IL-1β. It is not known whether IL-1β may alter the affinity and/or the number of BK B2 receptor or induce BK B1 receptor in bronchial smooth muscle cells. Therefore, we investigated BK receptors in Human bronchial smooth muscle cells and examined whether IL-1β might alter BK receptors by studying [3H]BK binding after different times of IL-1β treatment. Saturation experiments indicate a single population of binding sites (Kd = 0.29 ± 0.01 nM; Bmax = 109.6 ± 14.9 fmol/mg prot.). Competition experiments with B1 and B2 ligands indicated the order of potency: NPC 17761 = Hoe 140 > BK > WIN 64338 > des Arg9-BK > des Arg9[Leu8]-BK suggesting that BK receptors are of the B2 subtype. IL-1β treatment (10 U/ml) induced an increase of BK B2 receptor density. This increase started after 3 hrs (24.5%), reached a maximum after 6 hrs (86.6%) and still at 30% after 24 and 48 hrs of treatment. In any cases, the affinity of [3H]BK was not modified. We observed also an increase of the B2 mRNA after 2 and 3 hrs of IL-1β treatment. These data suggest (i) that Human bronchial smooth muscle cells express B2 receptors (ii) the mechanism of IL-1β induced airway hyperresponsiveness to BK does involve the induction of B2 receptors and an upregulation of the B2 mRNA.

P3T-365

CHANGES ON PLASMATIC BIOCHEMICAL PARAMETERS, INDUCED BY SALBUTAMOL AT THERAPEUTIC AND DOPING DOSAGES IN AEROBIC EXERCISED RATS

Mar Cepero, J. Carlos Cubría, Marta Lozano *, Rosa Reguera, Rafael Balafia-Fouce, David Ordoñez. Departamento de Fisiología, Farmacología y Toxicología. Universidad de León, Campus de Vegazana 24071 León, Spain

Salbutamol (Ventolin®) is a bronchodilator drug currently used in allergic induced asthma. However, the use of this compound and other β2-adrenergic drugs is not only restricted to therapeutic uses, but also in the physical performing of elite sportsmen. The repartitional effect of β-adrenergic drugs increases the protein deposition in skeletal and heart muscle, and depletes the lipidic content on fat and muscles. These effects are induced by means of adrenergic stimulation and can be prevented by using antagonists like propranolol. Present study concerns on the effect on plasmatic enzymic profiles of exercised and control rats treated with salbutamol at therapeutic and doping dosages.

The effect of salbutamol on heart and skeletal muscle was dose-dependent and completely abolished by co-administration of propranolol. This suggesting the adrenergic implications of both phenomena. The increase of muscular mass weight was higher in exercised rats than in no exercised. Muscular damage marker enzymes, CK, LDH and the cardiac isozyme CKmb were highly increased in a dose-dependent way in both trained or control rats after salbutamol administration.

Conversely salbutamol produced a fat removal from adipose tissue, paralleling to a dose-dependent depletion of plasmatic lipids, triglycerides and cholesterol. All these effects were abolished by the unspecific β-agonist propranolol.

Keywords: salbutamol; physical performance; β-agonist; muscle skeletal; plasmatics parameters; doping

P3T-366

CORNEAL DAMAGE INDUCED BY CONTINUOUS INFUSION IN RATS

Olivier Loget, Roy Forster *, Jean-François Le Bigot. CIT, Centre International de Toxicologie, Evreux, France

Preclinical studies by the intravenous route, via an indwelling catheter, are often performed to mimic the proposed clinical route. In such studies, increased incidence and severity of corneal lesions had been observed in rats continuously infused. Since the origin of this increased ocular damage was unknown, we performed a study in order to determine if these changes were related to anesthesia, infusion material or infusion method.

Sprague-Dawley rats were allocated to 4 groups. In group 4, the animals were anesthetized using an association of acepromazine and ketamine chlorhydrate and surgically prepared for continuous intravenous infusion by location of an indwelling catheter in the vena cava caudalis through one of the femoral veins. After location of the catheter, the animals were continuously infused (24 h/24 h) during 13 weeks with a sterile isotonic saline solution, at a rate of 1 ml/kg/hour. In group 3, the animals were anesthetized under the same conditions and dressed with the jacket usually used for infusion but not operated. In group 2, the animals were anesthetized under the same conditions and at the same occasions but without jacket. Group 1 animals consisted of absolute controls (not anesthetized). Gross examinations, indirect ophthalmoscopic and biomicroscopic examinations were performed before surgery as well as every week for the first four weeks and every 2 weeks thereafter.

Interestingly, corneal lesions were mainly observed in groups 4, 3 and 2 suggesting a relationship with the anaesthesia.

Keywords: continuous infusion; ketamine anesthesia; corneal lesions; rats

P3T-367

ONE-MONTH INTRAVENOUS TOXICITY STUDIES OF POLOXAMER 188 IN MALE SPRAGUE-DAWLEY RATS AND IN BEAGLE DOGS

Catherine Duvinage *, Stephanie Millecamps, Anne Sagner, Magali Guffroy, Jean-Pierre Sarsat, Vincent Belin. Département Sécurité du Médicament, CRVA, Rhône-Poulenc Rorer S.A., Vitry-sur-Seine, France

Poloxamer 188, a nonionic surface active agent used as a vehicle for pharmaceutical formulations, was administered by intravenous route for 1 month to male Sprague-Dawley rats at 100, 300 or 1000 mg/kg/day, and to beagle dogs at 250, 500 and 1000 mg/kg/day. Parameters evaluated included in-life observations, clinical pathology, necropsy and histopathologic evaluation of the liver, lungs, kidneys and injection sites. Treatment-related changes in rats were limited to accumulation of foamy macrophages in the lungs and intracytoplasmic vacuolation of proximal tubular epithelial cells in the kidneys at 300 and 1000 mg/kg/day, associated at 1000 mg/kg/day with increased organ weights. In dogs, mild body weight loss and irregular food intake were noted at 1000 mg/kg/day. Transient changes during and immediately after dosing in all treated dogs included increased heart rate and/or decreased blood cell parameters possibly related to increase in oncotic pressure by the compound and subsequent hypervolemia. Histopathologic changes were noted at 250 mg/kg/day and above in kidneys (intracytoplasmic vacuolation of proximal tubular epithelial cells, also noted at 1000 mg/kg/day in glomeruli). In both studies, local tolerance at the injection sites was good. Consequently, this vehicle should not be used in subchronic toxicity at 250 mg/kg/day or more in dogs, and at 300 mg/kg/day or more in rats. At lower dose levels in rats, poloxamer 188 was considered to be well tolerated. Other intravenous toxicity studies performed in cynomolgus monkeys at doses up to 30 mg/kg/day (and using poloxamer 188 as the vehicle for developmental drug) did not show evidence for specific toxic effects.

Keywords: intravenous poloxamer 188; Sprague-Dawley rats; beagle dogs; vaculuation; kidneys; lungs