CROATIAN MEDICAL JOURNAL
CMJ

43(5):628-630,2002

CROATIAN INTERNATIONAL PUBLICATIONS

Antica M, Kušić B, Hranilović D, Dietz AB, Vuk-Pavlović S. Cloning the cDNA for murine U2 snRNP-A' gene and its differential expression in lymphocyte development. Immunol Lett 2002;82:217-23.

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The authors studied genes differentially transcribed during development of murine thymocytes. By the use of differential display of mRNA by polymerase chain reaction (DD-PCR) they identified a cDNA for U2snRNP-A' from a transcript abundant in precursor thymocytes, but rare in mature T cells. The transcript was fully cloned and found to be 97% homologous to the human cDNA for U2 snRNP-A'. The authors found the gene most abundantly transcribed on day 15 of gestation and in adult prothymocytes, spleen, testis and liver. Further characterization of snRNP proteins in the mouse is warranted in an effort to establish animal models of autoimmunity relevant for studies of connective tissue diseases or systemic lupus erythematosus, where patients harbor autoantibodies reactive to snRNP.

Markotić A, Marušić A, Tomac J, Muthing J. Ganglioside expression in tissues of mice lacking beta2-microglobulin. Clin Exp Immunol 2002;128:27-35.

Department of Biochemistry, Split University School of Medicine, Split, Croatia

The study presents a comparative analysis of gangliosides from lymphoid (spleen and thymus) and other (brain, liver, lungs and muscle) tissues of C57BL/6 mice lacking the gene for beta2-microglobulin (beta2M), a constitutive component of the MHC class I molecule. Ganglioside fractions in the tissues of mice homozygous (beta2M-/-) and heterozygous (beta2M-/+) for the gene deletion were determined by high performance thin-layer chromatography (HPTLC), followed by immunostaining with specific polyclonal antibodies. Ubiquitous gangliosides GM3(Neu5Ac) and GM3(Neu5Gc) were the dominant gangliosides in the lungs of the control beta2M-/+ mice, whereas the homozygous knockout mice had substantially decreased expression of these structures. The lungs of the beta2M-/- mice also had reduced expression of T-lymphocyte-specific GM1b-type gangliosides (GM1b and GalNAc-GM1b). Beta2M-deficient mice also had more GM1a and GD1a gangliosides in the liver, and several neolacto-series gangliosides were increased in the brain and lungs. The study provides in vivo evidence that the beta2M molecule can influence the acquisition of a distinct ganglioside assembly in different mouse organs, implicating its non-immunological functions.

Bedenić B. Selection of Klebsiella pneumoniae mutants with high-level cefotaxime resistance during growth in serum containing therapeutic concentrations of cefotaxime. Chemotherapy 2002;48:10-4.

Department of Microbiology, Andrija Štampar School of Public Health, Zagreb University School of Medicine, Zagreb, Croatia

In this investigation, the author tried to develop hyperproducing variants from 8 low-level SHV-2 beta-lactamase-producing Klebsiella strains by subculturing them in serum containing therapeutic concentrations of cefotaxime (CTX). In most cases, there was a moderate increase in CTX resistance (twofold to threefold), except in one strain which displayed a 16-fold increase in the minimum inhibitory concentration (MIC) of CTX after incubation in the serum. That strain showed a marked increase in enzyme activity as well. The strains with a moderate increase in CTX MIC did not produce more enzyme after exposure to the serum, except for one strain which had a threefold rise in beta-lactamase activity after exposure to serum. In conclusion, the mutants with high-level CTX resistance developed very quickly in the biological fluids containing therapeutic concentrations of CTX. It is reasonable to expect that a similar process occurs in patients infected with an ESBL-producing K. pneumoniae strain during antibiotic treatment. Since most of the high-level CTX-resistant mutants did not have a marked rise in beta-lactamase activity after exposure to serum, it is possible that the elevated resistance was due to some other mechanism, such as reduced penicillin-binding protein affinity, changes in outer membrane proteins or efflux by multidrug efflux pumps.

Glavić Ž, Begić L, Rožman R. A new device for the detection and recognition of blood vessels in laparoscopic surgery. Surg Endosc 2002;16:1197-200.

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The endoscopic pulse detector is a new device that has been specially developed for the detection and recognition of blood vessels in laparoscopic surgery. The aim of this study was to investigate the performance and possibilities of the new instrument. The instrument's performance and features were tested in laboratory conditions by use of simulated circulation. The authors assessed the dependence of the signal amplitude recorded by the instrument over a given blood vessel on the blood pump frequency change (36-130/min), circulating systolic pressure change (40-180 mm Hg), and adjacent blood vessel pulsations. Clinically, the instrument was tested in a randomized study in 40 elective noncomplicated laparoscopic cholecystectomies, where the authors assessed the time needed for the preparation of Calot's triangle and positioning of the cystic artery. The results of laboratory testing showed that the instrument operated throughout the frequency range of 36-130/min and a circulating systolic pressure range of 40-180 mmHg, while the signal amplitude rose with pressure increase. The results of clinical testing showed that use of the pulse detector in laparoscopic cholecystectomy significantly reduced the time needed to prepare Calot's triangle ($t = 3.91$; $df = 38$; $p < 0.001$) and also made the positioning of the cystic artery more reliable. In conclusion, the study showed the new instrument to be very simple to use and potentially valuable in laparoscopic surgery, primarily for the differentiation of blood vessels of similar structures.

Crljen V, Volinia S, Banfić H. Hepatocyte growth factor activates phosphoinositide 3-kinase C2 beta in renal brush-border plasma membranes. Biochem J 2002;1;365(Pt 3):791-9.

Department of Physiology and Croatian Institute for Brain Research, Zagreb University School of Medicine, Zagreb, Croatia

Upon stimulation of renal cortical slices with hepatocyte growth factor (HGF), inositol lipid metabolism was studied in basal-lateral plasma membranes (BLM) and brush-border

plasma membranes (BBM). Whereas in BLM rapid increases in 1,2-diacylglycerol, PtdIns(3,4,5)P(3) and PtdIns(3,4)P(2) were observed, suggesting that in BLM HGF activates both phospholipase C (PLC) and phosphoinositide 3-kinase (PI3K), in BBM only HGF-induced transient accumulation of PtdIns3P was seen, which was temporarily delayed from signalling events in BLM and could be blocked by the PtdIns-specific-PLC inhibitor ET-18-OCH(3) and the calpain inhibitor calpeptin, suggesting that 3-kinase activation in BBM lies downstream of PLC activation in BLM and is a calpain-mediated event. Moreover, the increase in immunoprecipitable PI3K-C2 beta activity, which is sensitive to wortmannin (10 nM) and shows strong preference for PtdIns over PtdIns4P as a substrate, was observed only in BBM upon stimulation of renal cortical slices with HGF and could be mimicked by the Ca^{2+} ionophore A23187 and blocked by the cell-penetrant Ca^{2+} chelator BAPTA-AM [1,2-bis-(2-aminophenoxy)ethane-N,N,N′,N′-tetra-acetic acid tetrakis(acetoxymethyl ester)]. On Western blots PI3K-C2 beta revealed a single immunoreactive band of 180 kDa in BLM and BBM, while after stimulation with HGF a gel shift of 18 kDa was noticed only in BBM, suggesting that the observed enzyme activation is achieved by proteolysis. When BBM were subjected to short-term (15 min) exposure to mu-calpain, a similar gel shift together with an increase in PI3K-C2 beta activity was observed, when compared with the BBM harvested after HGF stimulation. The above-mentioned gel shift and increase in PI3K-C2 beta activity could be prevented by the calpain inhibitor calpeptin. The data presented in this report show that in renal cells there is a spatial separation of the inositol lipid signalling system between BLM and BBM, and that HGF causes activation of PLC and PI3K primarily in BLM, which leads to calpain-mediated activation of PI3K-C2 beta in BBM with a concomitant increase in PtdIns3P.

Hećimović S, Klepac N, Vlašić J, Vojta A, Janko D, Škarpa-Prpić I, et al. Genetic background of Huntington disease in Croatia: molecular analysis of CAG, CCG, and Delta2642 (E2642del) polymorphisms. Hum Mutat 2002;20:233.

Division of Molecular Medicine, Ruđer Bošković Institute, Zagreb, Croatia

This study presents the first molecular data on the basis and the origin of Huntington disease in Croatia and is the first such analysis performed among a Slavic population. The authors analyzed three trinucleotide polymorphisms in the HD gene: CAG, CCG and GAG Delta2642 (E2642del) triplets. Analysis of the CAG repeat size among 44 Huntington patients (39-66 CAGs) and 51 normal individuals (9-34 CAGs) showed that the range of the repeats was similar to previous findings. The frequency of the CCG and Delta2642 polymorphic alleles on N and HD chromosomes was found to correlate well with earlier reports for Western European populations. The authors found significance for both the CCG7 allele ($p = 0.004$) and the Delta2642 allele ($p < 0.001$) among HD chromosomes. The CCG7 allele was overpresented among affected chromosomes (94.6%), but was also the most frequent CCG allele among normal chromosomes (66.7%). Interestingly, the Delta2642 allele was present on 40.5% HD chromosomes compared to only 9.8% of control chromosomes. The results indicate that HD mutations in Croatia could be of the same origin as in Western populations and also support the multi-step hypothesis for generating new HD alleles. Similar frequencies and distributions of both the CCG and the Delta2642 polymorphisms in Croatia and Western European normal chromosomes indicate that the prevalence rate of HD in Croatia may be as high as in Western populations. Since the authors estimated a lower prevalence rate (1:100,000), they assume that there are still many misdiagnosed and/or unrecognized cases of Huntington disease in Croatia.

Gršković B, Ferenčak G, Rukavina AS, Karija M, Furač I, Kubat M. Mutation analysis of the MPZ and PMP22 genes in Croatian patients. Clin Chem Lab Med 2002;40:559-62.

Clinical Institute of Laboratory Diagnostics, Zagreb University School of Medicine and Clinical Hospital Center Zagreb, Zagreb, Croatia

The authors used single-strand conformation polymorphism analysis for mutational screening in two candidate genes, MPZ and PMP22, which have an important role in the pathogenesis of Charcot-Marie-Tooth disease (CMT) and related peripheral neuropathies. A novel Ser8Ser polymorphism was found in exon 1 of the MPZ gene in two heterozygous subjects, in a father with mild CMT2 phenotype and his daughter with normal clinical data. Thr118Met polymorphism was found in exon 5 of the PMP22 gene. The patient heterozygous for 118Met allele had CMT1 disease. It is concluded that the occurrence of the 118Met allele does not usually cause CMT1 and that it is not a clinically relevant disease marker.

Rožgaj R, Kasuba V, Šimić D. The frequency of dicentrics and acentrics and the incidence of rogue cells in radiation workers. Mutagenesis 2002;17:135-9.

Mutagenesis Unit, Institute for Medical Research and Occupational Health, Zagreb, Croatia

Occupational exposure to ionizing radiation causes chromosomal damage. Some of the damaged cells show a large number of aberrations such as dicentrics, polycentrics, rings and numerous acentric fragments. This paper describes an analysis of the frequency of dicentric chromosomes and acentric fragments in 1,260 subjects occupationally exposed to X-rays and 241 controls. Special attention was paid to the incidence of multi-aberrant cells. The 3 year cumulative dose was a significant predictor for all analyzed aberrations. The duration of exposure was a highly significant predictor of the frequency of rogue cells, but not of acentrics and dicentrics. Age and sex were not found to be significant predictors of the analyzed aberrations.

Mulić R, Ropac BD, Zorić I, Bradarić N. Epidemiologic and ecologic characteristics of some diseases transmitted by arthropods on the littoral of the Republic of Croatia. Mil Med 2002;167:321-5.

Naval Medicine Institute, Croatian Navy, Split, Croatia

The objective of this study was to show epidemiologic and ecologic characteristics of some diseases transmitted by arthropods on the littoral of the Republic of Croatia. The pattern of infectious diseases that are transmitted exclusively by vectors was monitored during the period 1985 to 1999. Data on the incidence of the diseases in the Republic of Croatia as a whole and available data on the presence of the vectors for the diseases on the littoral are presented for comparison. Mediterranean spotted fever, murine typhus, Lyme disease, cutaneous and visceral leishmaniasis, and pappataci fever occur sporadically on the Croatian littoral. Tick-borne meningoencephalitis is endemic in the northwestern part of Croatia but is not present on the littoral. Twelve cases of malaria are imported into Croatia per year on average. Diseases transmitted by arthropods are not a major public health problem in Croatia. The medically relevant entomofauna of Croatia has not yet been adequately investigated.

Turk Z, Šesto M, Skodlar J, Ferenčak G, Turk N, Stavljenić-Rukavina A. Soluble LDL-immune complexes in type 2 diabetes and vascular disease. Horm Metab Res 2002;34:196-201.

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The objective of our study was to analyse markers of LDL oxidation - soluble LDL containing immune complexes (LDL-ICs) in type 2 diabetes with micro- and macrovascular disease. The study included 69 diabetic patients with coronary artery disease (DM + CAD), 78 non-diabetics with CAD, 47 controls, and 27 diabetics with nephropathy and 36 free from complications.

OxLDL antibodies and advanced glycated end-products were measured by ELISA, and LDL-IC apo B content after PEG precipitation. Determination of a broad range of oxLDL antibody activity in all study groups showed no significant differences. In contrast, the content of apo B, a component of the antigen moiety of oxLDL-ICs, was higher in CAD and diabetes (+CAD) than in LDL-ICs isolated from controls ($p < 0.001$). LDL-ICs did not differ between patients with CAD+DM and CAD patients free from diabetes. LDL-IC levels in diabetic patients with or without microangiopathy were significantly higher than in healthy volunteers. However, there was no significant difference in the level of circulating LDL-ICs between the subgroup of diabetic patients with nephropathy/retinopathy and patients free of microvascular disease. There was a statistically significant positive correlation between AGE content and LDL-ICs ($r = 0.35$, $p < 0.009$). A significant but inverse correlation was recorded between triglyceride concentration and level of LDL-ICs in DM+CAD ($r = -0.32$, $p < 0.016$) and CAD patients ($r = -0.35$, $p < 0.002$). A highly significant negative correlation between triglycerides and circulating LDL-ICs ($r = -0.54$, $p < 0.039$) was observed in patients with early nephropathy, but not in those with physiological proteinuria. In conclusion, the increased level of circulating LDL-ICs is a risk factor for the general population, including those with diabetes. The results of this study suggested the contribution of LDL-ICs to the development of atherosclerosis to probably be more significant than the direct contribution of oxLDLAb itself.

Čulić O, Eraković V, Čepelak I, Barišić K, Brajša K, Ferenčić Ž, et al. Azithromycin modulates neutrophil function and circulating inflammatory mediators in healthy human subjects. Eur J Pharmacol 2002;450:277-89.

PLIVA d.d., Research Division, Zagreb, Croatia

Effects on human neutrophils and circulating inflammatory mediators were studied in 12 volunteers who received azithromycin (500 mg/day, p.o.) for 3 days. Blood was taken 1 h before treatment, 2.5, 24 h and 28 days after the last dose. An initial neutrophil degranulating effect of azithromycin was reflected in rapid decreases in azurophilic granule enzyme activities in cells and corresponding increases in serum. The oxidative response to a particulate stimulus was also acutely enhanced. These actions were associated with high plasma and neutrophil drug concentrations. A continuous fall in chemokine and interleukin-6 serum concentrations, within the non-pathological range, accompanied a delayed down-regulation of the oxidative burst and an increase in apoptosis of neutrophils up to 28 days after the last azithromycin dose. Neutrophils isolated from blood at this time point still contained detectable drug concentrations. Acute neutrophil stimulation could facilitate antibacterial effects of azithromycin, while delayed, potentially anti-inflammatory activity may curtail deleterious inflammation.

Ćelić R, Jerolimov V. Association of horizontal and vertical overlap with prevalence of temporomandibular disorders. J Oral Rehabil 2002;29:588-93.

Department of Prosthodontics, Zagreb University School of Dental Medicine, Zagreb, Croatia.

The aim of the study was to evaluate the relationship between horizontal (HO) and vertical overlap (VO) and prevalence of temporomandibular disorders (TMD) in a non-patient population. Out of the total number of subjects (230), 65.7% subjects were classified as asymptomatic, 5.7% subjects had muscle disorders (MD) (myalgia), 9.1% subjects had disc displacement with reduction (DDR), while 19.6% subjects had disc displacement with reduction associated with muscle disorder (DDR+MD) (myalgia). The HO equal to or greater than 5 mm was significantly more prevalent in the symptomatic patients (MD, DDR, DDR+MD) than in the asymptomatic group ($p < 0.001$). The VO equal to or >5 mm also demonstrated statistically significant difference ($p < 0.006$) between the symptomatic (suffering from DDR, and DDR+MD) and asymptomatic subjects. Conclusion of the study is that the HO and VO equal to or >5 mm was more present in the group of subjects with diagnosis of a TMD (DDR and MD), what distinguishes this group from the group of asymptomatic subjects.

Bosnar A, Stemberg V, Zamolo G, Štifter S. Increased suicide rate before and during the war in southwestern Croatia. Arch Med Res 2002;33:301-4.

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This paper analyzes the impact of war, with special regard for the intensity of changes in the suicide rate in wartime in the southwestern region of Croatia. The investigated region has an area of 7,993 km^2 with a population of 322,964. Most of the population lives in the city of Rijeka, the regional center. During the 10-year period from 1986-1995, a total of 4,172 deaths were investigated at the Institute of Forensic Medicine in Rijeka. Official reports based on forensic autopsies performed on the corpses note 602 cases of suicide, which have been confirmed by police investigation and forensic examination. As the investigated 10-year period encompasses two distinctly specific intervals – the peacetime 1986-1990 interval and wartime 1991-1995 – the differences in respective quantitative and qualitative features of suicides during these two intervals were analyzed. A total of 262 suicides were committed in peacetime at the rate of 16.22/100,000 inhabitants, as opposed to 340 suicides in wartime at a rate of 19.61/100,000 inhabitants. This increase in the wartime suicide rate was primarily due to the 83% increase in the number of suicide victims <40 years of age. The number of suicides by firearms in wartime was almost fourfold the number in peacetime. It can be concluded that wartime conditions had a direct impact on the change of quantitative and qualitative features of suicides.

Kaliterna L, Larsen ZP, Brkljačić T. Chronological and subjective age in relation to work demands: survey of Croatian workers. Exp Aging Res 2002;28:39-49.

Ivo Pilar Institute of Social Sciences, Zagreb, Croatia

The aim of the study was to investigate chronological and subjective age and gender differences in terms of the physical, mental, and social demands of work. Subjects, 88 women and 25 men, were workers in a medical setting who completed the Survey of Health Care Professionals. Subjects' reports of effort required by their work, tiredness resulting from their work, and personal skills needed to complete their work were used to assess the physical, mental, and social demands of work. Subjective age measures included cognitive age, desired age, and comparative age. Results showed that, compared to men, women reported being more tense as a result of the work. Age differences were found in the physical and mental skills required by the work. Older workers had lower estimates of their own skills than younger workers. Three measures of subjective age showed different patterns of relationships with aspects of work demands.

Grubić Z, Stingl K, Kaštelan A. Determination of polymorphism at eight STR loci in the Croatian population using automated detection. Forensic Sci Int 2002;127(1-2):147-9.

National Referral Organ Transplantation and Tissue Typing Center, University Hospital Center Zagreb, Zagreb, Croatia

Population study was carried out on the sample of 167 unrelated donors from the wider area of the Croatia's capital, Zagreb, using the short tandem reapet (STR) loci: TH01, VWFA31, FES/FPS, F13A01, D1S1656, D12S391, D18S535 and D22S683.