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Prevention of Renal Fibrin Deposition in Endotoxin-induced DIC through Inhibition of PAI- 1

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Plasminogen activator inhibitor-1 (PAI-1) increases in endotoxemia thus possibly cooperating in altering the hemostatic balance in a prothrombotic direction. The effect of the inhibition of PAI-1 with the monoclonal antibody MA-33B8 was studied systemically and in kidneys in a lapine model of endotoxin-induced disseminated intravascular coagulation (DIC). The increase in plasmatic PAL activity in the control group (n = 9) was inhibited in the MA-33B8 treated rabbits (n = 5). Control rabbits showed renal fibrin deposits, whereas only one of the MA-33B8 rabbits did so. These results were confirmed immunohistochemically in kidneys as PAI-1 immunostaining was seen inside the glomeruli and larger vessels in the control group, whereas MA-33B8 rabbits showed a remarkable decrease, demonstrating that MA-33B8 successfully inhibited PAI-1 in the kidneys as well. Therefore evidence for the important role of PAI-1 in fibrin generation in endotoxin-induced DIC is presented, suggesting that strategies aiming at its reduction can be useful in this pathology (9), there have been no attempts so far to study the effect of the inhibition of PAI activity, which is dramatically increased after a continuous infusion of gram negative endotoxin [lipopolysaccharide (LPS)] at a dose sufficient to induce disseminated intravascular coagulation (DIC) (10-16). MA-33B8 is a PAI-1-neutralizing monoclonal antibody (Mo Ab) raised against human PAI-1 and crossreacting with, as well as exerting a strong neutralizing activity against, rabbit PAI-1 (17). The PAI-1 neutralizing effect of MA-33B8 was shown to be associated with an accelerated conversion to the latent conformation (18, 19). In this study we investigated the effect of MA-33B8 in a well defined model of LPS-induced DIC (14-16), in which the Mo Ab is administered to rabbits receiving a continuous intravenous infusion of F. coli LPS. As kidneys are a target organ for the formation of fibrin polymers in the microvasculature in DIC and previous studies have postulated a role for PAI-1 in the renal pathology and also in the generation of fibrin deposits (20, 21), special attention is paid to the effect of treatment with MA-33B8 on PAI-1 activity in the renal microcirculation and its relationship with the formation of renal fibrin deposits.

Key words: PAI-1. Sepsis. Endotoxemia. Monoclonal antibodies. Kidney.

The nervous system of the chicken proventriculus: an immunocytochemical and ultrastructural study

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The proventriculus constitutes the glandular region of the chicken stomach. This organ is innervated by two parasympathetic networks, the myenteric and submucous plexus, and here we present a systematic study of this system by immunohistochemistry and electron microscopy. All the neurons and fibres were positive for the neural markers, protein gene product 9.5 and the amidating enzymes. Immunoreactivities for the constitutive neuronal isoform of the enzyme nitric oxide synthase and the vasoactive intestinal peptide were present in neuronal bodies suggesting an intrinsic origin for the similarly immunoreactive fibres found in the proventriculus. On the other hand. immunoreactivity to gastric inhibitory peptide was only found in varicose fibres making contact with the blood vessels and the glandular epithelium, but never in the neuronal somas, suggesting that this substance may be provided by an extrinsic nervous system whose neuronal bodies are located elsewhere. Electron microscopy revealed frequent neuromuscular and neuroepithelial connections in the muscle layers, the wall of the blood vessels and the epithelium. In addition, synapsis-like structures were identified in the proximity of cells belonging to the diffuse endocrine system. providing a new example of neuroendocrine contacts. No positivity was found for antibodies against other neural substances including somatostatin. peptide histidine-isoleucine. peptide tyrosine-tyrosine, neuropeptide tyrosine, bombesin, met-enkephalin, serotonin. substance P, galanin, calcitonin gene-related peptide and S-100 protein.

Haemostatic changes in systemic inflammatory response syndrome during continuous renal replacement therapy

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Abstract: Background. Endothelial damage and hemostatic imbalance play an important role in the evolution of the Systemic Inflammatory Response Syndrome (SIRS) into the Multiple Organ Dysfunction Syndrome (MODS). In Acute Renal Failure associated with SIRS, different types of Continuous Renal Replacement Therapies (CRRT) may give non-renal benefits by modifying the levels of some factors related to those disturbances.

Methods: Forty patients with SIRS-associated ARF were randomised to receive either continuous venovenous hemofiltration (CVVH) or continuous venovenous hemodiafiltration (CVVHDF) for the first 24 h. Afterwards the CRRT method was reversed. The group treated with CVVH moved to CVVHDF and that treated with CVVHDF to CVVH for the next 24 h. Plasma levels of: von Willebrand Factor (vWF), thrombomodulin, plasminogen activity inhibitor type 1 (PAI-1: antigen and activity), tissue type plasminogen activator (t-PA: antigen), prothrombin fragment 1+2 (F1+2) and thrombin-antithrombin complexes (TAT) were measured previously to CRRT (base-line), and after 24 and 48 hours of therapy. Multivariate ANOVA was the statistical method used.

Results: In the MANOVA study a significant decrease in PAI-1 activity during the treatment procedure was observed (horizontality $p < 0.05$).PAI-1 antigen showed a tendency to decrease although without statististical significance. There were no significantly different changes in the other factors analysed during either CRRT (parallelism $p > 0,05$). At the base-line point, all the factors were higher than normal values in healthy adults.

Conclusions: The present study suggests that CRRT, in patients with SIRS, may promote a decrease in PAI-1 and consequently, a better outcome. There were no differences between the CVVH and the CVVHDF methods regarding the factors analysed. The present data confirms that there is an important endothelial and hemostatic dysfunction in SIRS from the early phases.

Key words: Systemic inflammatary response Syndrome (SIRS). Acute renalfailure (ARF). Continuous renal replacement therapy (CRRT). Endothelial injury. Haemostatic disturbances. Plasminogen activity inhibitor type 1 (PAI-1)

Hemostatic Disturbances in Patients With Systemic Inflammatory Response Syndrome (SIRS) and Associated Acute Renal Failure (ARF)

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Endothelial damage plays a central role in the development of an SIRS-related Multiple Organ Dysfunction Syndrome (MODS) as a consequence of the establishment of a hemostatic imbalance between coagulation and fibrinolysis systems. Until now, sepsis is the SIRS model that has been most studied. The aim of this study was to assess the endothelial damage and the hemostatic imbalance in early stages of an SIRS of different origins, and to study if there are any differences in these disturbances between infectious and noninfectious SIRS. The endothelial damage and hemostatic changes were studied in 40 patients with SIRS (with less than 12 h of evolution) and an acute renal failure. Infectious SIRS was diagnosed in 19 cases and non-infectious SIRS in the remaining 21 patients. Patients with SIRS presented significantly higher values ($p < 0.001$) for factors related to endothelial damage [von Willebrand factor (vWF), thrombomodulin, tissue plasminogen activator (t-PA), and plasminogen activator inhibitor type 1 (PAI-1) antigen], hypercoagulability [prothrombin fragment 1 + 2 (FI + 2) and thrombin—antithrombin complexes (TAT)], and fibrinolysis (D-dimer and PAI activity) with respect to the control group. However, although the group with infectious SIRS presented higher values for all the factors except for the t-PA and D-dimer with respect to SIRS of other origins, none of these differences reached statistical significance ($p > 0,05$). Our data show that patients with SIRS and associated acute renal failure, irrespective of the origin (infectious or noninfectious), show signs of intense endothelial damage and bypercoagulability throughout the process.

Key words: Systemic Inflammatory Response Syndrome. Acute renal failure. Sepsis. Hemostatic disturbances. Endothelial damage.

Colocalization of numerous immunoreactivities in endocrine cells of the chicken proventriculus at hatching

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The colocalization of regulatory peptide immunoreactivities in endocrine cells of the chicken proventriculus at hatching has been investigated using the avidin-biotin technique in serial sections and double immunofluorescence in the same section for light microscopy, and double immunogold staining for electron microscopy. In addition to the eight immunoreactivities

previously described in this organ, cells immunoreactive for peptide histidine isoleucine (PHI), peptide gene product 9.5 (PGP). and the amidating enzyme, peptidylglycine α-amidating monooxygenase (PAM) were observed. All the cells immunoreactive to glucagon were also immunostained by the PHI antiserum. In addition, all the glucagon-like peptide 1, avian pancreatic polypeptide, and some of the neurotensin-like cells costored also glucagon- and PHI-immunoreactive substances. PGP- and PAM-immunoreactivities were also found in the glucagon-positive cells. A small proportion of the somatostatin-containing cells were positive for PHI but not for other regulatory peptides. These results could suggest either the existence of a very complex regulatory system or that the endocrine system of the newborn chickens is not yet fully developed.

The relationship between glycogen synthesis, biofilm formation and virulence in Salmonella enteritidis

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Salmonella enteritidis accumulated large quantities of intracellular polysaccharide when grown in unrestricted nutrient conditions. Dense, abundant cytoplasmic granules were observed by electron microscopy in sections stained by the periodic acid-chlorite technique, indicating that the polysaccharide was of the glycogen type. When biofilm-producing *S. enteritidis* was pre-incubated in media containing increasing levels of glucose concentration, the levels of both cytoplasmic glycogen and biofilm rose correlatively to a point where a ceiling effect was observed. Studies carried out with activators and inhibitors of glycogen biosynthesis confirmed that biofilm was formed from glycogen cell stores. On the other hand, the virulence of the biofilm-producing strain in infected chickens increased proportionally to the amount of stored glycogen, suggesting a possible role of the glycogen depot in the virulence of *S. enteritidis.*

Gentamicin encapsulation in PLA/PLGA microsphere in view of treating Brucella infections

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In view of treating intracellular Brucella infections, microspheres made of poly(lactide) (PLA) and poly(lactide-co-glycolide) (PLGA) were developed as delivery system for the cationic and highly hydrophilic antibiotic gentamicin sulphate. Drug microencapsulation by spray drying yielded microspheres with regular morphology, an average particle size of approximately 3 μm and encapsulation efficiencies of up to 45%. Different copolymers of similar molecular weights gave varying encapsulation efficiencies and particle size distributions. The encapsulation efficiency generally increased with polymer hydrophilicity, except for the hydrophilic copolymer PLGA5O:50H carrying carboxylic end groups. Encapsulation also depended on the pH value of the aqueous drug solution to be encapsulated. Moreover, increasing nominal gentamicin sulphate loading yielded lower efficiencies. For comparison, some formulations were also prepared by a (W_1/O)W 2-solvent evaporation method, which yielded lower encapsulation efficiencies, in the order of 13%. Finally, drug bioactivity was found to remain intact after microencapsulation, MS storage and MS incubation in aqueous medium. The results suggest that PLA/PLGA microspheres prepared by spray drying may be an appropriate delivery system for gentamicin sulphate to be used in the treatment of intracellular Brucella infections.

Key words: Gentamicin sulphate. Poly(lactide). Palv(lactide-co-glycolide). Microspheres. Brucellosis. Intracelullar infections.

Brucella abortus and Its Closest Phylogenetic Relative, Ochrobactrum spp., Differ in Outer Membrane Permeability and Cationic Peptide Resistance

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The outer membrane (OM) of the intracellular parasite *Brucella abortus* is permeable to hydrophobic probes and resistant to destabilization by polycationic peptides and EDTA. The significance of these unusual properties was investigated in a

comparative study with the opportunistic pathogens of the genus *Ochrobactrum*, the closest known *Brucella* relative. *Ochrobactrum* spp. OMs were impermeable to hydrophobic probes and sensitive to polymyxin B but resistant to EDTA. These properties were traced to lipopolysaccharide (LPS) because (i) insertion of B. *abortus* LPS, but not of *Escherichia coli* LPS, into *Ochrobactrum* OM increased its permeability; (ii) permeability and polymyxin B binding measured with LPS aggregates paralleled the results with live bacteria; and (iii) the predicted intermediate results were obtained with B. *abortus-Ochrobactrum anthropi* and *E. coli-O. anthropi* LPS hybrid aggregates. Although *Ochrobactrum* was sensitive to polymyxin, self-promoted uptake and bacterial lysis occurred without OM morphological changes, suggesting an unusual OM structural rigidity. *Ochrobactrum* and B. *abortus* LPSs showed no differences in phosphate, qualitative fatty acid composition, or acyl chain fluidity. However, *Ochrobactrum* LPS, but not B. *abortus* LPS, contained galacturonic acid. B. *abortus* and *Ochrobactrum* smooth LPS aggregates had similar size and zeta potential (-12 to -15 mV). Upon saturation with polymyxin, zeta potential became positive (1 mV) for *Ochrobactrum* smooth LPS while remaining negative (-5 mV) for B. *abortus* smooth LPS, suggesting hindered access to inner targets. These results show that although *Ochrobactrum* and *Brucella* share a basic OM pattern, subtle modifications in LPS core cause markedly different OM properties, possibly reflecting the adaptive evolution of B. *abortus* to pathogenicity.

Adenocarcinoma vesical primario: nuestra experiencia en los últimos diez años

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Los adenocarcinomas vesicales son tumores vesicales raros, que generalmente corresponden a lesiones metastásicas y con menor frecuencia a tumores vesicales primarios. Presentamos los adenocarcinomas primarios tratados en nuestro hospital en los últimos 10 años. El pronóstico de estos tumores está en relación con el momento del diagnóstico ya que su crecimiento intramural provoca que el diagnóstico se realice en estadios avanzados de la enfermedad. El tratamiento suele consistir en cirugía radical, a la que es necesario añadir otros tratamientos complementarios en casos de recidiva.

Palabras clave: Adenocarcicoma vesical. Uracal. No uracal.

Key words: Bladder adenocarcicoma. Urachal. No urachal.

First Break of Mania Associated with Topiramate Treatment

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Preliminary open-label data suggest the possible efficacy of the new anticonvulsant, topiramate (TPM) in the treatment of bipolar disorder.[1] Its mechanism of action involves the potentiation of -aminobutyric acid responses, inhibition of excitatory pathways through an action at the kainate aminomethyl phosphonic acid subtype of glutamate receptors, and voltage-dependent sodium channel blockade[2,3]. TPM also inhibits some carbonic anhydrase isoenzymes; however, this action may be related to side effects and not to anticonvulsant activity[4]. Reported treatment-emergent adverse events included dizziness, somnolence, ataxia, headaches, paresthesia, nystagmus, dysarthria, cognitive disfunction, visual disturbance, confusion, nervousness, mood lability, weight loss, and renal stones[5]. We report the emergence of manic symptoms associated with TPM treatment in a woman with a history of major depression but no prior reported history of mania.

Variations of serum eosinophil cationic protein and tryptase, measured in serum and saliva, during the course of immediate allergic reactions to foods

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Abstract of:
Allergy 2001: 56: 568-572

Background: Subjective complaints and reactions after placebo administration during food challenges (FC) may make their outcome difficult to interpret. We determined serum ECP and tryptase as tryptase in saliva during FC, looking for markers to support challenge outcomes.

Methods: Twelve patients with systemic reactions after food intake and nine presenting oral allergy syndrome (OAS) underwent skin tests; total and specific IgE determinatiom double-blind, placebo-controlled FC (DBPCFC); and open challenges.

Blood samples were collected before and 1, 2, and 5 h after challenge and saliva before and 5, 30, and 60 mm after challenge. ECP and tryptase were quantified by ImmunoCAP

(Pharmacia-Upjohn. Sweden). Serum tryptase of >10 μg/l was considered positive.

Results: After positive DBPCFC (n = 8), ECP rose significantly (P < 0.05) at 1-h -16.03 (12.8) μg/l (mean [standard deviation]) -and 2-h intervals -17.56 (10.7) μg/l -compared to basal level of 9 (6.4) μg/l. After negative DBPCFC (n = 6), ECP increased from basal 9.63 (3.9) μg/1 to 24.84 (14.17) μg/l at the 2-h time point. There were nonsignificant differences in ECP between patients with positive and negative FC. Two patients with positive challenge showed a tryptase level of > 10 μg/l and only one patient with OAS showed 5.6 μg/l of txyptase 5 mm after FC.

Conclusions: ECP and tryptase in serum and saliva were not useful markers for FC outcomes.

Key words: Eosinophil cationic protein (ECP). Food allergy. Food challenges. Oral allergy syndrome. Tryptase.

Differences in interleukins' patterns between dysthymia and major depression

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We assessed whether cytokine production-interleukin (IL)-1, IL-β and tumour necrosis factor α (TNα)-is affected in depressed patients, dysthymia (Dt) and major depression (MD), and its association with various parameters of severity and clinical course. We found a possible different pattern of interleukin production between Dt and MD.

Key words: interleukin-1β. Interleukin-6. Cytokine I dysthymia. Major depression. Monocyte.

Allergic vulvovaginitis in infancy: study of a case

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Abstract of:
Allergol et Immunopathol 2001;29(4):137-140.

Background: the role of dust mites (Dermatophagoides pt.) in the pathogenesis of allergic vulvovaginitsis is still controversial. Association between this mite and atopic dermatitis, conjunctivitis, rhinitis or asthma is already known.

Some authors study the possible relationship between some vulvovaginitis and local hypersensitivity. The aim of this study was to corroborate the allergic aetiology due to the mite Dermatophagoides pt. in a girl with vulvovaginitis and perennial rhinitis.

Methods and results: we studied a nine year-old patient with symptoms of perennial rhinitis and un-specific vulvovaginitis of torpid evolution. In vivo and in vitro allergologic tests were performed as well as complete analytic tests including immunoglobulins, urine tests, nasal culture, and parasitic test.

Skin test was positive for Dermatophagoides pt. as well specific IgE (99.5 kU/L). Total IgE was elevated for her age (492 kU/L). In the rest of the complementary tests, no values out of normality or pathological findings were obtained.

Conclusions: considering these results, it was suspected that the nasal symptoms and the vulvovaginitis presented by the patient are of allergic aetiology by hypersensitivity to the mite Dermatophagoides pt. The study did not prove relation with bacteria, parasites, Candida albicans or any inhalant allergens other than mites.

After three months of treatment with oral antihistamines and topical chromones, as well as environmental avoiding measures, the symptoms totally yielded.

Key words: Vulvovaginitis. Allergy. House dust mite Dermatophagoides pt. Childhood. Atopy. Rhinitis.