including GI and infectious of Mofeed hospital and chemotherapy in Aliasghar hospital. Stool culture was done once introduce to laboratory 48 hours after patient's admittance. Specimens were cultured on media such as Enterococcal agar and Bile Esculin agar. And then some specific tests were performed for their confirmation such as PYR test, growth in 15 C and 45 C in Mueller Hinton agar. Growth in 6.5% NaCl. E-test was performed on VRE strains. Selected antibiotics fro E-test were included Cephoretan, Rifampicin, Gentamycin, Ciprofloxacin, Ceftriaxon, Oxacilin, Cefotaxim, Amikacin, Clindamycin, Chloramphenicol, Imipenem. Susceptibility level was reported on the basis of NCCLS charts.

Results: In this research, we examine stool samples of 64 patients who had one or two samples. 13 percent of Enterococci were resistant. Antibiotic pattern was cefotetan 60%, Rifampicin 36%, Gentamycin 36%, ciprofloxacin 32%, tetracycline 53%, Ceftriaxon 86%, oxacilline 65%, cefotaxime 76%, Amikacin 48%, Clindamycin 23%, Chloramphenicol 24%, imipene 25%.

Conclusion: Antibiotic resistance in Enterococci is increasing even vancomycin which is the first choice for Enterococcal infections treatment, it could be suggested before effective treatment, disc diffusion test and E-test should be done.

P05

Interstitial Atypical Pneumonia

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Introduction: Pneumonia by mycoplasma usually affects young people age 1-30 years old. Elderly are not excluded. After an incubation period of 2-3 weeks the disease invades with sore throat, coughing and fever. Radiologically there is focal invasion, bilaterally or unilaterally, to one or more parts of the lower lobes of the lung.

Purpose: Patient with interstitial pneumonia, severe anemia, hypoxigenemia, and positive serum antibodies against mycoplasma pneumoniae is presented.

Material/Methods: 42 years old man, alcoholic, heavy smoker with known megaloblastic anemia came to our hospital suffering from coughing, fever, weakness and loss of power during the last 15 days. He was hypoxigenemic (sat: 90%, pO₂: 56 mm Hg) with pale skin. Also while palpating the belly hepatosplenomegaly was found. Blood test counts: Hct: 13%, Hgb: 4.5g/dl, Reticulocytes: 0.50%, MCV: 101fl, WBC: 5.210 (PMN: 60%) E.S.R.: 34 mm 1st hour. Ro lung: deeper outline of the interstitium of the lung bilaterally and mainly to the left laterally to the heart. The lesion was found also in CT of the lung. U/S: hepatosplenomegaly (spleen: 15 x 6.5 cm). Mayer test, direct coombs, electrophoresis of the albumins, control for collagenosis, hepatitis, HIV, Ca markers, ferritin, B12, all were normal. Serological test for antibodies against chlamydia, rickettsiae, coxackie virus, mycoplasma pn. were sent. In the same time he was submitted to gastroscopy where showed oesophageal candidiasis and hiatal hernia. Antibodies against candida albicans were sent.

Results: The antibodies against mycoplasma pn. were positive IgG:1/160, IgM:1/40 (normal values 1:20 and 1:20) all others were negative, therefore we concluded our diagnosis. He was treated with Avelox 1flx1, Primaxin 1flx4, targocid 1flx1 i.v. for 15 days and itraconazole p.o., plus pantoprazole for 2 months. The improvement of his health was satisfactory.

Conclusion: Pneumonia by mycoplasma pneumoniae is usually expressed with mild or intermediate severity of clinical manifestations (only 2-10% of the infected demand hospitalisation). In our case report the manifestation was very severe so it was necessary for the patient to be hospitalised for long time. The immunosuppression due to alcoholism could be the main reason.

P06

The Prevalence of Intestinal Parasitic Infections among Mentally Disabled Children and Adult of Urmia in 2007

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Introduction: Intestinal parasites including protozoa and nematodes can cause different clinical and symptoms in infected patients. Institutions where men live in groups with low of are a suitable environment for preservation and transmission of these infections. In this study, prevalence of intestinal parasites infection in mental retarded institutions of Urmia city investigated.

Materials and methods: This descriptive-cross sectional study was carried out in mental retarded institutions of Urmia city. 225 fecal samples of less than 29 years old mental retarded individuals were examined by using direct smear, formalin ether concentration and scotch tape.

Results: Of 225 mentally retarded persons, 118 (52/4%) and 107 (47/6%) were women and men. The overall prevalence of infection was 20/5% and that of male, and female were 9/7% and 10/6%. 39 (17/3%) of infected individuals had protozoa infection and 7 (3/1%) helminth including *Enterobius vermicularis*. The infection rate of detected intestinal parasite was: *Entamoeba coli* (33/3%), *Giardia lamblia* (21/5%), *Iodoamoeba butschlii* (20%), *Blastocystis hominis* (12/3%), *Enterobius vermicularis* (10/8%), *Entamoeba histolytica* (1/6%). Of 46 infected persons, 2 (4%) in persons aged 1-5; 11 (46%) in persons 5-14; 6 (13%) in persons aged 14-18; 17 (37%) in persons aged >18 years. Of 44 infected persons, 15 (34%) IQ <25, 15 (34%) IQ 50-75, 12 (27/2%) IQ 25-50, 2 (4/5%) IQ 75-90.

Conclusion: According to these findings, attention to suitable procedures in order to control and treatment in these institutions is necessary.

P07

Unusual Pathogens inducing Bacteremia during the Years 2006-2007 in the General Hospital of Chania

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We report the isolation of unusual bacterial pathogens in cases of bacteremia. For the incubation of the blood cultures the Bact Alert system was used and for the standardization and the MIC calculation the Vitek 2 system (Biomerieux).

The most interesting cases were the following:

- *Aeromonas hydrophila* in an 80 year-old patient. Outcome: death. It was sensitive to aminoglycosides, TSX, IMP, MER and quinolones.
- Abiotrophic defective in a 67 year-old patient who was receiving corticosteroids. Outcome: death. It was sensitive to P, VAN and E.
- Group A beta-hemolytic streptococcus in a pregnant woman. Outcome: cure. It was sensitive to P and it was resistance to E and CC.
- Group B beta-hemolytic streptococcus in a neonate. Outcome: cure. It was sensitive to P and AMP.
- Group B beta-hemolytic streptococcus in an adult. Outcome: cure. It was sensitive to P and AMP.
- *Achromobacter xylosoxidans* in a patient of the Intensive Care Unit. Outcome: death. It was sensitive IMP, MER, CIP, MIN, C and CAZ.
- *Sphingomonas paucimobilis* in an oncological patient. Outcome: death. It was sensitive IMP, AKN, TSX and MIN.
- *Bordetella bronchiseptica* in an adult patient. Outcome: cure. It was sensitive RA, TSX, MIN and TE.
- *Chryseobacterium meningosepticum* in a adult patient. Outcome: cure. It was sensitive RA, TSX and CIP.
- *Saccharomyces cerevisiae* in a patient of the neurosurgical department. Outcome: cure.

(TSX: Trimethoprim/Sulfamethoxazole, IMP: Imipenem, MER: Meropenem, P: Penicillin, VAN: Vancomycin, E: Erythromycin, CC: Clindamycin, AMP: Ampicillin, CIP: Ciprofloxacin, MIN: Minocyclin, C: Chloramphenicol, CAZ: Cefazidime, AKN: Amikacin, RA: Rifampin, TE: Tetracycline)

Provided that, during the last years, new pathogens are described as a cause of bacteremia the laboratories should be alerted and a close collaboration with the clinicians should be established.

P08

Treatment of Diabetic Foot Complications

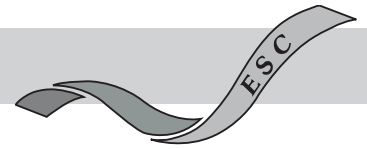
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Purpose: To analyze retrospectively the infectious complications of the lower extremity in patients with diabetes mellitus, to identify the most frequent etiologic agents and to determine the surgical strategy for management of the problem.

Materials and Methods: Hospital records of patients with diabetic foot infection treated in the Surgery Clinic of Medical Institute – Ministry of the Interior during 2002–2008 we studied. Wound swabs or exudate and tissue biopsy were submitted for Microbiologic investigation. Culture was followed by microbial identification by routine tests, as well as with API, Biomérieux (France) and Crystal, BD, USA biochemical panels. Antimicrobial susceptibility was determined by disk diffusion method according to CLSI, USA. Four different groups of Surgical procedures were performed: A. necrectomy, incision and drainage; B. amputation of one or more fingers or part of the foot; C. below-knee amputation; D. amputation of the hip.

Results: One hundred and twenty two patients (mean age 64 year; 95 male and 27 female) with diabetic foot infection have been treated during the 6-year period. Seventy six patients (62%) have been operated. Number of patients that have undergone surgical interventions of group A, B, C, D was respectively: 24, 37, 5 and 10. The crude mortality was 2.5% (3 patients). Microbiology results showed different mixed flora (2 to 5 isolates). The most frequent etiological agents were combination of *Staphylococcus aureus*, *Enterococcus faecalis* or beta-hemolytic *Streptococcus* gr. B with Gram-negative microorganism: *Proteus mirabilis*, *Pseudomonas aeruginosa* and others. Majority of microorganisms was susceptible to antibiotics: only one MRSA and one *Enterobacter cloacae* with ESBL have been identified, however the recovery of infection was delayed and re-infection was frequent.

Conclusion: The optimal strategy in diabetic foot infection should have a multidisciplinary character. Early recognition of problems, periodic and adequate surgical care of wound, control of hyperglycemia and infectious agents represent the key in the management of disease.



P09

Methicillin-Resistant Staphylococcus Aureus (MRSA) and MLSB (Macrolide-Lincosamide-Stretogramin B) Isolated from Community and Hospital – Associated Infections (Ca/Ha-MRSA)

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Background: Hospital - associated infections are representing a major worldwide health public inquire. They are the result of the convergent action of the risk factors concerning the host, the microbial infectivity, and the hospital environment including the hospital personnel, the equipment, the wrong procedures and the sanitation level. These infections are important for their frequency and for the resistance to antibiotics of the bacteria which cause them. Illness gravity and high mortality, especially in newborn units, paediatrics, and intensive care units asked for the statement of national and international systems for control and surveillance of hospital infections. The purpose of this work it was the evaluation of the resistance to antibiotics of Methicillin-resistant Staphylococcus aureus (MRSA), MLSB (Macrolide-Lincosamide-Stretogramin B) and CoNS (Coagulase Negative Staphylococcus), isolated from Universities Hospitals in Cluj Napoca Romania.

Material and Methods: Isolation of Staphylococcus spp. were carried out from different pathological products. Identification of Staphylococcus spp. was performed by phenotypic patterns using the APY system (BioMerieux France) and the VITEK 2 System ATB (BioMerieux, France). The resistance profiles were performed according to NCCLS by disk diffusion method to betalactams, macrolides, lincosamides, quinolones, aminoglycosides, tetracyclines, glycopeptides, sulphamethoxazol & thrimetoprim, and in Mueller-Hinton agar supplemented with 2% NaCl for oxacillin, using antibiotic discs from Bioanalyse Ltd. For inducible MLSB detection has been used D-zone test according to NCCLS. Quality control was achieved using Staphylococcus aureus ATCC 25923.

Results and Discussions: In the period January 2005 - February 2008, 359 S. aureus strains were isolated from sputum (47%), bronchial lavages (20%), pharyngeal exudates (21%), pus (9%), others (3%). The isolated strains were MSSA (74%), MRSA (8%), MLSB (12%) and MRSA+MLSB (6%). From MRSA isolated strains, 6% were Erythromycin and Lyncomycin susceptible; 56% Erythromycin resistant, 38% were resistant to MLSB of which 14% were inducible MLSB and 24% were constitutive MLSB. Were isolated also 95 strains of

S. Aureus and CNS involved in urinary and surgical infections, especially in Surgery Hospitals, from which 8 (42%) were MRSA. In our study no VISA or VRSA was found. MRSA and MRSA & MLSB strains are the most frequent isolated from pus, whereas MLSB resistance phenotype is frequent isolated in sputum and pharyngeal secretions. If all the strains of S. aureus were sensitive to Vancomycin and Teichoplanin, those of CoNS were resistant in 12% to Vancomycin and in 4% to Teichoplanin. We have noticed (even if at a low rate) an increase toward glycopeptides, antibiotics used in severe forms of these infections. The prevalence of MRSA and CoNS is explained by the invasive diagnostic and therapeutic procedures performed. Proper surveillance of medical staff and a rational policy in prescribing antibiotics in this hospitals are therefore mandatory. Resistance to the macrolides (MLSB resistance) in staphylococci may be due to an active efflux mechanism (encoded by the mrsA gene) or to the modification of the ribosomal target (encoded by ermA or ermC genes). MLSB resistance is either constitutive or inducible by certain macrolides. S. aureus and CoNS strains are able to produce a wide number of virulence factors: biofilm, nonfimbrial adhesins, capsule, toxins, etc. Their detection by phenotypic methods is rather difficult and therefore the molecular methods are imposed. Genetic structures coding for the virulence factors, located on the chromosome as well as on mobile genetic elements, are the targets in the molecular methods. PCR technique, colony hybridization, RFLP for genetic regions of medical interest, ribotyping, macrorestriction profile (PFGE) and sequencing are the main tools used for identification of pathogenic strains.

Conclusions: Our data suggest that MLSB resistance is highly prevalent in S. aureus strains and infections caused by S. aureus, especially MRSA and MLSB, continue to be a major challenge in Romania. CoNS are more resistant to antibiotics than S. aureus and can cause serious infections. The combined use of molecular and conventional techniques provides physicians and health care workers with invaluable information that directly affects the treatment.

P10

Study of the Resistance of Enterococcus Faecalis and Enterococcus Faecium Isolated from Inpatients During 2007

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Introduction: Enterococci are regarded as causative agents of serious infections such as bacteremia, urinary tract infections, transfection of wounds. Enterococcal infections have become hard to cope with, due to acquired resistance to many antibiotics.

CultureMedia/Methods: 459 strains from inpatients including the ICU, were studied. The cultures were completed in the usual culture media. Identification was done by classic methods, colony morphology, catalase reaction(-), hydrolysis of esculin(+) and with Vitek2(BIOMERIEUX). Susceptibility testing was done with the disc diffusion method in agar (Kirby-Bauer) and with Vitek2. Glycopeptide resistance was confirmed with E-test.

E.faecalis_343strains-74.4%

E.faecium_116strains-25.2%

5% of strains were isolated from blood cultures, 68.8% from urine samples and 26.1% from other samples.

Results:

A.RESISTANCE(%) TO ANTIBIOTICS:

	PEN	AMP	Gen H	QD	Lnz	Van	Tec
Enterococcus faecalis	30.2	0	48.3	100	0	1.5	1
Enterococcus faecium	91.4	92	34.5	1.7	1.8	25	25

Enterococcus faecium high resistance to Pen, Amp, Amc was noticed. All strains of Enterococcus faecalis were sensitive to Amp and Lnz.

B. ENTEROCOCCI RESISTANCE TO GLYCOPEPTIDE(%)

		VAN	TEC	Phenotype
E.faecalis	4 strains	R	R	VAN-A
	1 strain	R	S	VAN-B
E.faecium	29 strains	R	R	VAN-A

From the 459 strains of enterococcus 34 strains VRE (7.4%) 29 strains VRA E.faecium (85.2%) 5 strains E.faecalis (14.8%) 33 strains belong to the phenotype VAN-A and one strain Enterococcus faecalis to the phenotype VAN-B.

Conclusion: Most enterococci were isolated from urine. E.faecalis is mostly isolated from nosocomial patients. Apart from its native resistance to Quin/Dalfo, presents, by far, higher sensitivity to Pen, Amp, Amc than Enterococcus faecium strains.

E.faecium (mainly VANA phenotype) strains present high resistance to various antibiotics even to glycopeptides.

P11

Strains of *Klebsiella Pneumoniae* Resistant to Carbapenemes During the Year 2007 in General Hospital of Chania

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Aim: In the present study we describe the appearance of infections due to *Klebsiella pneumoniae* species which are resistant to Imipenem (Imp) because of the production of metalloenzymes VIM or KPC. The fore mentioned species, consist a new important problem especially in intensive care unit.

Culture Media and Method: 353 strains of *Klebsiella pneumoniae* were isolated from different specimens. For the isolation the usual culture media were used. The identification took place according to the classical methods, comprised by API-20E and Vitek2 (BioMerieux). The test of antimicrobial drug sensitivity was done by the method of Kirby-Bauer disc diffusion and by the MIC estimation according to Vitek2. The production of the metallo- α 946;-lactamase (MBL) was realized by the test Imp+EDTA and Hodge test.

Results: During the year of 2007 were isolated in total 55 pathogens Imp R (resistant). 20 pathogens were isolated from the urine, 24 from bronchial secretions, 5 from pus, 5 from blood and tree from CSF. All the MBL pathogens were sensitive to Gentamycin. *Klebsiella pneumoniae* (14,7%) of wich 39 were isolated from the intensive care unit at a percentage of 75%.

Conclusion: The appearance of MBL or KPC producing *Klebsiella pneumoniae* pathogens represent a new threat, especially regarding the intensive care unit (75%) which could lead to serious infections with increased mortality. The immediate realization of problem and the adoption of means of control of spreading are necessary for coping with the threat of MBL or KPC resistant *Klebsiella pneumoniae* pathogens.

P12

Detection of Integron Elements and Gene Groups encoding ESBLs and their Prevalence in *E.coli* and *Klebsiella* Isolated from Urine and Stool Samples of Patients who Referred to Mofeed Children Hospital

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Introduction: Antibiotic resistance pertaining of extended spectrum beta lactamases recently has highly increased it could be seen its daily enhancement. The most observations are for Enterobacteriaceae and early studies were for this family of bacteria. Because, *E.coli* and *Klebsiella* species are amongst the usual clinical isolated bacteria, this study has been performed on these two bacteria. Integron elements are short sequences of DNA like transposons. They can transfer genes such antibiotic resistance among the bacteria. Recent studies have revealed these elements could bear the ESBL genes.

Objectives: Determination of prevalence *E.coli* and *Klebsiella* species including ESBL and their relation with integron sequences.

Method: 100 *E.coli* and 100 *Klebsiella* have been isolated from urine and stool samples. Then susceptibility antibiotic testing such epsilometry test (E-test), disc diffusion test and MIC has done with prototypical antibiotics indicative for ESBLs like Ceftriaxon, Cefotaxim, Cefepime, Aztreonam and Clavulanate added to Cefotaxim and Cefepime. ESBL bacteria have been preserved in -70 α 61616;C freezer and PCR performed on them. The studied gene groups for ESBL were CTX, TEM, SHV and Int.

Results: 33 of *E.coli* were positive for ESBL with frequency include: CTX (30), TEM (15), SHV (25), Int (18). 29 *Klebsiella* were positive for ESBL with frequency such: CTX (28), TEM (24), SHV (20) and Int (14).

Conclusion: It could be perceived that among these strains 48% – 54% concomitantly have ESBL genes and Integrons.

P13

Epidemiological Investigation of Multiresistant *A.baumannii* Clinical Isolates in Intensive Care Unit

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Purpose: Emergence of multiresistant *A.baumannii* presents a problem for hospital infection control and institutional antibiotic policies. The surveillance of *A. baumannii* nosocomial infections must be strongly effective especially in Intensive Care Unit, because this area is frequently the epidemiological epicenter for transmission of multidrug-resistance. In the present study we examined the REP-PCR-determined genotypes of *A. baumannii* multiresistant strains.

Materials and Methods: A total of 63 *A. baumannii* strains were isolated from 31 patients from the Intensive Care Unit of Alexandroupolis University Hospital during the period 1.01.07 to 31.12.07. The sources of the isolates were as follow: blood 26, body fluid 1, sputum/respiratory aspiration 16, catheter 17, pus/wound swab 3. Biochemical identification and susceptibility to ampicillin (AM), gentamicin (GM), amikacin (AN), cefuroxime (CXM), ceftazidime (CAZ), ceftriaxone (CRO), ciprofloxacin (CIP), imipenem (IPM), meropenem (MEM) and colistin (CL) were determined by the automated system VITEK2 (BioMerieux). Typing of the isolates was performed by REP-PCR.

Results: The antimicrobial resistance of the 63 *A.baumannii* strains was as follow: AM 63 (100%), CXM 63 (100%), CAZ 63 (100%), CRO 63 (100%), CIP 63 (100%), AN 58 (90%), GM 47 (75%), IPM 59 (90%), MEM 6 (11%) and CL 0 (0%). REP-PCR DNA fingerprints of all isolates revealed the presence of three genotypes: 50 (79%) strains of genotype II, 12 (19,5%) strains of genotype III and only one strain of genotype I (1,5%),

Conclusion: *A.baumannii* isolates presented a high level of antimicrobial resistance. In some cases the strains were sensitive only to colistin. Examination of the REP-PCR DNA fingerprints of the strains revealed the presence of one predominant genotype. The genotypes were not presented intrinsic variety in susceptibility profiles. Operative procedures and precautions to avoid the spread of multidrug – resistant *A.baumannii* requires to confirm or exclude the genetic relationship among the isolates. Rep-PCR method provided high degree of discrimination among the strains and may be used as a rapid technique for epidemiological investigation of *A.baumannii* infections.

P14

Evaluation of Antimicrobial Resistance in *Enterococcus* spp. Isolated in Romania from January 2006 to August 2008

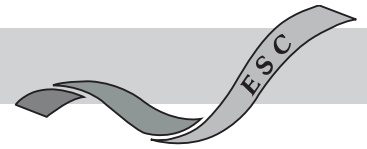
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Purpose: to study the antibiotic resistance in enterococci.

Material and Methods: A total of 216 *Enterococci* strains isolated between January 2006 – August 2008 were collected from two hospitals, at National Reference Center for Enterococci. The samples came from urine, wounds, drain, blood, bile, catheter, sputum, peritoneal fluid, CSF. The strains were characterized by using standard protocols and tested for beta-lactamase. The enterococci were studied for susceptibility to 5 antibiotics: penicillin (Pc), gentamycin (Gn), levofloxacin (Lev), linezolid (Lzd), vancomycin (Va) by seeding agar plates and agar dilution (MIC standard dilution and E test) according to CLSI recommendations.

Results: The data were analysed according to CLSI 2008 and showed the following aspects. 139 strains were identified as *E.faecalis*, 77 as *E.faecium* and 1 strain as *E. avium*. Penicillin resistance was seen at Coltea hospital 78% in *E. faecium* and 3.5% in *E. faecalis*, against Babes hospital with 92.6% in *E. faecium* and 8.1% in *E. faecalis*. High level resistance to Gn revealed the following resistance: at Coltea hospital 72% in *E. faecium* and 21.2% in *E. faecalis*, against Babes hospital with 81.5% in *E. faecium* and 36.7% in *E. faecalis*. Resistance to Lev in *E. faecium* showed 72% resistance at Coltea hospital and 59.3% at Babes hospital. No resistant strain to Va or Lzd was found.

Conclusions: A high resistant percent to penicillin and gentamycin was found in *E. faecium* strains at both hospitals. In the future, more prudent policy for antibiotics, but preserving for seriously ill and immunosuppressed patients is needed.



P15
Antimicrobial Susceptibility of Prevotella Strains Isolated from Abscesses of Fascial Spaces of the Face and Neck

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Purpose: The oral and maxillofacial infections are mainly mixed infections, involving aerobic and anaerobic bacteria, which are often beta-lactamase producers. The aims of the present study were: 1) to identify at species level the Prevotella isolates from pus samples collected by needle aspiration in Romanian patients with abscesses of fascial spaces of the face and neck; 2) to test the susceptibility of these isolates to some commonly used antibiotics.

Materials and Methods: Microscopy of Gram-stained smears and cultures on selective and nonselective media – incubated aerobically and anaerobically – were performed in each pus sample. All strictly anaerobic Gram-negative bacilli strains were identified to genus and species level using the conventional methods of diagnosis and the Rapid ID 32 A system (BioMérieux, Marcy-l’Etoile, France). The Prevotella isolates were examined for the in vitro susceptibility to five antibiotics using the E-test (AB Biodisk, Solna, Sweden). In addition, the beta-lactamase production was investigated by the nitrocefin disk.

Results: The Prevotella strains were isolated in more than 50% of the samples, either alone or in association with other microorganisms and belonged to five species. Prevotella was the main strictly anaerobic bacteria isolated and P. melaninogenica predominated. The results of the antibiotic susceptibility testing indicated that 22% of the isolates were resistant to penicillin due to the beta-lactamase production and 13% of the isolates were resistant to tetracycline.

Conclusions: 1) The Prevotella strains isolated from the investigated abscesses belonged to different species, but no P. intermedia strain was identified; 2) amoxicillin/clavulanic acid, clindamycin and metronidazole were fully active against these anaerobic bacteria isolates and might remain good empirical choices in oral and maxillofacial infections in which these bacteria are involved.

P16
Study of the Occurrence and Resistance of Ouropathogenic Strains of Klebsiella Pneumoniae in Inpatients and Outpatients during the Year 2007 in the General Hospital of Chania

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Aim: To study the incidence of Klebsiella pneumoniae in urine samples and to compare the resistant strains to antibiotics between inpatients and outpatients.

Material and Methods: 128 Klebsiella pneumoniae strains were isolated, in a percentage of 5,6% of a total of positive urine cultures. The cultures were done in the usual culture mediums. The sensitivity test was done by the disk diffusion method in Kirby-Bauer agar and the MIC was calculated by VITEK 2 (Bio-merieux). The standardization was done with API 20E and VITEK 2. The ESBL production was tested with disks of CTX (cefotaxime), CTX-CLA (cefotaxime-clavulanic acid), CAZ (ceftazidime), CAZ-CLA (ceftazidime-clavulanic acid) by VITEK 2.

Results: 18 strains (4%) of Kl. pneumoniae were isolated from the outpatient urine samples and 110 strains (86%) from the inpatients’ samples. From the total of 128 strains, 7 were ESBL(+) in a percentage of 5,4% and a priori, were resistant to penicilline, cephalosporins and aztreonam. One strain was found resistant to imipenem. The seasonal occurrence of ESBL(+) was as follows: January-February: 4 strains July-August: 3 strains.

	AN	Amc	Cfm	Caz	SXT	Cip
R% Kl. pneumoniae strains in inpatients	2,3%	20%	10,2%	8,6%	27,3%	11,7%
R% Kl. pneumoniae strains in outpatients	0%	7%	0%	0%	11,1%	5,6%

Conclusions: Klebsiella pneumoniae as an ouropathogen showed competent sensitivity to aminoglycosids, 2nd and 3rd generation cephalosporins and cinolons. SXT (trimethoprim-sulphamethoxazole) and Amc (Amoxicillinclavulanic acid) still remain a reliable choice of treatment among outpatients. ESBL(+) strains should not be treated with β-lactames, cephalosporins and aztreonam.

P17
Use of the PhenePlate Biochemical Fingerprinting System in Epidemiological Studies of Nosocomial Outbreaks with Extended-Spectrum β-Lactamase (ESBL) Producing Klebsiella Species Strains

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Introduction: Rapid and sensitive methods are essential for typing and monitoring of nosocomial infections. The PhenePlate™ System (DiaTeam, Austria) biochemical fingerprinting system is a method differentiation of bacteria, based on the evaluation of the kinetics of biochemical reactions (carbon sources), and performed in microtitre plates. Aim of the study was to evaluate the usefulness and accuracy of the PhenePlate™ System for typing bacterial strains, with subsequent analysis of the investigated isolates referred to nosocomial outbreak-monitoring.

Methods: A total of 43 extended-spectrum-β-lactamase (ESBL+) producing Klebsiella spp. isolates (30 K. pneumoniae and 13 K. oxytoca) strains were investigated.

The strains were collected over a time period of 3 years (2005-2007) at the University hospital in Graz, Austria, and originated from three nosocomial outbreaks. For testing the PhenePlate™ 16R PhP-KL Kit was used. Results were analysed, cluster formation and dendrograms were generated using the PhenePlate™ analysis software. As reference method PFGE was performed and interpreted according to published procedures.

Results: In the Phene Plate System 37 of the ESBL+ Klebsiella strains clustered in three different trees, with each tree corresponding to one outbreak. Three strains, each detected in one the investigated outbreaks, were identified as unique strains, showing that they were independent of the investigated outbreaks. Furthermore, three Klebsiella spp. strains of the outbreak in 2006 did not cluster in the 2006 tree but in the 2005 tree. Clusteranalysis in the PFGE showed only 2 different clusters, one corresponding to the outbreak in 2005 and one to the outbreak in 2006, the unique strains were identical to those of the PhenePlate System.

Conclusion: The PhenePlate™ System is a rapid and easy to handle method for differentiation of bacterial strains. Compared to PFGE results can be obtained in a short time-period (24 h), a fact, which may be an essential advantage in rapid analysis and monitoring of nosocomial outbreaks. Concerning the studied Klebsiella spp strains, the discriminatory power of the PhenePlate™ system in this study was similar or even slightly better than that of PFGE.

Further investigations have to be conducted to assess the reliability of the PhenePlate™ system.

P18

Evolution of Antibiotic Resistance Patterns of *Pseudomonas* Species Strains Isolated from Different Pathological Products

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Background: One of the most difficult problems in hospitals is the appearance of an increased number of *Pseudomonas* antibiotic resistant strains. The objective of our study is to describe the resistance pattern of *Pseudomonas* strains, against the most frequently used antibiotics.

Methods and results: The sensitivity to antibiotics of 229 strains of *Pseudomonas aeruginosa* isolated from different pathological products in 2003 (95 strains) and 2008 (134 strains) was tested. 77.3% of strains were obtained from patients with urinary infections. Antimicrobial susceptibility testing of *Pseudomonas aeruginosa* isolates was performed by disk diffusion method as recommended by the National Committee for Clinical Laboratory Standards (NCCLS), using a panel of 29 antibiotics. *P. aeruginosa* strains isolated from urine in 2003 showed a high resistance to some beta lactams: Ampicillin, Amoxicilin/Clavulanic acid, Piperacillin/Tazobactam, Aztreonam, to first generation cefalosporines (Cefazolin, Cephalotin), second generation (Cefuroxime, Cefoxitin), and even third generation (Cefotaxime), to Cefpodoxime, Cefixime, and remain susceptible only to Carbenicillin, Carbapenems (Imipenem, Meropenem) and third generation (Ceftazidime) and fourth generation cephalosporins (Cefepime). The Aminoglycosides resistance pattern of isolated strains showed high resistance to Amikacin, Netilmycin, and a low resistance to Gentamycin and Tobramycine. All *P. aeruginosa* strains were resistant to Pefloxacin, showed a high resistance to Nalidixic acid, Ciprofloxacin (75%), Ofloxacin (50%) and a low resistance to Levofloxacin (25%). The resistance of *P. aeruginosa* isolates to Trimethoprim-Sulfamethoxazole and to Tetracycline is also significantly high (75%). Most of the strains isolated from urine were resistant to 8 to 10 antibiotics. Strains isolated from other pathological products were resistant to 5 to 8 antibiotics. The resistance to cefalosporines was similar to that of the strains isolated from urine, the resistance to quinolones oscillated between 33.1% and 51.9%; 29.7% of the strains were resistant to Amikacin, 29.6% were resistant to Colistin and 18.6% to Gentamycine. The resistance percents of strains isolated in 2003 are higher in comparison with the results obtained in 2001. More than 80% of the strains were resistant to third generation cephalosporines, while for the strains isolated in 2001, only 63.6% were resistant. The same situation was found for Quinolones: more than 80% resistant strains in 2003 and between 46.5%-77.2% in 2001. There were found 71.4% resistant strains to Amikacin, in 2003 and 93.69% in 2008. Strains isolated from other pathological products demonstrate a smaller growing resistance trend.

Conclusions: Our results could reflect the implication of some hospital multi resistant strains in nosocomial urinary infections. *P. aeruginosa* strains showed adequate sensitivity only to a limited number of antibiotics. Third generation Aminoglycosides and Quinolones (except Levofloxacin) are no longer of first choice in the treatment of urinary tract infections, because of the resistance pattern of *P. aeruginosa* strains. Resistance to third generation cephalosporins is very important due to the frequent use of these antimicrobials in hospitals. Recommended drugs are broad spectrum penicillins (Carbenicillin), some of third or fourth generation cephalosporins (Ceftazidime, Cefepime, Cefpirome) and carbapenems (Imipenem, Meropenem), Levofloxacin and second-generation aminoglycosides.

P19

Fungemia with *Candida Glabrata* in a Diabetic Patient and Conversion of a Renal Cyst to Fungus Ball

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Introduction: 80% of patients with candidiasis have renal infection. *Candida glabrata* is accounted for 5–21% of them.

Objective: To present a case of a superinfected renal cyst with *Candida Glabrata* in a diabetic patient with fever and candidemia.

Material and methods: A 77-year-old man (with coronary artery disease, diabetes mellitus, hypertension, known bilateral renal cysts and recent hospitalization for atypical infection and acute renal failure 4 months ago) presented with 15-day fever, productive cough, weakness and 24-hour oliguria. Laboratory tests showed elevated white blood cells (WBC:19,9 K/α956;l,neutr.: 90,5%, lymph: 6,1%), normocellular, orthochromatic anemia (RBC: 3,82 x 10⁶/α956;l, Ht:33,9%, MCV:88,7fl, MCH:29,8pg), RAS=101 mm., acute renal deficiency (Ur:95mg/dl, Cr:1,8mg/dl) and urine culture negative for bacteria. Even though the patient's clinical condition gradually improved, the results of 3 consecutive blood cultures showed fungemia with *Candida Glabrata*, so antifungal treatment was given according to susceptibility testing. For the detection of the fungemia's location we performed: chest CT(-), brain CT(-), ophthalmologic examination (-), cardiac u/s and transoesophageal echocardiography (-), renal u/s α954; α945; α953; CT of the abdomen showed complicated renal cyst of the right kidney (containing an echogenic material reported at the time of to be either blood clot or a fungus ball). Under CT guidance fluid was aspirated from the renal cyst of the right kidney and the cystic fluid cultures yielded *Candida Glabrata*, the same strain which was obtained from 3 blood cultures. 1,5 month after discharge his clinical and laboratory condition were restored (normalisation of renal function Ur: 73 mg/dl, Cr: 1,1 mg/dl, the right kidney's cyst decreased in size d=2,6 cm and RAS: 34 mm.)

Results: In this case what is notable is the superinfection of a pre-existing renal cyst with the same strain of fungus that was developed in the blood culture (*Candida Glabrata*) and the successful, conservative treatment with the proper adjunct antifungal therapy.

P20

Fatal Case of Brain Abscess due to Fungus *Scedosporium Apiospermum*

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Introduction: *Scedosporium apiospermum* the asexual form of *pseudallescheria Boydii* are ubiquitous saprophytic fungi that commonly cause cutaneous infections. However in certain circumstances can also cause invasive disease which can involve the central nervous system (CNS) When the CNS becomes involved, treatment is difficult, therapeutic options are limited and the prognosis is poor.

Patients and Methods: An 86 years old previously healthy man developed acute sinusitis and was treated in ENT dept. Cultures of sinus drainage specimens were negative for both fungi and bacteria. Ten days later the patient presented again in the emergency department lethargic with a right sided hemiparesis. Cranial C/T and Brain MRI with contrast medium revealed a left parietal intracerebral ring-enhanced lesion with surrounding brain edema. Surgical Drainage of the abscess was performed via burr hole and purulent material was sent for culture and cytological examination. Microscopic examination showed the presence of septate hyaline hyphae and single-celled oval conidia with typical truncated bases. These conidia were formed singly or in small groups on annellidic conidiophores. Based on macroscopic and microscopic characters, the fungus was identified as *Scedosporium apiospermum* Voriconazole 400 mg/day every 12 h for 2 doses was initiated after these results but despite this the patient's condition worsened more and finally he died due to multiple organ failure 20 days after his first admission.

Conclusions: The correct microbiological diagnosis is essential to the treatment of infections due to *S. apiospermum*. We believe that voriconazole despite of our patient's outcome is the treatment of choice for *S. apiospermum* infections. We suggest an early high dose voriconazole treatment of patients at risk (such as immunosuppressed patients, near-drowning victims) together with mycological monitoring to reduce high mortality score of over 75%.

P21
Invasive Candidiasis, Candida Colonization and Antifungal Treatment in Intensive Care Patients after Cardiothoracic Surgery
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Purpose: To improve the diagnosis, the timing of diagnostic procedures and antifungal treatment of invasive *Candida* infections in intensive care patients, a prospective surveillance study was performed at our cardiothoracic intensive care unit (ICU) of the Medical University of Vienna.

Methods: Patients admitted to the cardiothoracic ICU between December 2006 and November 2007 were enrolled into the study. Two times weekly surveillance cultures (n=2413) of inguinal swabs (17%), axillary swabs (17%), pharyngeal swabs (17%), urine (14%), nasal swabs (13%), bronchial lavage (11%), surgical wounds (4%) and anal swabs (2%) were taken. At each time point material from at least five different body sites was analyzed for presence of *Candida*. The Candida Colonization Index (CI) was calculated for each patient by the number of *Candida* positive samples per all samples. Patients were grouped as follows: no colonization (CI α 8804; 0.2; n=17), colonization (CI $>0.2 <0.6$; n=31) and severe colonization (CI α 8805; 0.6; n=37). As risk factors time of stay on ICU, number and type of surgeries, sex and age of patients and start of antifungal treatment were investigated.

Results: A total of 85 patients were included into the study, the overall mortality was 36.5%. The different types of surgeries included valve replacement, aortocoronary bypass, heart or lung transplantation, implantation of left ventricular assist device or artificial heart with a mean of 2.1 surgeries per patient. During the first four weeks on ICU the dominating pathogen was *Candida albicans* (39-49% of all cultures). The percentage of Non-*albicans* species, particularly *Candida parapsilosis* and *Candida glabrata* increased over time. Antifungals were applied in 55.3%. Sex, age of patients, the type of surgery and the start of antifungal treatment were no significant risk factors for severe colonization. A significant difference between colonized and severely colonized patients were detected for the time of stay on ICU (mean = 21.8 days versus 46.8 days; p<0.001).

And the number of surgeries (mean = 1.7 versus 2.6; p=0.02). All patients who developed Candidemia (n=4) were previously severely colonized with *Candida*.

Conclusion: Prolonged stay on ICU and number of surgical interventions of >2 predicted for severe colonization in patients at the cardiothoracic ICU. Candidemia was exclusively detected in patients who were previously severely colonized with *Candida*.

P22
Effect of Disinfectants and Caspofungin Against Planktonic and Sessile Cells of Candida spp.
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Objectives: The aim of this study was to evaluate the activity of 3 different disinfectants chlorhexidine digluconate, Akacid plus® and hydrogen peroxide compared to the antifungal caspofungin against planktonic and sessile cells of *Candida* spp.

Methods: As test strains 40 clinical isolates of *Candida* spp. including *C. albicans*, *C. krusei* and *C. tropicalis* were used. The activity of Akacid plus®, chlorhexidine, hydrogen peroxide and caspofungin was determined against planktonic cells according to CLSI guidelines for antifungals using broth microdilution method. For antifungal susceptibility testing of sessile cells, isolates were incubated overnight in yeast peptone dextrose resuspended in RPMI 1640 to a cellular density equivalent to 1.0x10⁶ CFU/ml. Cells were grown for 48 h in 96-well-microtiter plates, and then treated with 100 µl of Akacid plus®, chlorhexidine and hydrogen peroxide at final concentrations of 0.25, 0.5, 1, 2 and 4% compared to caspofungin at concentrations of 64, 128, 256 and 512 mg/l for 48 h at 35 °C. The cells were fixed and stained with crystal violet. The mean optical density was used for quantification using a routine microtiter-plate-reader at 490 nm. Additionally, fungal growth following antimicrobial treatment was examined.

Results: MICs of Akacid plus® and caspofungin against planktonic cells of *Candida* spp. were comparable and reached MIC values of 0.03-8 mg/l, whereas MICs of chlorhexidine and hydrogen peroxide ranged from 16 to 32 mg/l and from 128 to 256 mg/l. Low concentrations of caspofungin at 64 mg/l caused a 62% reduction of the sessile cells of *Candida* spp. Treatment with 0.25% chlorhexidine and Akacid plus® led to reduction of the sessile cells in 59% and 74%, whereas hydrogen peroxide showed no effect. No viable cells of *Candida* spp. were detected in biofilms treated with 0.25% Akacid plus®, 0.25% chlorhexidine and 0.1% caspofungin.

Conclusions: Caspofungin and cationic antimicrobials showed high activity against sessile and planktonic cells of *Candida* spp., whereas hydrogen peroxide was found to be ineffective.

P23
Prevalence and Antifungal Resistance of Candida Isolates
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Purpose: The study of prevalence and antifungal resistance of *Candida* isolates from an group of haematological malignancies patients.

Materials and methods: The authors have retrospectively analyzed an group of 34 patients with haematological malignancies that received chemotherapy, admitted in Department of Infectious Diseases. The positive diagnosis was established based on physical examination (fever, weight loss, white depots on tongue, headache, anorexia, asthenia etc.), and biological samples (leukocyte values, fibrinogen, CRP, ESR, blood cultures, sputum cultures, ELISA, Western blot, glosal exudate and CHROM-AGAR cultures). We used identification with API *Candida* system and antifungal sensibility tests with ATB Fungus 2 system testing 5-flucytosine, amphotericin B, fluconazole, itraconazole and direct microscopy examination of fungal specimens.

Results: The female-to-male ratio was 14/20, mean age of patients was 36.2 years; were diagnosed 11 patients (32.35%) with *Candida* infections; 9 patients with glossal candidiasis and 2 have had oropharyngeal candidiasis. There have been isolated and identified 5 strain of *Candida albicans* (45.45%) and 6 strain of *Candida non albicans* (54.54%) from which 2 *Candida glabrata*, 2 *Candida kefyr*, 1 *Candida tropicalis* and 1 *Candida parapsilosis*. Out of 5 strain of *Candida albicans*, 3 have been resistance to fluconazole and all with sensitivity to itraconazole. Out of 6 strain of *Candida non albicans* 4 have been resistance to fluconazole and 2 to itraconazole. The isolation of these species, along with the sensitivity at antifungal drugs test have allowed an antifungal treatment with satisfying results.

Conclusion: *Candida albicans* remain the main etiologic agent for the oropharyngeal fungal infections in haematological malignancies patients. It requires the modification of the therapeutic strategy and the introduction of new antifungal drugs based on antifungal sensibility.

P24
Psychosocial and Economic Support Given to HIV Positive Women by their Husbands in Sero Discordant Marriages
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Background: The burden of HIV causes psychological, social and economic stress on couples and more so when the two involved are sero discordant, because they generally face greater adversity. Several studies have analysed sero discordant marriages and enumerated factors that influence marital stability: longer duration of marriage, extended family members, children and economic constraints are just a few. This study aims to assess the psychological and economic support HIV positive women receive from their HIV negative husbands.

Methods: A cross sectional descriptive study was carried out using data collection from self administered questionnaires to the women. Study population included HIV positive women in sero discordant marriages from 3 HIV centres in Abuja, Nigeria.

Results: Of the 51 women sampled, modal class-interval for age group in both men and women was 35-44 years at 53% and 43.1% respectively. Mean age for women was 37.3 years and men was 42.0 years. 68.6% had been married for >5 years and 29.6% have had HIV >5 years. 76.4% of the men were breadwinners. 50.9% of the women had their husbands follow them to the clinic or remind them of their ARV's and clinic appointments always 19.6% never did. 74.4% sometimes had their husbands collect their drugs for them by proxy. Before HIV diagnosis, 54.9% of the women's husbands paid their medical bills always, 9.8% did not and after diagnosis, 60% paid and 11.7% did not. Before diagnosis 19.6% of the husbands used to buy their wives gifts and take them out always, 29.4% never did. After diagnosis 15.6% did and 37.2% did not. Sexual intercourse before diagnosis was frequent in 49%, few times in 37.2% and hardly in 15.7% had. It become 21%, 52.9% and 25.4% respectively after diagnosis.

Conclusion: This study shows that the husbands in these marriages offer an appreciable amount of support to their HIV positive wives as evidenced by the slight variation seen between the support their wives received before and after diagnosis. However, our results may reflect the high literacy and socio-economic status of the clients being mostly residents of a developed city.

P25

Impact of the National Immunization Program for PCV7 on IPD among German Children

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Background and aims: Since 1997 the German National Reference Center for Streptococci has monitored the epidemiology of invasive pneumococcal disease (IPD) in German children up to 15 years of age. In July 2006 evidence of the beneficial effects of a heptavalent conjugate vaccine on the incidence of IPD led to a general recommendation for the vaccination with a 7-valent pneumococcal conjugate vaccine (PCV7) for all children up to the age of 24 months. In this study we have monitored the effects of PCV7 on the incidence of IPD in Germany.

Methods: Cases of IPD in children were reported by microbiological laboratories and pediatric hospitals. Species confirmation was done by optochin testing, bile solubility and serotyping.

Results: As from January 2007 approximately 70% of all newborns in Germany have been vaccinated with at least one dose of PCV7. In the second half of 2007 we saw the first effects of the PCV7 vaccination. In children under 2 years of age the number of cases caused by vaccine serotypes reported per month decreased by 50%. The decrease was mainly caused by serotypes 14 and 23F. The number of non-vaccine serotype cases slightly increased. Significant changes could not be observed in age groups 2-5 and 5-15.

Conclusions: First effects of PCV7 are seen in IPD cases in German children under 2 years of age. A possible replacement with other serotypes was observed.

P26

Epidemiology and Treatment of Pneumonia in HIV-Infected Individuals in Vienna from October 2007 to February 2008

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Purpose: Pneumonia is a major cause of illness and death among HIV-infected patients with large spectrum of common and uncommon causative pathogens. We report 19 cases of radiologically confirmed pneumonia in HIV-positive individuals admitted to the Department of Immunodermatology of the Medical University of Vienna from October 2007 to February 2008.

Methods: To evaluate the specific pathogen causing the infection, several blood and sputum cultures for bacteria and mycobacteria and pneumococcal and legionella antigen detection were performed for all patients. In special cases CT-scans or bronchoalveolar lavage for pneumocystis, Herpes simplex or cytomegalovirus PCR were performed.

Results: The distribution of the causative agents was as follows: Four cases of pneumococcal pneumonia, two cases of pneumocystis jirovecii pneumonia, two cases of lung tuberculosis, two cases of atypical mycobacteriosis, two cases of Gram-negative sepsis, two cases of herpes simplex virus pneumonia, one case of Enterococcus faecalis sepsis, and one case of systemic cytomegalovirus infection were detected. In three cases more than one single pathogen per patient could be identified. As first line empirical treatment most patients with community-acquired pneumonia received moxifloxacin 400 mg monotherapy, which was well tolerated by all patients. Moxifloxacin was effective in 67% and increased transaminase levels only in 1 of 9 patients, even in patients with chronic active viral hepatitis.

Conclusion: The predominating pathogen of definite community-acquired pneumonia in HIV-positive was Streptococcus pneumoniae, followed by Pneumocystis jirovecii pneumonia. In nosocomial pneumonia a total different spectrum of causative agents, especially multi-resistant gram-negative bacteria, were detected. Most patients received moxifloxacin intravenously, which was well tolerated in all patients, but was only a sufficient treatment in patients without evidence of pneumococci, multi-resistant Gram-negative bacteria, mycobacteria, pneumocystis or cytomegalovirus infection.

P27

JC Papovavirus Leukencephalopathy in a Patient with B-CLL Receiving Long Term Chemotherapy and Rituximab

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Progressive multifocal leukencephalopathy (PML) is a rare demyelinating infection of the central nervous system caused by viruses (CMV, JC, BK). It is almost exclusively described in profound immunocompromised patients developed as a primary infection or by reactivation of the virus after a latent infection. PML is typically observed among patients with severe immunodeficiency like HIV or in the setting of autologous or allogenic transplantation.

Our patient was a 64-year-old woman with B-CLL diagnosed in 2000 in state B of Binet treated with 12 cycles of fludarabine until July 2007 in a B-cell and lymphoma-reducing manner. For the reason of progressive disease 3 cycles of rituximab, fludarabine, mitoxantrone and cyclophosphamide were added (dose reduced causing hematologic toxicity) until October 2007 continuing rituximab once a month up to Mai 2008. In a CT scan mixed response was observed with progressive disease infradiafragmatic and regression supradiafragmatic. Corticosteroids could stop B-symptoms and leads to a distinct shrinkage of the lymphomas within 2 weeks. Later on she presented with confusion and disorientation. An MRI of the brain showed subcortical and periventricular lesions with increased signal on T2-weighted and fluid attenuated inversion recovery (FLAIR) suggestive of a vascular origin. Her symptoms progressed by aphasia and weakness of the right leg corresponding to increased size and number of cerebral lesions. A lumbar puncture showed no malignant cells and no evidence of inflammation. A PCR analysis was negative for Toxoplasma gondii, HSV, VCV, CMV, EBV, and enterovirus but positive for JC-DNA (215.000 copies/ml). A rapid progression leads to death two months after onset of symptoms. Antibody deficiency syndrome corresponding to CLL under treatment with chemotherapy and rituximab lead to a severe immunosuppression enabling unusual viral infections such as PML. A case report and review of the literature of PML in CLL well be presented.

P28

Disorders of Lipid Metabolism in Patients with HIV/AIDS Under Antiretroviral Therapy

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Purpose: the study of disorders of lipid metabolism in Patients with HIV/AIDS under antiretroviral therapy.

Methods: The authors have studied a group of 24 patients with HIV/AIDS under antiretroviral treatment, hospitalized in Clinic of Infectious Diseases Timisoara. Biologically were determined the following samples: cholesterol, HDL cholesterol, VLDL cholesterol, triglyceride, glycemia etc. Anthropometrical measurements included weight, height and body mass index (BMI) with immunological and virological status for each patients. The fat distribution disorders were established by repeated objective examinations, measuring of skin fold at abdominal level, thigh, triceps muscle, thorax, face and neck. All patients were following a treatment with diverse regimes of NRTI, NNRTI, IP of over two years, being included in the clinical database.

Results: Lipid metabolism abnormalities was recorded in 15 patients (62.50%); 11 patients (45.83%) with high levels of total cholesterol; 16 patients (66.66%) with low levels of high-density-lipoprotein (HDL); 14 patients (58.33%) with high level of low-density-lipoprotein (LDL) and 12 patients with high triglyceride levels; 8 patients with hyperglycemia. Fat redistribution was recorded in 15 patients (62.50%): 8 patients (33.33%) were diagnosticated with lipatrophy, 6 patients (25.00%) with lipohipertrophy at abdominal and trunk level and 1 patient with mixed syndrome; we mention that out of a total of 15 patients with lipid abnormalities, 12 were under stavudine or zidovudine (p=0.04) and 3 used protease inhibitors (indinavir, nelfinavir) for over two years.

Conclusion: Ours results demonstrate that stavudine and zidovudine used in therapeutical regimens increase the risk for lipid metabolism abnormalities in patients with HIV/AIDS.



P29

Liver Toxicity of Nevirapine and Efavirenz in Patients with HIV/AIDS

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Purpose: to assess the liver toxicity of nevirapine (NVP) and efavirenz (EFV) in patients (pat.) with HIV/AIDS or coinfection with VHB or VHC.

Materials and methods: The authors studied a group of 32 patients with HIV/AIDS treated with NVP, EFV+reverse transcriptase inhibitors (RTI) or protease inhibitors (PI) between 2-3 years, hospitalized in Department of Infectious Diseases Timisoara. Biologically were determined: ESR, glycemia, amylasemia, amylasuria, ALT, AST, BD, BT, AP, Gamma-GT, Ab IgM HAV, Ab IgM HBe, Ab HCV, HBs Ag, Ab HDV, cholesterol, triglyceride etc. The statistic data was processed with the EpiInfo 5 program.

Results: 15 pat. (46.87%) were treated with NVP and 17 pat. (53.12%) with EFV; 5 pat. treated with NVP (3 pat. NVP+RTI and 2 pat. NVP+PI) and 8 pat. treated with EFV (5 pat. EFV+RTI and 3 pat. with EFV+PI) was recorded with liver toxicity (ALT α 8805; 2-fold, p=ns); 86.4 \pm 2.1 UI/ml is mean ALT values in pac with NVP+RTI versus 122.7 \pm 11.2 UI/ml in pac NVP+PI (p=ns); 115.6 \pm 3.2 UI/ml is mean ALT values in pac with EFV+RTI versus 145.8 \pm 12.2 UI/ml in pac with EFV+PI (p=ns); We mention 3 pac with NVP and coinfection VHB, VHC and 4 pac with EFV and the seem coinfection. 130.6 \pm 9.4 UI/ml is mean ALT values in pac with NVP+coinfection VHB, VHC versus 160.7 \pm 5.2 UI/ml in pac with EFV+the seem coinfection (p=ns);

Conclusion: The liver toxicity of NVP and EFV is higher in patients HIV with coinfection VHB, VHC; is necessary the control of liver enzyme in all patients with NVP or EFV therapy.

P30

Infectious Complications Following Autologous Stem Cell Transplantation in Patients with Lymphoma

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Purpose: Infections remain a major cause of morbidity and mortality following autologous hematopoietic stem cell transplantation. Knowledge of local epidemiology and patients' risk factors are essential for their optimal management.

Methods: 72 consecutive patients (median age 45, range 19-68 yrs) with relapsed or refractory Non-Hodgkin's Lymphoma (NHL, n=53) and Hodgkin's Disease (HD, n=19) treated with autologous peripheral blood stem cell transplantation (PBSCT) in a single center have been evaluated for infectious complications following PBSCT.

Results: Febrile neutropenia (FN) occurred in 45 (62.5%) patients at a mean of 6 days after transplantation (range 1-9, SD 1.8). Twelve patients (26.7%) had proven bacteremias, 11 (24.4%) other microbiologically documented infections (MDIs) and 22 (48.9%) had clinically documented infections (CDIs). Gram+ microorganisms were responsible for the majority of all documented infections (58.3% of bacteremias and 54.5% of other MDIs). One patient died as a consequence of sepsis. Correlations of a number of variables available at time of FN onset with the duration of febrile episode have been investigated. Patients with higher CRP levels at FN onset and those with earlier onset of fever following PBSCT had significantly longer (p=0.03 and p=0.002, respectively) duration of fever. Other variables, including age, No. of previous lines of therapy, No. of stem cells reinfused, ANC and monocyte counts as well as peak temperature values at day of fever onset did not show statistically significant correlations with duration of fever. All patients with diabetes developed FN and, although not reaching statistical significance (p=0.08), its duration was longer in comparison to other patients.

Conclusions: Infections are serious but manageable complications of PBSCT. Gram+ microorganisms remain the major cause of documented infections.

P31

Urinary Tract Tuberculosis in a Patient with Anti-TNF-Induced Immunosuppression

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Introduction: Tumor Necrosis Factor (T.N.F) is a proinflammatory cytokine, which has a key role in the pathogenesis of various autoimmune disorders, including Rheumatoid Arthritis. During the last years, the use of anti-T.N.F. agents in the therapy of several immune-inflammatory diseases has been proven as particularly efficient. Their use however has been related to a variety of opportunistic infections. Tuberculosis in such patients often presents as extrapulmonary or disseminated, potentially life-threatening, disease.

Aim: Presentation of a patient who developed urinary tract tuberculosis, after initiation of therapy with anti-T.N.F. agent.

Material and Method: A 74-year old female patient was admitted in our clinic because of fever during the last 17 days. Rheumatoid arthritis had been diagnosed 25 years ago. Since 4 months the patient was on an anti-T.N.F. agent (Humira). She also received tb. Prednisolone 5 mg daily and tb. Methotrexate 12.5 mg weekly. The physical examination revealed no abnormal signs. The laboratory tests showed slight lymphopenia and mild orthochromatic anemia, ESR 66 mm/1st h. PPD TEST (Mantoux) was positive (10cm), whereas one year before it had been negative. The imaging studies revealed fluid in Douglas pouch and the paracentesis proved lymphocytic exudation. The endoscopic examinations were negative. The patient refused to undergo IVP.

Result: 15 days after the admission, while the fever still persisted, anti-TBC treatment was commenced. 3 days later complete apyrexia was noted. 10 days later, the patient left the hospital afebrile and in good clinical condition. After one month, while the cultures of gastric fluid, ascetic fluid and sputum were negative, the urine culture was positive for mycobacterium tuberculosis. Urinary tract tuberculosis was diagnosed. The patient received anti-TBC treatment for 9 months and is in excellent general condition.

Conclusion: The clinician should be alerted to perform diagnostic tests for TBC in every patient who receives treatment with an anti-TNF agent and who presents with fever, weight loss or cough.

P32

Frequency of Pneumonia Development in Hospital Patients with Rheumatic Diseases

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Purpose: Pneumonia (PN) is one of the complications deteriorating prognosis in immunocompromised patients (pts) including pts with rheumatic diseases (RD). Information about prevalence and course features of PN in hospital pts with rheumatoid arthritis (RA), systemic lupus erythematosus (SLE) and primary Sjogren α 8217;s syndrome (PSS) in Russian Federation is lacking.

Objectives: To perform retrospective evaluation of PN frequency and course features in hospital pts with rheumatic diseases (RD).

Methods: 9203 pts hospitalized in the hospital during 6 years were analyzed. Results: Clinical and radiological signs of PN were revealed in 104 pts (1,1%) Table. Characterization of patients with PN

	RA (n=6818)	SLE (n=2038)	PSS (n=347)
Number of pts with PN abs (%)	58 (0,8)	40 (2)	6 (1,7)
Mean age (M \pm SD)	56,1 \pm 9,8	38,4 \pm 14,0	60,0 \pm 18,2
Sex male/female	12/45	7/33	0/6
Median duration of the disease (years)	8 [4; 13]	6 [3; 14]	21 [10; 30]
Steroids (number of pts) total	23 (39)	34 (85)	2 (33)
dose α 8805;10 mg/day	7	28	0
Immunosuppressive drugs abs (%)	18 (33)	19 (22)	2 (33)
NSAIDs abs (%)	52 (90)	6 (15)	1 (17)

Fever was absent in 69% of RA, 50% SLE and 33% PSS pts, productive cough - in 52%, 37%, 67%, typical radiological picture in 55%, 55% and 67% of pts respectively. Leucocytosis was revealed in only 28% of RA and 16% PSS pts. Stab neutrophil shift was present in 45% of SLE pts. 40% of SLE, 32% of RA and 17% of PSS pts fulfilled definite PN criteria. In other cases the diagnosis was considered as probable. Antibiotics of different classes were administered (cephalosporins, macrolides, fluoroquinolones). Mortality of pts with PN was 3%.

Conclusion: PN frequency in hospital pts with RD was 1,1%. Previous treatment with steroids and NSAIDs possibly changes clinic picture and course of concomitant PN. Steroid administration can be considered as possible risk factor of comorbid respiratory infection development in RD.

P33

The Prevalence of Respiratory Infections in Haematological Malignancies Patients

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Purpose: To assess the prevalence and the etiology of the respiratory infections in a group of haematological malignancies patients.

Materials and methods: The authors have retrospectively analyzed a group of 42 patients with haematological malignancies diseases hospitalised in the Clinic of Infectious Diseases Timisoara. The positive diagnosis of respiratory infections was based on clinical elements (fever, cough with expectoration, bronchoalveolar rales, sweats, chest pain, dispnea, anorexia etc.), biological (ESR, leukocyte values, Fibrinogen, CRP, ASLO, electrophoresis, sputum smear testing, sputum cultur testing, blood cultures, etc.) and paraclinic elements (chest radiography, spirometry, computer tomography). The results of the biological and paraclinical investigations were recorded in the individual patient file. All patients were in data base of Hematology Clinic and undergone chemotherapy or corticosteroids treatment. The data obtained was processed with Epi Info 5 statistical programme.

Results: Out of study group was recorded 13 patients (30.95%) with respiratory infections ($p=0.01$): 3 patients with interstitial pneumonia, 4 with bacterial pneumonia, 1 with acute bronchitis, 1 with chronic bronchitis, 2 with bronchopneumonia, 1 with chronic obstructive pulmonary disease, and 1 with pulmonary tuberculosis. Only for 8 patients the ethiological agent was established: 2 patients were discovered with Streptococcus pneumoniae, 3 patients with Staphylococcus aureus, 2 patients (21.87%) with Klebsiella pneumoniae and 1 patient with Mycobacterium tuberculosis. We mention 11 patients with glossal candidiasis associated ($p=0.03$). The evolution was unfavorable in 2 cases. The mean duration of hospitalization was 17.8 days.

Conclusions: By high prevalence (30.95%) of respiratory diseases the patients with haematological malignancies represent a population group with high risk; it's necessary improving of prophylaxis measures and treatment.

P34

Invasive Streptococcus Pneumoniae Disease in Children below 5 Years of Age in 2007 and 2008 in Austria

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We prospectively collected surveillance data on invasive Streptococcus pneumoniae disease (IPD) in children below 5 years of age from all Austrian paediatric hospitals and collaborating microbial laboratories. Streptococcus pneumoniae had to be confirmed by culture/PCR from blood, cerebro-spinal fluid or other normally sterile body sites. Additionally isolates of the strains were stored for serotyping. From January 2007 to September 2008 a total of 43 IPDs was reported with an overall fatality rate of 4.7% (including 9 cases of meningitis, fatality rate of 22.2%). The incidence for Austria in 2007 was 7.3/100 000 in children below 5 years, and 12.1/100 000 in children below 24 months of age. During the period from January to September 2008 the incidence rate was 4.7/100 000 and 5.1/100 000 for the two age groups, respectively. Two children suffering from meningitis had a permanent hearing loss as sequelae. Two other infants died of meningitis. We could identify some risk factors in our 43 IPD cases: 2 preterm infants (1 meningitis), 1 child with lack of pyruvate dehydrogenase, one infant had a defect of the cardiac septum.

Two children had received one vaccination against IPDs. Another 2 infants were vaccinated twice against IPDs. One of the children who died (from a non-vaccine serotype) had received 3 vaccinations against IPDs. Identification of serotypes was available in 18 cases. In our analysis we found the serotypes 14 (4 cases), 1 (2 cases), 3 (2 cases), and 7F (2 cases, 1 meningitis). Additionally, we identified the serotypes 5, 9N (1 meningitis), 9V, 10A, 16F, 19A (1 meningitis), 19F and 25A once each.

Incidence rates of IPDs were similar to the rates observed earlier. This emphasizes that a general and subsidized immunization program for Austria is urgently needed.

P35

Laboratory Diagnosis of Viral Infections - Thematic Course from Post Graduate Studies for acquiring a major in "Virology"

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The knowledge of epidemiology and pathogenesis of viral diseases is due, to a great extent, to the laboratory studies, obtained by conventional methods of viral diagnosis. They include both the classic methods for isolating of viruses in cell cultures, chicken embryos and laboratory animals, as well as serological tests. However, great attention is paid to the development of quickly and highly effective, easily applicable methods for diagnosis through direct usage of clinical material for a viral study. So, scientists today can easily find out not only the possible etiological agent, but also the quantitative alterations in the course of the infection and viral monitoring. It is already a fact in the treatment process of a number of chronically leaking viral infections. The quantitative viral markers, studied in the development of the illness, give some idea for the alterations in the population of the viral agent. There is a simultaneous routine quantitative studying of the antibody titers after an infection, prophylactic or chemotherapy. This report is centered on a thematic course of the "Laboratory of Cell Cultures" of the National Centre of Infectious and Parasitic Diseases and has been developed and put into practice for the last five years in the aforementioned Laboratory. It is a part of post - graduate studies for medical and non - medical specialists. The schedule of the discipline includes 60 teaching hours. This course aims to enlarge the knowledge and skills of the students on the methods of early diagnosis and on the epidemiology of a number of viral diseases. The qualification of those completed the course will allow them to accomplish specialized work as a "doctor - specialist of Virology".

P36

Acute Suppurative Thyroiditis, as a Cause of Neck Infection: Diagnostic Approach and Treatment of Two Cases

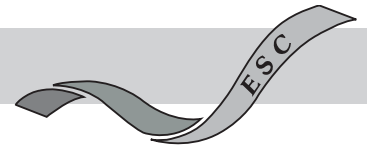
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Purpose: Acute suppurative thyroiditis is an extremely rare, usually bacterial thyroid infection with abscess formation. It typically presents in the course of an upper respiratory tract infection. The treatment of choice is surgical drainage and parenteral antibiotics. In our presentation we would like to demonstrate the rarity and severity of the disease, as well as, the life threatening complications that imposes carefully planned diagnostic procedures.

Material and Methods: We present two male patients admitted to the outpatient department of our hospital with symptoms of high fever, difficulty in swallowing and painful neck masses. Both patients presented with prolonged infections. The first had been unattended, while the second was referred to our clinic from another department where he was recovered for acute renal failure. Both patients received intravenous antibiotics and hydration and underwent laboratory tests, FNA biopsy and CT scan of the neck, which demonstrated the position of the abscess into the visceral space. Thyroid hormones and antithyroid antibodies showed no pathological elevation.

Results: The first patient had a rare complication of tracheocutaneous fistula, which was treated with surgical drainage and tight closure of the external opening. The second patient had a spread of the infection from the parapharyngeal space unilaterally and underwent surgical drainage also. Diagnosis was based on CT scan and FNA results.

Conclusion: Acute suppurative thyroiditis can be the cause of neck infection, either primarily or secondary to other neck infection. Clinical manifestations are atypical and differential diagnosis is based mainly on FNA biopsy.



P37

Chronic Phase of Schistosomiasis in a Patient with Pulmonary Involvement

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Introduction: Human schistosomiasis is caused by five species of this parasitic trematode, which belong to the phylum Platyelminthes. The five species are estimated to infect 200 to 300 million people in South America, the Caribbean, Africa, the Middle East, and Southeast Asia.

Clinical presentation: Incidence of a patient having disease in the lungs during chronic schistosomiasis.

Material: A patient, 27 years old, from Egypt comes to our hospital because of chest pain (at the right lower ribs), fever and hemoptysis. The patient took three months ago, in Egypt, treatment with praziquantel because the presence of schistosome ova in excreta was established. Also he was given directions for repeating the praziquantel treatment in 6 months, which never happened.

Process: He was afebrile with excellent blood pressure and oxygen saturation. The examination of the CNS, ear-nose-pharynx, heart, lungs and abdomen was free of symptoms. CBC, total biochemical profile, CEA, Ca19-9, aFP, RF, immunologic tests, antibody detection tests for *Leptospira* spp, *Salmonella typhi*, dengue virus, Hepatitis A, B, C, D viruses, HIV, *Rickettsia rickettsii*, *Plasmodium* spp were normal. Multiple cultures of sputum and urine were negative for common microorganisms and for mycobacterium TBC. Sputum cytology was negative except from a few ferrum-biting cells. The only abnormal findings were D-Dimers=1.9 and mild microscopic hematuria (35/.....). Arterial blood gases were between normal levels. U/S of the abdomen revealed only splenomegaly (13.5 cm). Thoracic HRCT was unremarkable.

Conclusions: Evaluating the personal and geographic history, the clinical presentation and the exclusion of other probable etiologies we conclude that the patient, during chronic schistosomiasis, has developed disease in the lungs too.

P38

Bacteremia with Pasteurella Multocida in an Elderly Patient

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Introduction: *Pasteurella multocida* (P.M.) is a Gram-negative coccobacillus found in 70-90% of oral cavities of cats, and as well, is isolated from the digestive systems of dogs, rats, rabbits, monkeys, and other animals. However, this organism can cause serious soft tissue infection. Systematic disease is rare. *P. multocida* often acts as an opportunistic pathogen in the very young or elderly, or in patients with liver dysfunction, causing bacteremia, septic arthritis in damaged joints, osteomyelitis, meningitis, pneumonia and pulmonary colonization.

Objective: To point out the role of P.M. as etiologic agent provoking infections to pet owners. **Patients and methods:** A 94-year-old woman was admitted for high fever up to 40C with rigors, weakness and subcrepitations bilateral in lower lung fields in the auscultation. The laboratory tests showed elevated white blood cells, anaemia, elevated liver enzymes, elevated acute phase inflammation index and normal liver ultrasound image. The patient gradually ameliorated her clinical and laboratory condition (under amoxicilline-clavulanic acid) but the kind of the infection was not determined yet.

Results: The blood culture result showed bacteremia with gram (-) pathogen *Pasteurella multocida* sensitive to ampicilline and doxycilline. It was also revealed from patient's history that the old lady had domestic animals (cats). Animal scratches or bites, or local wound infections were not found. The fever went down after the patient receiving the proper antibiotic therapy and returned home in a very good condition.

Conclusions: Infections with P.M. should be taken into account in every Patient pet-owner, even though there is no apparent soft tissue infection.

P39

Infections of Anaplasma Phagocytophilum

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Aim: The discussion of a short series of patients, presenting with non-specific, flu like symptoms, secondary to infection with human Ehrlichiosis.

Cases presentation: Four patients (3 males), with a mean age of 41 years, presented to the Accident and Emergency department with persisting pyrexia. All four patients reported close contact with either cattle or mice. On clinical examination three of them exhibited a generalised maculopapular rash. The remainder of the examination was unremarkable. A full blood count revealed anaemia in one of the subjects while three of them had evidence of leucopenia and thrombocytopenia. Two patients exhibited granulocytosis with associated lemphopenia. Three of the patients had abnormal liver function tests with elevated transaminases. Acute phase proteins were elevated in all subjects.

The history, clinical presentation and laboratory investigations raised the suspicion of possible rickettsial infection and serum was sent to the reference infectious diseases laboratory for further tests. The patients were commenced on a double antibiotic regime with Doxycycline and Penicillin or Ciprofloxacin. In all four subjects there was significant clinical improvement and the fever subsided by day five. Polymerase chain reaction (PCR) based testing confirmed a diagnosis of *Anaplasma Phagocytophilum* infection.

Conclusion: Ehrlichiosis is a relatively rare tick borne infection. However, it should always be included in the differential diagnosis of patients presenting with a non-specific febrile illness, in endemic areas.

P40

Two Cases of Visceral Leishmaniasis with Different Clinical Picture

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Aim: The discussion of a short series of patients, presenting with non-specific, flu like symptoms, secondary to infection with human Ehrlichiosis.

Cases presentation: Four patients (3 males), with a mean age of 41 years, presented to the Accident and Emergency department with persisting pyrexia. All four patients reported close contact with either cattle or mice. On clinical examination three of them exhibited a generalised maculopapular rash. The remainder of the examination was unremarkable. A full blood count revealed anaemia in one of the subjects while three of them had evidence of leucopenia and thrombocytopenia. Two patients exhibited granulocytosis with associated lemphopenia. Three of the patients had abnormal liver function tests with elevated transaminases. Acute phase proteins were elevated in all subjects.

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Conclusion: Ehrlichiosis is a relatively rare tick borne infection. However, it should always be included in the differential diagnosis of patients presenting with a non-specific febrile illness, in endemic areas.

P41

Infectious Endocarditis in an Adult Caused by *Abiotrophia Defectiva*

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Introduction: *Abiotrophia defectiva* belongs to the nutritionally defective streptococci. It causes septicemia, bacteremia and endocarditis, as Streptococci viridans.

Case Report: A 64 year-old man with a history of rheumatoid arthritis, mitral valve prolapse, thyroiditis and anemia treated with Plaquenil 1x2, T4 50 1x1 and Medrol 16 mg, presents with intermittent fever of 6-month duration. Laboratory Findings: Ht 27.9%, Hb 8.7 gr/dl, WBC 7.650 mm³, CRP 8.9 mg/dl. The blood cultures were incubated at the BacT/Alert system. During the first 24 hours, in all blood cultures a gram(+)coccus and a gram(+)coccobacillus with polymorphism and richer growth in Chocolate agar, were detected. The microorganism identified by Vitek2 (BIOMERIEUX), was *Abiotrophia defectiva*. The susceptibility testing was done with E-test, as CLSI defines for streptococci. It was sensitive to Pen, Ery, CRO, VAN. Treatment included high doses of penicilline and garmycine. 5 days later the patient underwent a myocardial infarction treated with thrombolysis. A CT was done due to focal neurological findings: an old infarct on the left and an infarct on the right side were revealed. The patient expired one day later due to cerebral hemorrhage.

Conclusion: *Abiotrophia defectiva* is a rare causative agent of bacterial endocarditis, in a percentage of 5-6%. The oral cavity is the main entrance. In relation with other streptococci, it poorly responds to antibiotic therapy, it has higher mortality, slow growth in selective culture media.

P42

Localized Infection Caused by *Pasteurella Multocida* After a Dog Bite

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Introduction: *Pasteurella multocida* is a frequent causative of localized infection after animal bites, but it rarely induces generalized manifestations.

Case Report: Male, 32 year-old presents, after being bitten by a dog, with an infected wound over the left tibia. Exudate was received for culture. A gram(-) coccobacillary bacterium was isolated, oxidase(+) and identified by API-20NE and Vitek2 (BIOMERIEUX).

The strain was sensitive to penicillin, ampicillin, ampicillin/sulbactam and aminoglycosides.

Discussion: *Pasteurella multocida* is a small, non-motile, gram(-) coccobacillary bacterium. It is an opportunistic saprophyte of the upper respiratory tract of domestic animals (cats, dogs), as well as of wild animals (lions, tigers). According to the international bibliography, 10-20% of the wounds caused by animal bites are transinfected having *Pasteurella multocida* as the most frequent agent. It is noteworthy that, although *Pasteurella multocida* is a gram(-) bacterium, penicillin remains the first choice antibiotic.

P43

Septicemia Caused by *Aeromonas Hydrophila* in an Adult Patient

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Introduction: *Aeromonas* genus includes hydrophilic Gram (-) microorganisms, pathogenic to humans causing gastroenteritis, skin and soft tissue infections, following direct contact with polluted water. Though, they can cause septicemia to immunosuppressed individuals (malignancies, pancreatic disorders, diabetes mellitus, heart failure).

Case Report: An 80 year-old man suffering from heart failure, hypertension and solitary kidney, presents with severe dyspnea, tachypnea, anuria and septic shock clinical manifestations. Lab results: WBC 11,000/mm³ (95% PMN), PLT: 66,000/mm³, Urea: 110mg/dl, Creatinine: 2.8mg/dl, CRPLX: 30.9 mg/dl, Total Bilirubin: 5.59 mg/dl (indirect: 4.90 mg/dl), Fibrinogen: 768 mg/dl, D.Dimers: 10.7α956;g/ml. Blood and urine cultures were obtained and the blood cultures were incubated in BacTAlert system(bioMerieux). During the first 24 hours the growth of a Gram (-) microorganism was detected. The identification, as well as the susceptibility testing of the microorganism were achieved by Vitek2 system(bioMerieux). In all three blood cultures *Aeromonas hydrophila* was isolated, oxidase(+), catalase(+), indole(+) and sensitive to Imipenem, Meropenem, Aminoglycosides, Trimethoprim-Sulfamethoxazole, Quinolones and 3rd-4th generation of cephalosporins.

The patient expired on the 4th day of hospitalization due to multiorgan failure.

Discussion: Septicemia due to *Aeromonas hydrophila* predominantly occurs in adults and children less than 2 year-old. Clinical symptoms include fever, chills, jaundice, dyspnoea, tachypnoea, acute renal failure and disseminated intravascular coagulation. *Aeromonas hydrophila* is isolated more frequently than other *Aeromonas* spp in septicemias with a single causative agent. Moreover, it is seldom observed in immune suppressed patients, it has a critical pathogenic action and it induces higher mortality.

P44

Septic Shock with Jaundice to Old Patient Caused by *Leptospirosis*

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Introduction: *Leptospirosis* due to *Leptospira interrogans*, a zoonosis occurring in many domestic and wild animal hosts, varies from inapparent illness to fatal disease. Human infections are acquired by direct contact with an infected animal's urine or tissue, or indirectly by contact with contaminated water or soil.

Purpose: An old man with sepsis, jaundice, anemia and fever caused by leptospira infection is presented.

Material and Methods: A 80 year-old man with free past medical history came to our hospital in stupor and signs of sepsis (B.P.: 80/50 mmHg, pulse: 110/min, T:390C). During last week he had vomiting, diarrhea and he was febrile.

Results: During the physical examination he was feeling pain when palpating his right belly also his skin and conjunctivae had icteric appearance. Blood tests count: WBC: 24.700, PLT: 24.000, CRP: 20 mg/dl, Ur: 289 mg/dl, cr: 3.5 mg/dl, Tbil: 20.78 (Dbil: 14.90) SGOT: 150, SGPT: 108, ALP: 190, α947; GT: 92, Pα932; 18, INR: 1.85, fib: 1330. He was treated with i.v. antibiotics, penicilline 2.000.000U x4 and doxycycline p.o. 100 mg x2/day. Hepatitis antibodies, Ca markers, antibodies for toxoplasma, cmv and leptospira were sent. Because of him not responding well to the therapy during the first week, and while waiting the immunology tests, Primaxin was added to his therapy (1fl x4/day) for time of 15 days. The patient was hospitalised for 21 days with periodical and slow reduction of bilirubin levels in his blood, while the improvement of his health was satisfactory. In the same time he was found positive in antibodies against leptospira (IgG: 120 normal value: <7, IgM: 47 normal value <17) therefore we concluded our diagnosis.

Conclusion: The hepatic manifestations of the disease (jaundice, hepatomegaly, right hypochondrium pain) caused due to endohepatic cholestasis. Kidney affection has to do with proteinuria, hematuria, azothemia and can be followed by tubullary necrosis and anuria. In patients with severe kidney affection, hemorrhagic manifestations are common due to mechanism of vasculitis. In our case we didnt have this kind of clinical signs. Despite of the patient's severe health condition his transfer to S.C.U. or in hemodialysis was not considered nessecary. The timely diagnosis and treatment in such kind of elder patients are definite factors regarding the outcome and normal rehabilitation of their health.