

CONCENTRATION OF CARPROFEN IN THE AQUEOUS HUMOR OF DOGS WITH UVEITIS. PRELIMINARY RESULTS.

Allgewer¹, A. Fischer¹, L. Brunnberg¹, U. Knoll², F.R. Ungemach³
 (Small Animal Clinic, Free University Berlin, Germany¹; Inst. of Pharmacology, Pharmacy and Toxicology, Faculty of Veterinary Medicine at the University of Leipzig, Germany²)

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Purpose: To determine the concentration of carprofen in the aqueous humor of dogs with and without uveitis after intravenous carprofen application.

Methods: 37 canine paired samples (aqueous humor and serum) were collected. Patients were divided into three groups: group I (patients without clinical signs of uveitis, n=11), group II (dogs with lens induced uveitis at the time of cataract surgery and dogs with lens luxations at the time of lens extraction, n=19), group III (72 hours after intraocular surgery, n=7). All patients received carprofen (4,4 mg/kg) intravenously 40 minutes before sample collection. Carprofen concentrations were determined in aqueous and serum, while protein and prostaglandin (PGE₂) levels were measured in aqueous humor only.

Quantification of carprofen (as racemate) in serum and aqueous humor was carried out using high-performance liquid chromatography (HPLC) with fluorescence detection (excitation at 300 nm, emission at 375 nm). Sample preparation for serum and aqueous humor (50 µl each) before HPLC was done by addition of buffer (0,1 mol/l citrate, pH 4,8) and internal standard (S-(+)-naprofen) was 0,02 and 0,10 µg/ml in extraction with methyl-tert-butyl ether. The limit of quantification (LOQ) was 0,02 and 0,10 µg/ml in aqueous humor and serum, respectively. (Method: Internal laboratory procedure of the Inst. of Pharmacology, Pharmacy and Toxicology, Faculty of Veterinary Medicine at the University of Leipzig, Germany).

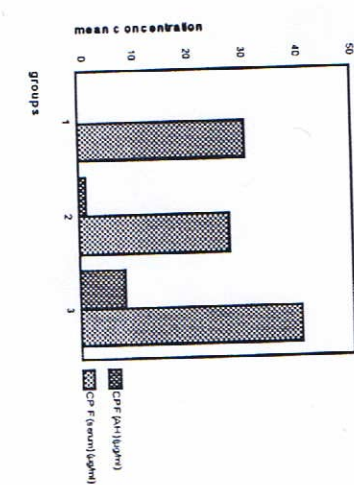
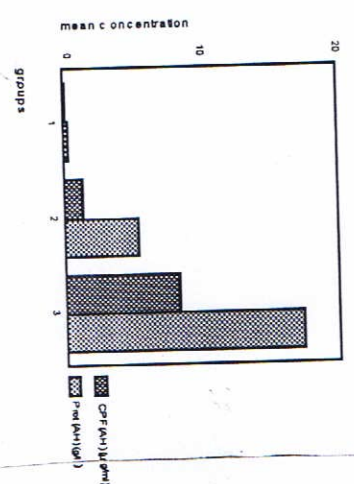
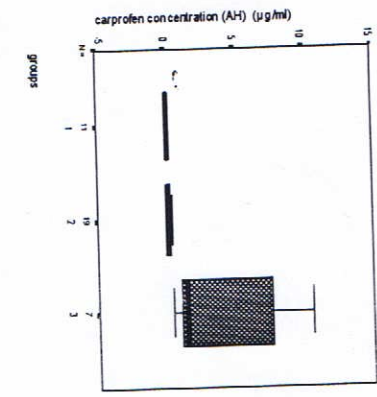
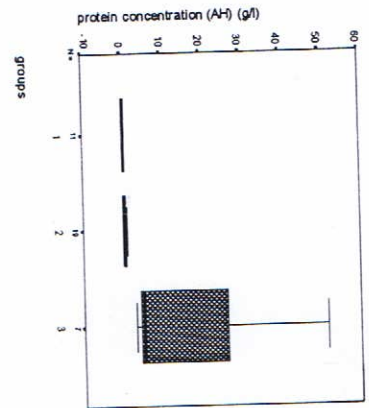
Quantification of total proteins in aqueous humor was done using a modified micro method according to a reported technique (Lowry, 1953). 10 µl aqueous samples were analyzed in duplicate after dilution with isotonic saline solution (dilution ratio between 1:4 and 1:160, dependent on aqueous protein levels). LOQ: 0,12 g/l.

Determination of PGE₂ was carried out by enzymeimmunoassay (EIA) using a commercial test system (RPN 222, Amersham Pharmacia Biotech, UK). Assay was performed in principle according Test protocol 5 (using novel lysis reagents), using 50 µl aqueous samples (in duplicate). PGE₂ curve range: 2,5-320 pg/well, accordant 0,05-6,40 ng/ml. Extension of the upper limit to 12,8 ng/ml was achieved by aqueous sample dilution with assay buffer.

Results: In group I the mean aqueous carprofen concentration (CPF_{AH}) was 0,14 µg/ml and the mean serum carprofen concentration (CPF_{serum}) 31,71 µg/ml. The mean aqueous protein concentration (Prot_{AH}) was 0,356 g/l, the mean aqueous PGE₂ concentration (PGE_{2,AH}) ranged around 5,68 pg/well. The mean ratio of CPF_{serum} / CPF_{AH} was 226,5.

In group II the mean CPF_{AH} was 1,26 µg/ml and mean CPF_{serum} was 32,98 µg/ml, mean Prot_{AH} was 5,61 g/l, mean PGE_{2,AH} was around 30,27 pg/well. The mean ratio of CPF_{serum} / CPF_{AH} was 26,17.

In group III the mean CPF_{AH} was 8,45 µg/ml and mean CPF_{serum} was 40,60 µg/ml, mean Prot_{AH} was 17,31 g/l, mean PGE_{2,AH} was around 34,70 pg/well. The mean ratio of CPF_{serum} / CPF_{AH} was 7,31.



Conclusions: Carprofen was detected in all aqueous samples. The carprofen concentration increased in eyes with uveitis. These results suggest a positive correlation between inflammatory parameters and carprofen in the aqueous humor.