XLI REUNION ANUAL
DE LA SOCIEDAD ARGENTINA
DE FARMACOLOGÍA EXPERIMENTAL

PROGRAMA RESUMENES AUTORES

24 al 26 de Noviembre de 2009

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OIII-17

COPAXONE REVERTED CHRONIC STRESS-INDUCED ALTERATIONS IN BEHAVIOUR AND TH1/TH2 BALANCE IN BALB/C MICE.

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Stress has been related to cognitive deficit. The hippocampus, a limbic area involved in learning and memory, is particularly sensitive to the effects of chronic stress. Cytokines have been shown to affect some behaviour, including memory. Moreover, IL-2, IFN-γ and IL-6 has been implicated in psychiatric disorders. Glatiramer acetate (Copaxone®) is a synthetic amino acid polymer that can weakly cross-react with CNS-resident autoantigens and can safely simulate the protective and reparative effects of autoreactive T cells. The aim of the present work was to study copaxone effects in the behaviour and in the TH1/TH2 balance induced by chronic stress in BALB/c mice. We found that BALB/c mice exposed to chronic stress had a poor learning performance respect to control mice in both, alternation behaviour in Y-maze task and habituation in open field. The lymphoid production of cytokines analysed by ELISA showed a decrease of IFN-γ and not changes in IL-2 (TH1-cytokines) and an increase of IL-6, IL-4 and IL-10 (TH2-cytokines) in stressed BALB/c mice. These effects induced by chronic stress were reverted by administration of copaxone (100ug per injection s.c. to four times during three weeks). These results indicate that copaxone is able to reverse both the memory impairment and the TH1/ TH2 cytokine balance. These results suggest that TH1 response could constitute a protective mechanism preventing behaviour impairment.

OIII-18

SHIFT IN THE BAX/BCL-XL BALANCED MAY ACTIVATE CASPASE 3 AND MODULATE APOPTOSIS/NECROSIS ASSOCIATED WITH PROGRESSIVE RENAL DELETION THROUGH INOS, HSP60 AND HIF-1α IN MURINE NEPHROTIC SYNDROME.

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Adryamicin (ADR) - induced nephropathy in mice is a commonly used experimental model for pharmacological studies of human renal diseases. However, its molecular mechanism remains unclear. The primary aim of this study was to explore the relationship between the nephrotic syndrome with renal Bax/Bcl-xl ratio, Caspase-3, Hsp60, iNOS and HIF-1α expressions following ADR administration along 30 days.

CF-1 mice were injected with a single dose of ADR (15 mg/kg, ip). Histological evaluations (PAS/HE) and scanning electronic microscopy were used for morphological descriptions of renal outer cortex. HIF-1α, Bax, Bcl-xl, Caspase-3, Hsp60 and iNOS expressions were determined by western blotting. Blood urea nitrogen (BUN) and Serum Creatinine (sCr) increased by 10 days ($p<0.01$), indicating severe loss of renal function that was associated with widespread ultrastructural abnormalities. Histological semiquantitative scores showed glomerulosclerosis, tubular damage and intestitial infitrates ($p<0.01$). The Bcl-xl/Bax ratio decreased within the first days ($p<0.01$) accompanied by active caspase-3 expression as a hallmark of apoptotic signalling. iNOS, Hsp60 and HIF-1α were up regulated on day 15.

Our findings suggest that changes in the ratio Bcl-xL/Bax triggers caspase-3 activation which modulates renal apoptosis; while iNOS, Hsp60 and HIF-1α are associated with renal inflammation and fibrosis, tubular atrophy; linked to necrosis in this experimental model.

OIII-19

GASTROPROTECTION AND ANTIMICROBIAL ACTIVITY OF *Lithraea molleoides* AGAINST *Helicobacter pylori*

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Lithraea molleoides (Vell.) Engl. (Anacardiaceae) is known popularly as "molle". *L. molleoides* is used in folk medicine as a diuretic and digestive. In one study, the antibacterial activity of *L. molleoides* was assayed on two clinical strains of *Helicobacter pylori* isolated from gastric biopsies and the reference strain *H. pylori* NCTC 11638, and was evaluated by agar diffusion method in well. An inoculum of 10^6 CFU/ml was plated on Mueller-Hinton agar supplemented with 7% horse blood. The aqueous extract of *L. molleoides* was tested in the following concentrations 10, 100 and 250 mg/ml and 15 µl were added on each well. The plates are incubated at 37ºC in microaerophilic conditions for 3-5 d. The MIC was determined by measuring the zone of inhibition at each concentration. *L. molleoides* exhibed antimicrobial activity in all strains tested with an inhibition zone range of 17-23 mm for 10 mg/ml; 25-33 mm for 100 mg/ml and 24-35 mm for 250 mg/ml. The results indicate that *L. molleoides* have activity antimicrobial against *H. pylori* strains.

In other study, absolute ethanol was employed as ulcerogenic agent in Wistar rats (Method of Robert y col.). Infusion 20% of *L. molleoides* reduced ethanol-induced gastric damage in rats ($p<0.01$). The results presented indicate that *L. molleoides* prevents the formation of gastric lesions and has significant antimicrobial properties against *H. pylori*. *L. molleoides* could represent an useful tool in relieving digestive disorders.

OIII-20

CHEMICAL ASSESSMENT OF THE NEW ANTIMICROBIAL PEPTIDE AP-CECT7121

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The emergence of multi-resistant bacteria, involves a serious therapeutic concern in clinical practice. Antimicrobial peptide (AP) CECT7121 was isolated from an environmental strain of *Enterococcus faecalis CECT712,* showing high *in vitro* efficacy against most of the pathogens recalcitrant to conventional treatments. The goals of this research work were the isolation, purification and identification by chromatography (RP-HPLC, TLC and HPLC-MS/MS) of the peptide AP-CECT7121. The BHI broth was inoculated with of an overnight culture of *E. faecalis* CECT7121. After incubating and chemical processes, the active compound obtained, was loaded on C_{18} cartridges previous to RP-HPLC analysis. AP-CECT7121 was identified as a lipophylic compound by a peak at 24 min of retention time. The same fraction was used for the aminoacidic assessment using TLC, where a sequence of six aminoacids was obtained (proline, treonine, cistine, valine, tirosine y leucine). This antimicrobial compound after HPLC-MS/MS analysis showed a low molecular weight of 910 Da. Developing of AP-CECT7121 may be a potential tool for the treatment of Human and Veterinary multi-resistant bacterial infectious diseases.