EDITORIAL

A COMPARATIVE STUDY OF BIPOLAR DISORDER AND ATTENTION DEFICIT HYPERACTIVITY DISORDER THROUGH THE MEASUREMENT OF REGIONAL CEREBRAL BLOOD FLOW

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Attention Deficit Hyperactivity Disorder (ADHD) and Bipolar Disorder (BPD) are two common neuropsychological disorders which are often present in a comorbid state. I used the results of cerebral blood flow studies made with Single Photon Emission Computer Tomography (SPECT), Positron Emission Tomography (PET) and Functional Magnetic Resonance Imaging (fMRI), to investigate a possible relationship between ADHD and BPD. The common areas of the brain involved in both BPD and ADHD appears to be the prefrontal cortex in its various components, the basal ganglia and possibly the cerebellum which, especially in the past, has been little studied by researchers in relation to ADHD and BPD. Among the differences the blood flow lateralization, present in BPD in states of altered mood, is evident with left hypoperfusion and right hyperperfusion during depression, the opposite in the case of manic state; in ADHD, the lateralization is less constant and of questionable interpretation. In BPD the involvement of a greater number of brain areas, especially the temporal lobe, is common. I advance the hypothesis that BPD progresses from ADHD secondary to expansion of perturbation in cerebral blood flow.

EDITORIAL

IL-36 A NEW MEMBER OF THE IL-1 FAMILY CYTOKINES

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Interleukin-36 (IL-36) is a pro-inflammatory cytokine which plays an important role in innate and adaptive immunity. IL-36 activates MAPK and NF-KB pathways and is produced by many different cells. This cytokine is a family member of interleukin-1 (IL-1) and plays an important role in the pathophysiology of several diseases. Here we summarise and review the new aspects of this important pro-inflammatory cytokine.
TREADMILL EXERCISE IMPROVED ADRIAMYCIN-INDUCED NEPHROPATHY

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Adriamycin nephropathy (AN) or doxorubicin-induced chronic kidney disease (DRCKD) has several strengths as an experimental model of renal diseases involving glomerulosclerosis, tubulointerstitial inflammation and fibrosis. Exercise has shown to be beneficial to many chronic diseases. We hypothesize that treadmill exercise may improve AN, and an investigation was carried out with the AN SD rat model. Treadmill exercise was conducted three times per week, each time for 30 and 60 min. DR induced swelling of glomeruli, collagen deposition in the interstitium and renal cortex, and increased the serum levels of MDA, IL-6, PDGF-BB, MMP-2, MMP-9, TGF-β, p-PDGF, uric acid, serum cholesterol, triglycerides, BUN, creatinine, blood platelet count, ratio of kidney to body weight, glomerular volume, and urinary BUN and protein. Conversely, levels of serum SOD, TNF-α, p-PI3K, p-Akt, albumin, WBC, RBC, and urinary creatinine were decreased. Treadmill exercise ameliorated most of these damaging effects, better outcome was found for the 60-min exercise training. Conclusively, the endurance exercise is more associated with the normalization of signaling expressions involving TGF-β, PDGF-BB, p-PDGF, p-PI3K, and p-Akt, which may help CKD patients to restore cell survival, proliferation, and growth. As rehabilitation is a personalized medicine, an appropriate design to fit individual feasibility has to be well figured out.

EARLY ANGIOGENIC RESPONSE TO SHOCK WAVES IN A THREE-DIMENSIONAL MODEL OF HUMAN MICROVASCULAR ENDOTHELIAL CELL CULTURE (HMEC-1)

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The exact nature of shock wave (SW) action is not, as yet, fully understood, although a possible hypothesis may be that shock waves induce neoangiogenesis. To test this hypothesis, a three-dimensional (3D) culture model on Matrigel was developed employing a human microvascular endothelial cell line (HMEC-1) which was stimulated with low energy soft-focused SW generated by an SW lithotripter. After 12 hours we observed a statistically significant increase in capillary connections subsequent to shock-wave treatment in respect to the control group and a marked 3-hour down-regulation in genes involved in the apoptotic processes (BAX, BCL2LI, GADD45A, PRKCA), in cell cycle (CDKN2C, CEBPB, HK2, IRF1, PRKCA), oncogenes (JUN, WNT1), cell adhesion (ICAM-1), and proteolytic systems (CTSD, KLK2, MMP10). Our preliminary results indicate that microvascular endothelial cells in vitro quickly respond to SW, proliferating and forming vessel-like structures, depending on the energy level employed and the number of shocks released. The early decreased expression in the analysed genes could be interpreted as the “first reactive response” of the endothelial cells to the external stimuli and the prelude to the events characterizing the neo-angiogenic sequence.
IN VITRO EXPOSURE OF HUMAN OSTEOARTHRITIC CHONDROCYTES TO ELF FIELDS AND NEW THERAPEUTIC APPLICATION OF MUSICALLY MODULATED ELECTROMAGNETIC FIELDS: BIOLOGICAL EVIDENCE

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Osteoarthritis (OA) is the most frequently occurring rheumatic disease, caused by metabolic changes in chondrocytes, the cells that maintain cartilage. Treatment with electromagnetic fields (MF) produces benefits in patients affected by this pathology. Isolated human osteoarthritic (OA) chondrocytes were cultured in vitro under standard conditions or stimulated with IL-1ß or IGF-1, to mimic the imbalance between chondroformation and chondroresorption processes observed in OA cartilage in vivo. The cells were exposed for a specific time to extremely low frequency (ELF; 100-Hz) electromagnetic fields and to the Therapeutic Application of Musically Modulated Electromagnetic Fields (TAMMEF), which are characterized by variable frequencies, intensities, and waveforms. Using flow cytometry, we tested the effects of the different types of exposure on chondrocyte metabolism. The exposure of the cells to both systems enhances cell proliferation, does not generate reactive oxygen species, does not cause glutathione depletion or changes in mitochondrial transmembrane potential and does not induce apoptosis. This study presents scientific support to the fact that MF could influence OA chondrocytes from different points of view (viability, ROS production and apoptosis). We can conclude that both ELF and TAMMEF systems could be recommended for OA therapy and represent a valid non-pharmacological approach to the treatment of this pathology.
Tissue Distribution and Metabolism of Guanosine in Rats Following Intraperitoneal Injection

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Guanosine has long been known as an endogenous purine nucleoside deeply involved in the modulation of several intracellular processes, especially G-protein activity. More recently, it has been reported to act as an extracellular signaling molecule released from neurons and, more markedly, from astrocytes either in basal conditions or after different kinds of stimulation including hypoxia. Moreover, in vivo studies have shown that guanosine plays an important role as both a neuroprotective and neurotrophic agent in the central nervous system. Specific high-affinity binding sites for this nucleoside have been found on membrane preparations from rat brain. The present study was undertaken to investigate the distribution and metabolic profiles of guanosine after administering the nucleoside to gain a better understanding of the biological effects of this potential drug candidate. Rats were given an intraperitoneal (i.p.) injection of 2, 4, 8 or 16 mg/kg of guanosine combined with 0.05% of [3H]guanosine. Plasma samples were collected 7.5, 15, 30, 60 and 90 min after the guanosine-mixture administration and analyzed by either a liquid scintillation counter or by HPLC connected to a UV and to an on-line radiochemical detector to measure the levels of guanosine and its metabolic products guanine, xanthine and uric acid. The levels of guanosine, guanine and xanthine were also measured in brain, lung, heart, kidney and liver tissue homogenates at the defined time points after the injection of 8 mg/kg of the guanosine-mixture. We found that the levels of radioactivity in plasma increased linearly in a dose- and time-dependent manner. Guanosine was widely distributed in all tissues examined in the present study, at almost twice its usual levels. In addition, guanine levels dramatically increased in all the organs. Interestingly, enzymatic analysis of the plasma samples showed the presence of a soluble purine nucleoside phosphorylase, a key enzyme in the purine salvage pathway and nucleoside catabolism. Since guanosine has been shown to be neuroprotective and astrocytes have been reported to play critical roles in mediating neuronal survival and functions in different neurodegenerative disorders, we also performed uptake and release
Peripheral arterial disease (PAD) is a chronic condition caused by atherosclerosis and a severe complication of type 2 diabetes (T2D). We hypothesised that chronic condition of arterial disease engenders inflammation and endothelial damage in response to circulating cytokines released in the blood stream of PAD patients. We explored the levels of circulating cytokines in PAD patients with and without diabetes by multiplex cytokine array compared with non-PAD controls. Serum from PAD patients with or without diabetes showed high levels of VEGF, IFN-γ, TNF-α, MCP-1, and EGF. VEGF levels correlated with TNF-α and IFN-γ, significantly. Endothelial cells (ECs) were exposed to the different altered cytokines to evaluate changes in cell growth, migration and tubule-like formation, displaying impairment on proliferation, migration and tubule formation. Our findings demonstrate that a set of cytokines is significantly increased in the serum of PAD patients. These cytokines act to induce endothelial dysfunction synergistically. VEGF strongly correlated with TNF-α and IFN-γ, opening new therapeutic perspectives.
CONCOMITANT EVALUATION OF FLOW-MEDIATED VASODILATION AND EPICARDIAL FAT THICKNESS IN IDIOPATHIC DEEP VENOUS THROMBOSIS

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Flow mediated vasodilation (FMD) evaluates the endothelium-dependent vasodilation, is a reliable marker of arterial endothelial dysfunction and is related to coronary artery disease. Visceral fat predicts an unfavorable cardiovascular and metabolic risk profile in humans and echocardiographic assessment of epicardial fat (EF) is a reliable marker of visceral adiposity. We measured the FMD and EF thickness in 77 subjects, 38 without idiopathic deep vein thrombosis (DVT) (mean age 65.95±16.29 years) and 39 with idiopathic DVT (mean age 65.49±17.22 years). The purpose of this work is to investigate the presence of statistical association between FMD and DVT and between EF thickness and DVT. Furthermore, to account for possible atherosclerosis risk factor unbalances, comparison between FMD and DVT (and between EF and DVT) was assessed using a multivariate logistic regression model which included the following covariates: FMD, EF, age, sex, smoking and the presence of obesity. Subjects without DVT showed significant lower values of EF thickness (9.07±1.89 mm vs 12.32±1.73 mm, p=0.005) and borderline-significant greater values of FMD (9.01±2.77% vs 7.47±5.37%, p=0.058) as compared to those with DVT. In conclusion, the data presented indicate that subjects affected by spontaneous deep vein thrombosis may have an impaired endothelium-dependent vasodilation, a marker of arterial endothelial dysfunction related to coronary artery disease, and an increased epicardial adipose tissue, a marker of cardiometabolic risk.

COMPARISON OF THE PRIMARY STABILITIES OF CONICAL AND CYLINDRICAL ENDOSEOUS DENTAL IMPLANTS: AN IN-VITRO STUDY

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The aim of this study is to determine the differences in primary stability between conical and cylindrical dental implants. The insertion and removal torques were the parameters used to measure the primary stability of the implants. Ten conical and cylindrical dental implants were positioned in polyurethane foam blocks to simulate bone density classes D1, D2, D3 and D4. The insertion and removal torques were quantified using a digital torque gauge. The maximum insertion torque and the maximum removal torque measured for the D1 and D4 synthetic bone were significantly higher for the conical implants than the cylindrical implants. In this in-vitro model, conical implants show significantly higher primary stability than cylindrical implants for the D1 and D4 synthetic bone classes.
Saporin-S6 is a single-chain ribosome-inactivating protein (RIP) that has low toxicity in cells and animals. When the protein is bound to a carrier that facilitates cellular uptake, the protein becomes highly and selectively toxic to the cellular target of the carrier. Thus, saporin-S6 is one of the most widely used RIPs in the preparation of immunoconjugates for anti-cancer therapy. The endocytosis of saporin-S6 by the neoplastic HeLa cells and the subsequent intracellular trafficking were investigated by confocal microscopy that utilises indirect immunofluorescence analysis and transmission electron microscopy that utilises a direct assay with gold-conjugated saporin-S6 and an indirect immunoelectron microscopy assay. Our results indicate that saporin-S6 was taken up by cells mainly through receptor-independent endocytosis. Confocal microscopy analysis showed around 30% co-localisation of saporin-S6 with the endosomal compartment and less than 10% co-localisation with the Golgi apparatus. The pathway identified by the immunofluorescence assay and transmission electron microscopy displayed a progressive accumulation of saporin-S6 in perinuclear vesicular structures. The main findings of this work are the following: i) the nuclear localisation of saporin-S6 and ii) the presence of DNA gaps resulting from abasic sites in HeLa nuclei after intoxication with saporin-S6.
Circadian and seasonal rhythms in daylight affect many physiological processes. In the eye, energy of intense visible light not only initiates a well-studied neural reaction in the retina that modulates the secretory function of the hypothalamus and pineal gland, but also activates the heme oxygenase (HO) to produce carbon monoxide (CO). This study was designed to determine whether the concentration of carbon monoxide (CO) in the ophthalmic venous blood changes depending on the phase of the day and differing extremely light intensity seasons: summer and winter. The concentration of CO in the venous blood flowing out from the nasal cavity, where heme oxygenase (HO) is expressed, but no photoreceptors, was used as a control. Sixteen mature males of a wild boar and pig crossbreed were used for this study. Samples of ophthalmic and nasal venous blood and systemic arterial and venous blood were collected repeatedly for two consecutive days during the longest days of the summer and the shortest days of the winter. The concentrations of CO in blood samples was measured using a standard addition method. During the longest days of the summer the concentration of CO in ophthalmic venous blood averaged 3.32±0.71 and 3.43±0.8 nmol/ml in the morning and afternoon, respectively, and was significantly higher than in the night averaging 0.89±0.12 nmol/ml (p<0.001). During the shortest day of the winter CO concentration in ophthalmic venous blood was 1.11±0.10 and 1.13±0.14 nmol/ml during the light and nocturnal phase, respectively, and did not differ between phases, but was lower than in the light phase of the summer (p<0.01). The CO concentration in the control nasal venous blood did not differ between seasons and day phases and was lower than in ophthalmic venous blood during the summer (p<0.01) and winter (p<0.05). The results indicate that the gaseous messenger carbon monoxide is released from the eye into the ophthalmic venous blood depending on the intensity of sunlight.
SKELETAL MODIFICATIONS IN MUCOPOLYSACCHARIDOSES: AN OVERVIEW

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The mucopolysaccharidoses (MPS) are a group of rare diseases characterized by deficiencies in different enzymes required for degradation of complex carbohydrates. The enzymatic deficiencies lead to lysosomal accumulation of dermatan sulphate, heparan sulphate, and keratan sulphate in different tissue resulting in multi-system complications. Six different principal types are described. Most MPS types, with the exception of MPS III, are associated with widespread skeletal abnormalities and joint disease. Authors analyzed clinical pathological and radiographical features of mucopolissaccharidoses focusing on pelvic and spine pathologies that generally limit activity and normal life so they have to be treated at the beginning of their manifestations in order to avoid major complication and improve quality of life.

ADVERSE REACTION TO IRRIGATION WITH POVIDONE-IODINE AFTER DEEP-IMPACTED, LOWER THIRD MOLAR EXTRACTION

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Povidone-iodine is most commonly used worldwide because of its germicidal activity, relatively low irritancy or toxicity and low cost. Frequently, povidone-iodine is used as a topical antiseptic for treating and preventing wound infection. In rare cases skin irritation or iododerma-like eruption could represent possible adverse effects due to the oxidative effects of iodine and allergic hypersensitivity reaction. In this report we describe a case of a massive adverse reaction to the irrigation of surgical wound dehiscence with 10% povidone-iodine solution after deep-impacted, lower third molar extraction. This reaction was related to a central neurotrophic reflex involving three trigeminal branches and probably due to a peripheral chemical insult of mandible nerve. This adverse reaction determined a severe edema and diffuse skin lesions, involving the whole left side of the face mimicking an iododerma-like eruption. These violent symptoms were solved after 60 days. Furthermore, we report a small permanent skin scar in the zygomatic area and transient alterations of facial sensitivity on the affected side which completely disappeared in 6 months.
PLATELET-RICH PLASMA IMPROVES WOUND HEALING IN MULTIPLE MYELOMA BISPHOSPHONATE-ASSOCIATED OSTEONECROSIS OF THE JAW PATIENTS

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Bisphosphonates are drugs used to treat various metabolic and malignant bone diseases. In the past 10 years intravenous bisphosphonates have been associated with increased risk of osteonecrosis of the jaw (ONJ). The aim of the present study is to evaluate platelet-rich plasma (PRP) wound healing benefits in multiple myeloma (MM) patients who developed ONJ after surgical tooth extraction. The study included 7 patients, 2 males and 5 females. All individuals had been taking zoledronate or pamidronate followed by zoledronate for an average of 5 years. Four subjects had only standard surgical debridement and sequestrectomy to treat the ONJ and three had additional autologous PRP. The patients were followed-up for 3 months. The use of PRP to enhance wound healing and reduce bone exposure seems to be a good treatment protocol in ONJ MM subjects.

SCLEROSTIN CONCENTRATIONS IN ATHLETES: ROLE OF LOAD AND GENDER

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Bone mass is the net product of formation and resorption, which are closely regulated by the equilibrium between endogenous/exogenous factors. Sclerostin inhibits the Wnt canonical signaling and is considered an anti-anabolic factor. We compared sclerostin serum concentrations between genders in athletes belonging to different sport disciplines, characterized by a different weight-bearing, and in their sedentary counterparts in order to study the possible link between bone metabolism in athletes and its peripheral concentration. We also compared sclerostin levels with bone alkaline phosphatase activity, a marker of bone formation. Sixty-one elite athletes, belonging to weight-bearing (15 male rugby players, 11 male enduro racers, 8 female basketball players), high-impact (6 male tennis players, 8 female ice skaters), non weight-bearing sports (13 male cyclists) and 16 sedentary controls were enrolled. Higher levels of sclerostin were found in females. Sclerostin was higher in weight-bearing than in non-weight-bearing disciplines in males. Significant inverse age-related correlation was found. Higher bone alkaline phosphatase activity was observed in females. The young adult elite athlete represents a peculiar physiologic model for studying sclerostin behavior: the applied load increased the marker concentrations, testifying a high bone turnover rate; however, a gender effect is evident.