WHY ARE NEUTROPHILS POLYMORPHONUCLEAR?

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Most cells in the human body have a spherical or ovoid nucleus. The mature human neutrophil, unlike most other cells, exhibits a distinctly non-spherical nucleus, which is segmented into three to five lobes. The possible mechanisms underlying this segmented nuclear shape have been explored. The structure of the nuclear envelope, composition of lamins and lamin-B receptor seems to have an important role in shaping the nucleus. Being the first line of defense, neutrophils migrate rapidly to the site of infection and destroy the invading pathogen. This requires negotiation through narrow capillaries, transmigration across the vessel wall and passage through tight tissue spaces. Segmented shape confers increased nuclear flexibility, thereby easing the migration of neutrophils through narrow channels. The segmented shape of the nucleus may also play a role in intranuclear chromatin organization and gene expression. The unique shape of the neutrophil nucleus seems to be an adaptation to facilitate its function.
EDITORIAL

INFLAMMATORY MARKERS: SERUM AMYLOID A, FIBRINOGEN AND C-REACTIVE PROTEIN – A REVISITED STUDY

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The acute phase response is the part of the innate defence system of an animal against trauma, inflammation or infection. During this response, there is increased production and release of certain plasma proteins known as acute phase proteins, which include C-reactive protein (CRP), serum amyloid A (SAA) and fibrinogen (Fg). CRP consists of five identical subunits of 206 amino acids with a molecular weight of approximately 23 kDa. There is strong evidence from numerous studies that CRP is a predictor of inflammation. The acute-phase protein serum amyloid A (SAA) is a clinically useful marker of inflammation. SAA plays not only an important role in the development of AA amyloidosis (an important complication of rheumatoid arthritis) but also interacts with events closely involved in the metabolic syndrome as a high- and low-grade inflammatory modulator. Fibrinogen (Fg) is a high molecular weight plasma adhesion protein and is a biomarker of inflammation. It is synthesized and assembled in hepatocytes and fibroblasts and when secreted into the circulation, its plasma half-life ranges from 3 to 4 days. Several cytokines, are involved in the induction of acute phase protein synthesis, but the mutual importance of these cytokines seems to be cell-type specific and to vary in various experimental settings. Here we revisited the classic acute phase proteins SAA, C-Reactive protein and fibrinogen in their role in inflammation and their interrelationship with some cytokines.
EDITORIAL

WHAT YOU SHOULD KNOW ABOUT ESCHERICHIA COLI INFECTION

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As in Northern Germany there have recently been several deaths caused by *Escherichia coli* (*E. coli*), we decided to revisit the effects of *E. coli* infection. Since this bacteria is the most numerous facultative and aerobic germ in the human intestine, we would like to warn the population of its pathogenicity. In fact, *E. coli* can be pathogenic both in humans and in animals and can start an inflammatory process, activating some factors of the cell nucleus such as NFkB, with the consequent production of cytokines. *E. coli* can appear in several strains and can be very aggressive and can contaminate food, water and the environment, causing severe disease, and in some cases death.
IN VIVO EFFECTS OF A GINKGO BILOBA EXTRACT ON PLATELET ACTIVATING FACTOR METABOLISM IN TWO ASYMPTOMATIC HIV-INFECTED PATIENTS

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Ginkgo biloba products seem to protect from several pathological conditions, including HIV manifestations, where Platelet Activating Factor (PAF) is implicated. In the present study, we examined for the first time the in vivo effects of a standardized formulation of Ginkgo biloba extract (150 mg daily, per os) on PAF metabolism in blood cells and plasma of two male, asymptomatic HIV-infected patients, not receiving antiretroviral treatment, during a 9-month period. These patients differed at baseline in terms of duration of HIV infection, viral load levels, CD4 cell counts and Highly Active Antiretroviral Therapy (HAART) experience. In the first patient with early HIV infection, after an initial transient increase, a return of both plasma viral load and PAF biosynthetic enzyme activities in leukocytes to their baseline levels was observed during Ginkgo biloba use. As a result PAF degradation also remained low in this patient. The second patient with late but not advanced HIV infection, had higher levels of viral load and a lower CD4 cell count at baseline. The use of 150 mg of a Ginkgo biloba extract was probably insufficient to induce PAF degradation and/or to suppress the induction of PAF biosynthesis observed. At the same time, the initial high levels of viral load were further increased and CD4 cell counts were finally decreased during the study. The observed differences in PAF metabolism during Ginkgo use seem to be related to the initial heterogeneity of these patients. It appears that in some HIV-infected patients inhibition of the PAF/PAF-receptor system, along with a decrease/down-regulation of PAF-biosynthesis, illustrates a new potential role for Ginkgo biloba compounds in the treatment of HIV infection and its manifestations. However, more tests on a larger number of patients are needed in order to support these preliminary observations.
IN VITRO STUDIES OF THE IMMUNOMODULATORY EFFECTS OF STATINS ALONE AND IN COMBINATION WITH IMMUNOSUPPRESSIVE DRUGS

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The effects of statins go beyond their lipid-lowering properties and include immunomodulatory and anti-inflammatory effects. Unfortunately, there is a lack of in vitro assays that study the immunomodulatory effect of statins at therapeutic concentrations and the possible synergism with immunosuppressive drugs. Besides, they are mostly evaluated on isolated peripheral blood mononuclear cells instead of using whole blood as a matrix. The aim of this study is to perform in vitro experiments to evaluate the effect of atorvastatin, simvastatin and fluvastatin at therapeutic concentrations alone and in combination with everolimus or tacrolimus on immunosuppressive response, using whole blood as a matrix by investigating lymphocyte proliferation and production of the soluble cytokines interleukin (IL)-2, IL-10 and interferon (IFN)-γ. Statins (0.1 μM) inhibited T cell proliferation by 12-16% in a dose-dependent manner and when statins at 0.1 μM were combined with each immunosuppressive drug at 8 ng/ml, inhibition increased by 6-9% (p<0.05) for everolimus and 8-15% (p<0.05) for tacrolimus, but not for atorvastatin. At a dose of 0.1 μM, all three statins inhibited soluble IFN-γ production by approximately 5-9% (p<0.02). IL-2 and IL-10 production were unaltered by the presence of statins. These findings suggest that statins seem to exert a mild anti-inflammatory effect that might potentially be used to treat autoimmune diseases.
MIF EXPRESSION IN INDUCED PERIPHERAL BLOOD MONONUCLEAR CELLS BY VITAMIN D3 AND ITS POTENTIAL CORRELATION WITH RESTING METABOLIC RATE IN OBESITY

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Macrophage migration inhibitory factor (MIF) plays a pivotal role in systemic and local inflammatory and immune responses. The aim of this study is to assess MIF gene expression in PBMCs harvested from obese individuals and to compare it with that of lean subjects to analyze any potential relationship between resting metabolic rate as well as several different parameters and MIF expression in PBMC. We also aim to assess anti-inflammatory characteristics of vitamin D3 on MIF expression in vitro. Participants were 30 obese and 18 non-obese subjects who were assessed following an overnight fasting for RMR by means of indirect calorimetry. Body composition was measured using a Bodystat device. The PBMCs were separated from whole blood by the Ficoll-hypaque technique. The mRNA was extracted and the cDNA was synthesized. This process was followed by real-time PCR using primer pairs specific for MIF mRNA and beta actin as internal control. Our findings clearly demonstrate that there were significant differences in terms of BMI, BMR predict, fat proportion, fat mass, free fat mass, TBW, visceral fat, fasting serum glucose, TG, HDL, Hs-CRP and RMR between the two groups. Moreover, the level of MIF expression in the obese group was approximately 2.5 times higher compared to the lean group. An increased level of MIF expression in the obese group and a decreased expression of that non-obese was observed after inducing PBMCs with vitamin D3. One of the intriguing results of this study was the observed reverse correlation between MIF expression and fat mass as well as fat proportion after PBMCs were cultured in the presence of vitamin D3. Therefore, it could be concluded that MIF expression, which is in turn influenced by vitamin D3, has a role in the hyperactive immune profile and the pro-inflammatory state observed in obese individuals which is suggested to have a causal relationship with obesity.
THE EFFECTS OF INTERFERON-α2b ON INTESTINAL FLORA IN PERITONEAL FIBROSIS

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Peritonitis is an important cause of the mortality and morbidity of peritoneal dialysis patients. The present study aims to investigate the effects of IFN-α2b on intestinal flora in peritoneal fibrosis. Twenty-four Wistar albino rats were divided into three groups. The control group received 0.9% saline (3 ml/d) intraperitoneally; the chlorhexidine gluconate (CH) group received 3 ml daily injections of 0.1% CH intraperitoneally; the CH+IFN group received 3 ml daily injections of 0.1% CH intraperitoneally and pegylated IFN-α2b 1.5 µg/kg per week subcutaneously on days 0, 7, 14. On the twenty-first day rats were sacrificed and visceral peritoneum samples were obtained from the liver. Blood samples were obtained from the abdominal aorta and intestinal flora samples were obtained from distal small intestine and transverse colon. Histopathologic control of CH, CH+IFN groups peritoneal thickness were 6.04±2.32, 135.4±22.24, and 42.56±11.6 µm, respectively. The decrease in thickness of parietal peritoneum in the CH+IFN group was statistically significant when compared to the CH group. Escherechia coli (E. Coli) had grown in cultures of the small intestine and colon samples of all the rats in the control group, whereas Proteus species (spp) had grown in one and Enterobacter spp. in seven cultures of the CH group. E. Coli had grown in four cultures, Proteus spp in three culture and Enterobacter spp. in one culture obtained from small intestine and transverse colon of the control group. The intestinal flora changed as the peritoneal thickness increased. The intestinal flora in the CH group completely changed compared with the control group (p<0.001). There was no correlation between visceral peritoneal thickness and intestinal flora change in the IFN group (p>0.05). IFN-α2b recovers the intestinal flora and the intestinal motility, thus reducing the experimental peritoneal fibrosis.
EVALUATION OF NERIDRONATE ON THE OSSEOINTEGRATION PROCESS OF ENDEOUS TITANIUM IMPLANTS IN ANIMAL MODELS

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Bisphosphonates are compounds that inhibit bone reabsorption mediated by osteoclasts. The use of bisphosphonates in oral implantology is still in the experimental stage. The aim of this study is to evaluate the efficacy of an aminobisphosphonate to increase the ability of the drug to act on the implant and bone surfaces in the development of the osseointegration in sheep. Forty SLA titanium implants were used on sheep iliac crests. Neridronate added to connective gel (test 1) or to physiological solution (test 2) was used in order to increase the bone and implant adhesiveness. Physiological solution (control 1) or connective gel (control 2) alone was given to the control groups. A topical administration of Neridronate was made on the implant surface and in the implant site. Four Bergamasca sheep were used and were sacrificed by intravenous injection of 10 cc Tanax after 8 weeks from implantation. Histologic and histomorphometric analyses were carried out. The results did not show significant differences between the test group and control group. Our data are different from other similar studies obtaining statistically significant differences. These differences could depend on the procedure of application of the drug on the implant. This study demonstrates the poor efficacy of neridronate applied topically to the implant and implant site during surgery. Further studies using different fixation techniques of the drug may be necessary to confirm the present data.
SERUM INTERLEUKIN-18 IN CHILDREN WITH HENOCH-SCHÖNLEIN PURPURA: A PROMISING MARKER OF DISEASE ACTIVITY?

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Henoch-Schönlein purpura (HSp) is the most common systemic vasculitis of childhood with typical skin involvement and concurrent signs involving joints, gastrointestinal tract, and kidney. HSp pathogenesis is still far from being completely understood, though a knotty cytokine complex is believed to contribute to its intimate processes. The aim of our evaluation is to establish the relationship between serum levels of interleukin (IL)-18 and disease outcome and establish its feasibility to provide a marker of disease activity or even a prognostic tool in clinical practice. We examined clinical/laboratory variables and serum IL-18 in 17 children hospitalized during a year for HSp, diagnosed by EULAR/PRINTO/PRES criteria; the same patients were re-evaluated after 6 months. All results were compared with 25 age-matched healthy controls. IL-12 and IL-6 were also evaluated in a cohort of the same patients and compared with controls. General and clinical variables (sex, edema of the extremities, gastrointestinal or renal complications, relapses and renal involvement at 6 months) had no relationship with cytokine levels. Serum IL-18 and IL-6 levels were found significantly increased at diagnosis in HSp patients when compared with healthy controls. After 6 months, serum IL-18 and IL-12 levels were significantly decreased in patients, while IL-12 and IL-6 levels were significantly increased compared to healthy controls. Though preliminary and expecting further confirmation on a larger sample, our data support the conclusion that serum IL-18 levels reflect HSp activity.
APOCYNIN, A PLANT-DERIVED DRUG, MIGHT BE USEFUL IN THE TREATMENT OF MYOCARDIAL ISCHEMIA REPERFUSION INJURY IN RAT HEARTS

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Apocynin is a potent and selective inhibitor of the NADPH oxidase-dependent production of ROS by stimulated human PMNs. Apocynin was isolated by activity-guided isolation from Picrorhiza kurroa, and in the years following its discovery it has been used in many laboratories around the world. Reactive oxygen species (ROS) production by activated polymorphonuclear neutrophils (PMNs) plays an important role in many cardiovascular disease states, including myocardial ischemia reperfusion injury. The purpose of this study is to examine the beneficial effects of apocynin on myocardial ischemia reperfusion injury. Myocardial I/R injury was caused by clamping the left anterior descending (LAD) coronary artery for 20 min followed by release of the clamp allowing reperfusion for 1 h. Administration of apocynin i.p. (5mg/kg i.p. 10%DMSO) 15 min after ischemia significantly reduced: 1) histological evidence of myocardial injury; 2) pro-inflammatory cytokines (TNF-α, IL-1β); 3) adhesion molecules (ICAM-1, P-Selectin); 4) nitrotyrosine formation; 5) NF-kB expression; 6) PAR formation; and 7) apoptosis (Bax, Bcl-2, Fas-L and tunel). Based on these findings we propose that apocynin would be useful in the treatment of various ischemia and reperfusion diseases.
THE USE OF TOPICAL HYALURONIC ACID AND SILVER SULFADIAZINE (ALTERGEN®) IN PATIENTS UNDERGOING TWO-STAGE ANTERIOR URETHROPLASTY WITH ORAL MUCOSAL GRAFT

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This investigative pilot study was conducted with the objective of evaluating the therapeutic effect of a cream containing hyaluronic acid (HA) and silver-sulfadiazine (Altergen®, IBSA Farmaceutici Italia, S.r.l.) in adult patients undergoing two-stage urethroplasty using an oral mucosal graft for anterior urethral strictures due to failed hypospadias repair. We used a cream (Altergen®) containing the following active ingredients: 100 g HA, 0.20 mg sodium salt, and 1 g silver-sulfadiazine. During the period of January 2009 – May 2010, 40 patients, mean age 24 years, who had undergone hypospadias surgery during childhood were enrolled in the study, according to some inclusion and exclusion criteria (age >18 years, with the absence of diabetes, neurovascular disease, lichen sclerosus or other dermatological lesions). All 40 patients had navicularis or penile urethral strictures and underwent two-stage urethroplasty using an oral mucosa graft. Post-operatively, 20 patients (50%) were treated with a cream containing HA and silver-sulfadiazine (Altergen®) and 20 patients (50%) were treated using a standard topical treatment (iodopovidone 10% gel). All patients underwent a follow-up at 6, 15, 30, 60 and 120 days after surgery. Out of the 20 patients treated with the HA and silver-sulfadiazine-containing cream (Altergen®), 18 (90%) showed good and complete healing of the graft implant 30 days after surgery. Of the 20 patients not treated with the HA and silver-sulfadiazine-containing cream (Altergen®), 13 (65%) showed good and complete healing of the graft implant 30 days after surgery. These results were constant in all follow-ups in both patient groups. In patients who underwent a navicularis or penile two-stage oral mucosal graft urethroplasty, the use of a HA and silver-sulfadiazine-containing cream was associated with a higher success rate of the graft implant and a lower incidence of post-surgical complications compared to the controls.
INFLAMMATORY AND IMMUNITARY MODIFICATIONS IN SALIVA OF SUBJECTS WITH LABIAL AND TONGUE PIERCING

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The first two authors contributed equally to this work

Piercing is the practice of puncturing some parts of the body, including the area of the stomach, to apply ornamental objects. The presence of oral and perioral piercings is a risk factor for numerous acute and chronic complications, such as chipping of the dental enamel, lesions of the gums and infection. The presence of piercings in the oral cavity may also act as a stimulant for inflammation and determine modifications in the components of the saliva. The aim of this study is to evaluate whether there is a variation in the inflammatory or immunitary components of the saliva of adult patients with labial and tongue piercings. Twenty-five adult patients were examined (11 males and 14 females with an average age of 23.4±3.6 years) who had had a minimum of one labial or tongue piercing for at least 1 year. A questionnaire was compiled for each patient and the composition of the saliva was examined, before and for the 72 hours after removal of the piercings. The data was analyzed using the student’s t-test for independent samples and the level of significance was placed at p< 0.05. The examination of the saliva showed a statistically significant increase (p< 0.05) of interleukin-1 and -8, of lysozyme and amylase. A more basic value of pH (p< 0.05) was also found and a decrease in the immunoglobulins, in particular of IgA (p< 0.05). After the removal of the piercings, the parameters of the saliva returned to the reference values, with the exception of the immunoglobulins, whose values remained the same as in the first saliva examination. The presence of piercings in labial areas or in the tongue is accompanied by chronic inflammation in the components of the saliva, with an increase in the level of the interleukins. Furthermore, in patients who have had an oral piercing for a longer period, the level of interleukins is even higher. This inflammatory situation, however, may recede in a few days after the removal of the irritating agent. Labial or tongue piercings also determine an increase in the flow of saliva, an increase in saliva enzymes, of lysozyme in particular, and a more basic pH value.
LETTER TO THE EDITOR

TANDEM REPEATS OF THE CATT ELEMENT OF MACROPHAGE MIGRATION INHIBITORY FACTOR GENE MAY PREDICT GESTATIONAL DIABETES MELLITUS SEVERITY

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Previous investigations have demonstrated the association of the CATT repeated allele in the Macrophage migration inhibitory factor (MIF) with obesity and diabetes. Since there are common risk factors and molecular pathways in Type 2 Diabetes and Gestational Diabetes Mellitus (GDM), we focused on the associations of MIF variation with GDM. In a case-control study we enrolled 157 GDM and 217 healthy pregnant women, referred to the outpatient clinic of Shariati Hospital. The different repeats of CATT in MIF promoter were determined. MIF relative gene expression was evaluated in Peripheral blood mononuclear cells of all the participants. The GDM group had higher mean age and pre-gravid BMI. Also fasting serum glucose, insulin and MIF gene expression were significantly higher in the GDM patients. The statistically significant difference was observed between GDM and a healthy group in carrying 7-CATT allele and MIF gene expression. Regarding GDM risk factors, MIF 7-CATT allele showed significant relation with pre-pregnancy obesity, as well as the need for insulin therapy. Our results indicate that an association between MIF genotypes and its expression with GDM, obesity and the need of insulin for management of GDM patients exists.
LETTER TO THE EDITOR

PREDICTION OF ALLERGY BY TOTAL SERUM IgE MEASUREMENTS IN INFANCY: A 10-YEAR FOLLOW-UP

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Asthma, allergic rhinitis and atopic dermatitis are multifactorial disorders influenced by various familial and environmental factors. Cord blood IgE levels may be predictive for subsequent allergy onset. Serum IgE assessment has been rarely investigated. This study is aimed at evaluating serum IgE levels during the first year of life and relating to subsequent allergies. Total serum IgE levels were assessed in 102 newborns in the first days of life (2-3) and then again at 3, 5, 6, 11 and 12 months of age. After ten years, paediatricians and general practitioners caring for these children were tracked through the Local Healthcare Agency (LHA) database and asked about possible allergy of their patients, including transient ones. Serum IgE increased in allergic infants at 1 year of life. The predictive model was significant. In conclusion, this study demonstrates that serum IgE may be predictive of subsequent allergy onset, preferably if assessed at 1 year of age.
LETTER TO THE EDITOR

CANCER IN ITALIAN PATIENTS WITH SYSTEMIC SCLEROSIS

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The association between cancer and systemic sclerosis (SSc) is known, although the underlying mechanisms remain unclear and epidemiological data is conflicting. Since no data exist on cancer in Italian SSc, we examined the frequency and characteristics of cancer in an Italian cohort of SSc patients to examine whether clinical and/or laboratory SSc-specific features represent a risk for developing malignancies in these patients. A retrospective chart review was carried out of 112 Italian SSc patients of whom 109 were women and were men, aged 63±13 years; 81 patients had limited SSc, 25 had diffuse SSc and 6 had sine scleroderma SSc. Fifteen cancers were found in 14 patients. The majority (60%) occurred after SSc onset (average 16 years), 40% occurred before the onset of SSc (average 14 years). The most frequent was breast cancer (prevalence: 4.5%, relative prevalence: 33.3%), followed by uterine cancer and lymphomas (prevalence: 2.7%, relative prevalence: 20% each). Lung cancer was not observed. Cancers were unrelated with SSc type, autoantibodies, organ involvement and treatments. In conclusion, clinical features do not seem to be linked with the risk of developing cancer in SSc patients. Interestingly, and in contrast with published data, no lung cancer was present in our patients, although lung involvement was observed in the majority of them. This finding, consistent with a lower prevalence of lung cancer in the Italian female general population, and the absence of associations between SSc-specific features and cancer, suggests that genetic and environmental factors might play a pivotal role in cancer risk in these patients.
LETTER TO THE EDITOR

SERUM IgE FOR Bet v 1 IN PATIENTS AFFECTED BY ATOPIC DERMATITIS

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Atopic dermatitis (AD) is a common allergic disorder and may be associated with respiratory allergy. In this study serum levels of IgE specific for the major allergen of birch (Bet v 1) were measured in two groups of AD patients with specific IgE to birch: the first with associated respiratory birch allergy and the second with cutaneous symptoms alone. The results show that the patients suffering from AD associated with respiratory allergy had the highest serum levels of IgE specific for Bet v 1. In conclusion, this study shows that the measurement of serum specific IgE for a major allergen may be useful in common practice as high levels are associated with complicated pathology.