

Barišić M, Korać J, Pavlinac I, Krželj V, Marušić E, Vulliamy T, et al. Characterization of G6PD deficiency in southern Croatia: description of a new variant, G6PD Split. *J Hum Genet.* 2005;50:547-9.

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Glucose-6-phosphate dehydrogenase (G6PD) deficiency protects from severe forms of malaria. It is interesting therefore to analyze the molecular basis underlying G6PD deficiency in regions such as the Mediterranean basin where malaria was present for a long time in history. Here the authors report on the genetic characterization of G6PD deficiency among inhabitants of one Mediterranean region-the Dalmatian region of south Croatia. They analyzed 24 unrelated G6PD-deficient male subjects. Molecular testing revealed several different mutations: G6PD Cosenza 9, G6PD Mediterranean 4, G6PD Seattle 3, G6PD Union 3, and G6PD Cassano 1. Furthermore, they have identified one novel G6PD variant named G6PD Split. This variant is caused by a nucleotide change 1442 C→G leading to the amino acid substitution 481 Pro→Arg and is characterized by moderate enzyme deficiency (class III variant). This study reveals a higher prevalence (37.5%) of the Cosenza mutation in the Dalmatian region than anywhere else previously investigated and overall shows the considerable molecular heterogeneity underlining G6PD deficiency that can be observed in Mediterranean populations.

Peričić D, Lazić J, Jazvinščak Jembrek M, Švob Štrac D. Stimulation of 5-HT 1A receptors increases the seizure threshold for picrotoxin in mice. *Eur J Pharmacol.* 2005;527:105-10.

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To evaluate the possible role of 5-HT 1A and 5-HT 2A receptors in the anticonvulsant effect of swim stress, mice were pre-treated with agonists and antagonists of these receptors prior to exposure to stress and the intravenous infusion of picrotoxin. 8-OH-DPAT [(+/-)-8-hydroxy-2-(di-n-propylamino) tetralin] and WAY-100635 (a selective agonist and antagonist of 5-HT 1A receptors), DOI [1-(2,5-dimethoxy-4-iodophenyl)-2-aminopropane] and ketanserin (a 5-HT 2A/2C receptor agonist and antagonist) were used. Results demonstrated that 1 and 3 mg/kg of 8-OH-DPAT increased the doses of picrotoxin producing running/bouncing clonus, tonic hindlimb ex-

tension and death in stressed and unstressed mice, respectively. Pre-treatment with WAY (0.3 mg/kg) prevented the effect of 8-OH-DPAT (3 mg/kg). DOI (2.5 mg/kg) and ketanserin (1 mg/kg) failed to affect the seizure threshold for picrotoxin. The results show that stimulation of 5-HT 1A receptors exerts anticonvulsant actions in stressed and unstressed mice, while stimulation of 5-HT 2A/2C receptors does not interfere with the effect of stress on picrotoxin-induced convulsions.

Puljiz I, Kuzman I, Đaković-Rode O, Schönwald N, Miše B. Chlamydia pneumoniae and Mycoplasma pneumoniae pneumonia: comparison of clinical, epidemiological characteristics and laboratory profiles. *Epidemiol Infect.* 2005;29:1-8.

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The purpose of this retrospective 3-year study was to analyse and compare clinical and epidemiological characteristics in hospitalized patients older than 6 years with community-acquired pneumonia (CAP) caused by Chlamydia pneumoniae (87 patients) and Mycoplasma pneumoniae (147 patients). C. pneumoniae and M. pneumoniae infection was confirmed by serology. C. pneumoniae patients were older (42.12 vs. 24.64 years), and were less likely to have a cough, rhinitis, and hoarseness (p<0.001). C. pneumoniae patients had higher levels of C-reactive protein (CRP), and aspartate aminotransferase (AST) than M. pneumoniae patients (p<0.001). Pleural effusion was recorded more frequently in patients with M. pneumoniae (8.84 vs. 3.37%). There were no characteristic epidemiological and clinical findings that would distinguish CAP caused by M. pneumoniae from C. pneumoniae. However, some factors are indicative for C. pneumoniae such as older age, lack of cough, rhinitis, hoarseness, and higher value of CRP, and AST.

Jajić Z, Malaise M, Nekam K, Koo E, Danko K, Kovacs M, et al. Gastrointestinal safety of amtolmetin guacyl in comparison with celecoxib in patients with rheumatoid arthritis. *Clin Exp Rheumatol.* 2005;23:809-18.

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Amtolmetin guacyl (AMG), a NSAID that inhibits both COX-1 and COX-2, has an anti-inflammatory effect compa-

able to that of traditional NSAIDs, with a better GI safety profile. The primary end-point of this study was to evaluate the gastrointestinal safety of amtolmetin guacyl in comparison with celecoxib in patients affected with rheumatoid arthritis. The assessment of efficacy was the secondary end-point. This study was a 24-week, randomized, parallel group, double-blind, double dummy, multicentre trial; 235 patients were enrolled and 180 patients (85 in the AMG group and 95 in the celecoxib group) completed the study. Each patient received twice daily amtolmetin guacyl 600 mg or celecoxib 200 mg. Assessment of safety was performed by upper GI endoscopy, gastrointestinal symptoms evaluation, electrocardiography, blood and urine laboratory tests, adverse events recording. Assessment of efficacy was performed by using the American College of Rheumatology (ACR-20) responder index. Neither amtolmetin guacyl nor celecoxib determined a worsening of baseline gastro-duodenal endoscopy findings. The percentage of patients with normal findings did not significantly change after treatment with both drugs, being virtually identical with AMG (i.e. 75.29%) and increasing from 75.79% to 77.66% with celecoxib. Evaluation of the other safety parameters did not reveal any difference between the two treatment groups. Therapeutic efficacy was equivalent in both groups, with no statistical difference between the two drugs at all time intervals. In patients affected with rheumatoid arthritis, AMG and celecoxib proved to be equivalent, showing comparable gastrointestinal safety and therapeutic efficacy of treatment.

Halassy B, Matelj S, Bouche FB, Putz MM, Muller CP, Frkanec R, et al. Immunogenicity of peptides of measles virus origin and influence of adjuvants. *Vaccine*. 2006;24:185-94.

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Epitope-based peptide antigens have been under development for protection against measles virus. The immunogenicity of five peptides composed of the same B cell epitope (BCE) (H236-250 of the measles virus hemagglutinin), and different T cell epitopes of measles virus fusion protein (F421-435, F256-270, F288-302) and nucleoprotein (NP335-345) was studied in mice (subcutaneous immunisation). The adjuvant effects of peptidoglycan monomer (PGM), Montanide ISA 720 and 206 were also investigated. Results showed basic differences in peptide immunogenicity that were consistent with already described structural differences. PGM elevated peptide-specific IgG when applied together with four of five tested peptides. A strong synergistic effect was observed after co-immunisation of mice with a mixture containing all five chimeric peptides in small and equal amounts. Results revealed for the first time that immunisation with several peptides having the com-

mon BCE generated significantly higher levels of both anti-peptide and anti-BCE IgG in comparison to those obtained after immunisation with a single peptide in much higher quantity. Further improvement of immune response was obtained after incorporation of such a peptide mixture into oil-based adjuvants.

Budimir A, Deurenberg RH, Plečko V, Vink C, Kalenić S, Stobberingh EE. Molecular characterization of methicillin-resistant *Staphylococcus aureus* bloodstream isolates from Croatia. *J Antimicrob Chemother*. 2006;57:331-4.

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The objectives of this study were (i) to investigate the genetic background of methicillin-resistant *Staphylococcus aureus* (MRSA) bloodstream isolates from Croatia and (ii) to monitor the prevalence of Panton-Valentine leucocidin (PVL) and toxic shock syndrome toxin-1 (TSST-1) among these isolates. Eighty-two hospital-acquired MRSA bloodstream isolates, collected in 2001 and 2002 in Croatia, were characterized by PFGE, staphylococcal cassette chromosome mec (SCCmec) typing and multilocus sequence typing (MLST). The presence of genes encoding PVL and TSST-1 was investigated by real-time PCR. All strains were multiresistant and were distributed among 16 different similarity groups as determined by PFGE. Two of the groups, groups H and K, harboured the majority of the MRSA strains with 52 and 12%, respectively. The predominant SCCmec type found among the isolates was type I (89%). Eleven per cent of the strains harboured a modified SCCmec type III, which contained, in contrast to the regular type III, an additional dcs region. One strain harboured a novel SCCmec type, containing the ccrC gene in combination with the mecI gene, the dcs region, the locus between pI258 and Tn554 (locus E) and the locus between Tn554 and orfX (locus F). MLST showed the presence of ST111-MRSA-I and ST247-MRSA-I among Croatian MRSA isolates. All isolates were negative for both PVL and TSST-1. These results indicate the emergence of ST111-MRSA-I and ST247-MRSA-I in Croatia among MRSA bloodstream isolates. The virulence factors PVL and TSST-1 were not present among these isolates.

Hudolin T, Juretić A, Spagnoli GC, Pasini J, Bandić D, Heberer M, et al. Immunohistochemical expression of tumor antigens MAGE-A1, MAGE-A3/4, and NY-ESO-1 in cancerous and benign prostatic tissue. *Prostate*. 2006;66:13-8.

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The aim of this study was to investigate immunohistochemical expression of MAGE-A and NY-ESO-1/LAGE-1, cancer testis antigens in prostate tissues showing evidence of malignant transformation or benign hyperplasia. A total of 112 prostate samples from patients undergoing surgery at the Urology Clinic at the Zagreb Clinical Hospital Center from 1995 to 2003 were investigated. Of these, 92 carcinoma samples were obtained by radical prostatectomy, and 20 benign prostatic hyperplasia samples by transvesical prostatectomy. Three monoclonal antibodies were used for immunohistochemical staining: 77B for MAGE-A1, 57B for multi-MAGE-A and D8.38 for NY-ESO-1 expression. Expression of MAGE-A1 was observed in 10.8% of carcinoma samples, whereas multi-MAGE-A and NY-ESO-1/LAGE-1 stained 85.9% and 84.8% of samples. Immunohistochemical staining was only detectable in the cytoplasm. A significant heterogeneity could be observed within a same tissue sample where areas with strong positivities coexisted with cancer testis antigens negative areas. Interestingly, a majority of 57B positive cases were also found to be D8.38 positive (correlation coefficient $r=0.727$ ($p<0.01$)). Cancer testis antigens expression was neither significantly correlated with PSA values nor with Gleason score. In benign prostatic hyperplasia tissues MAGE-A1 expression was detected in 5%, while 57B and D8.38 staining was observed in 15% samples, and in all cases percentages of positive cells were always $<10\%$. These data underline the peculiar relevance of cancer testis antigens expression in prostate cancers, with potential implications regarding both diagnosis and therapy.

Vučković D, Abram M, Bubonja M, Wraber B, Dorić M. Host resistance to primary and secondary Campylobacter jejuni infections in C57Bl/6 mice. Microb Pathog. 2006;40:35-9.

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Campylobacter jejuni has been known as a main causative agent of human enterocolitis for more than 30 years. This has prompted the research on defence mechanisms of the host involved. Although the humoral immune response to *C. jejuni* has been addressed in many studies, relatively little

is known about the role of T lymphocytes in campylobacteriosis. The current study was based on *in vivo* T-cell subsets depletion to evaluate the role of CD4⁺ and CD8⁺ T lymphocytes in disseminated *C. jejuni* infection in C57BL/6 mice. Depletion of either CD8⁺ or CD4⁺ cells did not change the overall infection kinetics of primary campylobacteriosis. To assess the role of T cells in acquired immunity that develops during primary infection in C57BL/6 mice, *in vivo* depletions were performed during reinfection. Depletion of CD4⁺ cells did not have any effect on secondary infection kinetics, whereas depletion of CD8⁺ cells resulted in secondary liver infection that failed to resolve during the observed period. This study showed that both CD8⁺ and CD4⁺ T cells contribute to protection of C57BL/6 mice against *C. jejuni*. However, the predominant role resides in the CD8⁺ cell subpopulation. The exact mechanisms by which CD8⁺ cells operate during the course of campylobacteriosis will be the subject of our further research.

Gjergja R, Stipoljev F, Hafner T, Težak N, Lužar-Stiffler V. Knowledge and use of folic acid in Croatian pregnant women—a need for health care education initiative. Reprod Toxicol. 2006;21:16-20.

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Adequate periconceptional folic acid (FA) intake reduces the risk of neural tube defects. There are still no official FA supplementation guidelines, FA fortification policies or larger studies of awareness regarding FA or number of planned pregnancies in Croatia. This study assesses the knowledge and practice regarding FA supplementation and reports the trends in pregnancy planning in Croatia. A total of 569 pregnant women completed an anonymous questionnaire and about 72% of them were aware of the benefits of FA. Despite 75.53% of planned pregnancies, only 14.41% of all women took FA appropriately. Croatian women get information about FA from the media, health professionals and friends, but 63.77% got this information too late. The present study showed low percentage of appropriate FA intake despite high number of planned pregnancies in Croatia. It emphasizes the need for immediate and continuous public health education initiative about FA intake targeted to the women of childbearing age before their pregnancies have occurred.