

# A STUDY ON VITREOUS HUMOR BETWEEN-EYE DIFFERENCES AND BASELINE VALUES OF POTASSIUM, CALCIUM, SODIUM AND GLUCOSE IMMEDIATELY AFTER DEATH IN DOGS

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## ABSTRACT

*The present study was conducted to study the vitreous humor between-eye differences and baseline values of potassium, sodium, calcium and glucose immediately after death in dogs. Eight canine carcasses brought for necropsy formed the material for collection of vitreous humour in the present study. The information regarding time of death was gathered from dog owners, clinicians and clinical records. Eyes were enucleated as soon as possible after receiving carcass for postmortem. Sampling of vitreous humor was done using 16 gauge needle and 20 ml syringe. The needle was inserted at the lateral angle of eyeball so that the tip of needle was placed at the center of the eyeball. About 50 µL vitreous humour was gently aspirated from each eye and transferred to eppendorf tubes. Sampling of vitreous humour was done within 1-4 hours after death. The aspirated vitreous humour samples were centrifuged at 13,000 rpm for 15 min at 5°C temperature and the supernatant was used for analysis. The average vitreous humour concentrations of potassium, sodium, calcium and glucose estimated within 1-4 hours after death were 8.18±0.31 mEq/L, 152.55±4.46 mEq/L, 9.52 ±0.20 mg/dL and 21.70±4.50 mg/dL respectively. The results of this study indicated that there were no significant differences between left and right eye for all of the vitreous biochemical constituents that were studied in present study (estimated within 1-4 hours after death).*

**Key words :** Dog, post-mortem, vitreous humour, biochemical, baseline values

## INTRODUCTION

Vitreous humor is a fluid that is relatively well protected from postmortem degradation and contamination. Due to its postmortem stability, vitreous humor has high utility in forensic pathology. Vitreous humor biochemical constituents, especially potassium, have been widely used in the postmortem

interval (PMI) estimations. The time dependent rise of vitreous potassium levels in the postmortem period has been considered to be helpful in PMI determinations. The relative stability of vitreous biochemistry is also useful in assessing the antemortem metabolic status and in predicting the antemortem serum biochemistry of an individual. However, the validity of vitreous biochemistry in forensic

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applications has been questioned in light of the reported concentration differences of various biochemical constituents in the same pair of eyes at identical PMI (Mulla *et al.* 2005). The availability of literature related normal antemortem or baseline values of various vitreous humour biochemical parameters immediately after death of animals is limited. Hence, the present study was conducted to study the vitreous humor between-eye differences and baseline values of potassium, sodium, calcium and glucose immediately after death in dogs.

#### MATERIALS AND METHODS

Eight canine carcasses brought for necropsy to the department of Veterinary Pathology, Madras Veterinary College, Chennai formed the material for collection of vitreous humour in the present study. The information regarding time of death was gathered from dog owners, clinicians and clinical records.

Eyes were enucleated as soon as possible after receiving the carcasses for postmortem. Sampling of vitreous humor was done using 16 gauge needle and 20 ml syringe. The needle was inserted at the lateral angle of eyeball so that the tip of the needle was placed at the centre of the eyeball. About 50  $\mu$ L vitreous humour was gently aspirated from each eye and transferred to eppendorf tubes. Sampling of vitreous humour was done within 1-4 hours after death. The aspirated vitreous humour samples were centrifuged at 13,000 rpm for 15 min at 5°C temperature and the supernatant was used for analysis.

The estimation of vitreous potassium, calcium, sodium and glucose concentrations were carried out immediately after collection on semi-autoanalyzer (MISHPHA BT-320)

using readymade kits supplied by Span Diagnostics Limited, Surat, India. The results were subjected to statistical analysis by using IBM SPSS Statistics (version 20.0) software. The data was processed for paired t-test.

#### RESULTS AND DISCUSSION

The average vitreous humour concentration of potassium, sodium, calcium and glucose estimated within 1-4 hours after death is presented in Table 1. The average vitreous humour concentrations of potassium, sodium, calcium and glucose estimated within 1-4 hours after death were  $8.18 \pm 0.31$  mEq/L,  $152.55 \pm 4.46$  mEq/L,  $9.52 \pm 0.20$  mg/dL and  $21.70 \pm 4.50$  mg/dL respectively. The results of this study indicated that there were no significant differences between left and right eye for all of the vitreous biochemical constituents that were studied (estimated within 1-4 hours after death).

The results of present study are useful in establishing baseline postmortem vitreous biochemical values in canines. The validity of vitreous biochemistry in forensic applications has been questioned in light of the reported concentration differences of various biochemical constituents in the same pair of eyes at identical PMI. Some earlier studies (Balasooriya *et al.*, 1984 and Pounder *et al.* 1998) on vitreous humour reported the variation in potassium level between right and left eye but in present study no such significant difference observed within 1-4 hours after death which is in consonance with some earlier reports (Adjutantis and Coutselinis, 1972; Agrawal *et al.*, 1983; Hughes, 1965; Mulla *et al.*, 2005; & Woolf and Gremillion-Smith, 1983). The reasons quoted in several studies regarding the variation in potassium level between left and right eye includes less

volume of sample, sampling technique, undue pressure during aspiration, sample quality, equipment & method used for estimation (Agrawal *et al.*, 1983; Coe and Apple, 1985; Brzezinski and Godlewski, 2004; Coe, 1969; McNeil *et al.*, 1999; & Woolf and Gremillion-Smith, 1983).

To conclude, the study has given the baseline reference values for vitreous humour potassium, sodium, calcium and glucose. There was no significant difference between-eye values for the vitreous biochemical constituents estimated within 1-4 hours after death.

**Table-1** : Mean ( $\pm$ S.E.) concentrations (within 1-4 hours after death) of various biochemical parameters in vitreous humour of dogs

Parameter	Eye	Mean $\pm$ SE (n=8)	Range	Overall Mean $\pm$ SE	Overall Range
<b>Potassium</b> (mEq/L)	Left	8.12 $\pm$ 0.42	6.22 - 9.46	8.18 $\pm$ 0.31	6.22 – 9.84
	Right	8.25 $\pm$ 0.49	6.82 - 9.84		
<b>Sodium</b> (mEq/L)	Left	151.62 $\pm$ 6.03	126.80-184.00	152.55 $\pm$ 4.46	126.80 – 193.69
	Right	153.49 $\pm$ 6.97	131.83-193.69		
<b>Calcium</b> (mg/dL)	Left	9.45 $\pm$ 0.26	8.69 - 10.99	9.52 $\pm$ 0.20	8.69 – 11.69
	Right	9.59 $\pm$ 0.33	8.72 - 11.69		
<b>Glucose</b> (mg/dL)	Left	22.52 $\pm$ 7.24	5.37 - 68.06	21.70 $\pm$ 4.50	5.07 – 68.06
	Right	20.88 $\pm$ 5.87	5.07 - 56.72		

Mean values within a same column for particular parameter did not differ significantly ( $P>0.05$ ) by paired t-test.

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