

Role of vitreous humor electrolytes in determining time since death

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Abstract

Background: Estimation of time of death is one of the most important requirements in medico-legal autopsies. Role of electrolytes is fundamental to the management in determining time since death. Vitreous humor was preferred as it was easy to obtain in sufficient quantity and without contamination. Vitreous humor potassium acts as a valuable marker in the determination of post-mortem interval. After death, post-mortem vitreous potassium levels change because of cellular hypoxia which induces the depletion of ATP and loss of selective membrane permeability for ions, after which intracellular potassium diffuses with the passive diffusion into the vitreous body leading to increase in vitreous potassium levels. Role of electrolytes is essential and were introduced to assist precise estimation of the time since death. With this background, this study is carried out to evaluate the role of electrolytes in determining time since death. **Aims and Objectives:** To evaluate the role of electrolytes in determining time since death. **Materials and Methods:** 80 deaths of age group 5-83 yrs with dead bodies of the patients died outside the hospital brought by police whose exact time of death is not known and with the bodies of the patients who died in the hospital and whose exact time of death is known were included in the study. Their sodium, Potassium and chloride levels were estimated. **Results:** The study showed vitreous potassium levels in right and left eyes of cases and control were associated significantly with post-mortem interval. Whereas there was no significant association found between vitreous levels of sodium and chloride with PMI. As PMI increases, level of vitreous sodium and chloride were not found to be significantly changed. **Conclusion:** There is linear relationship between vitreous potassium concentration and time since death.

Key Words: Vitreous, post-mortem interval (PMI), potassium.

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INTRODUCTION

Estimation of time of death is one of the most important requirements in medico-legal autopsies. Role of electrolytes is fundamental to the management in determining time since death. The incidence of unnatural

deaths is found to be persistently increasing. Pattern of unnatural deaths is a reflection of the prevailing social set up and mental health status of the population. The interval between death and time of examination of body is called post-mortem interval. This is important to know when the crime was committed. The chemical tests to determine the PMI have been largely developed in the past few decades. Various body fluids which are available for the chemical examination are whole blood, serum, Cerebrospinal fluid, pericardial fluid, aqueous humor, vitreous humor and synovial fluid. Amongst these, the most widely used method is vitreous humor which is investigated in this study. Vitreous humor potassium acts as a valuable marker in the determination of post-mortem interval. The linear relationship of increase in vitreous potassium concentration to lengthening post-mortem interval is arithmetic rather than logarithmic. After death, post-

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mortem vitreous potassium levels change because of cellular hypoxia which induces the depletion of ATP and loss of selective membrane permeability for ions, after which intracellular potassium diffuses with the passive diffusion into the vitreous body leading to increase in vitreous potassium levels. Role of electrolytes is essential and were introduced to assist precise estimation of the time since death. With this background, this study is carried out to evaluate the role of electrolytes in determining time since death

MATERIAL AND METHODS

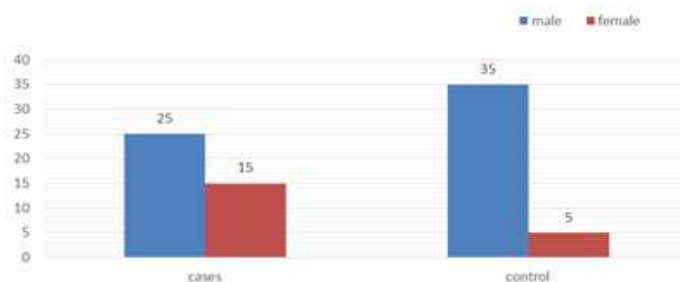
This was a case control study carried out for a period of 2 months in 2019 at Forensic Medicine Department, Government Medical College, Nagpur. The study population comprised of 80 patients (40 cases and 40 controls) of age group 5-83 yrs. Cases included the Study group with unknown time of death that consists of dead bodies, died outside the hospital brought by police whose

exact time of death is not known and the control group consists of the bodies of the patients who died in the hospital and whose exact time of death is known. Dead bodies in state of decomposition were excluded from the study. Cases were included in group with various causes of death on the basis of confirmation by investigating officer and cor-roborative findings at medico-legal examination. Vitreous humor was collected during autopsy with the help of 10 ml. sterile syringe and 20 gauge needle and the sample was collected in a plain rubber stopper bottle. Two vitreous samples were drawn, first from the right eye and the second from the left eye. Samples contaminated with blood, turbid sample and sample with known electrolyte imbalance were discarded. Vitreous Potassium, Sodium and Chloride were analysed on XD-683 electrolyte analyzer. The principle of analyzer is based on Ion selective electrode technique.

Approval from institutional ethical committee was taken. Statistical analysis was done by Microsoft Excel SPSS version 21.

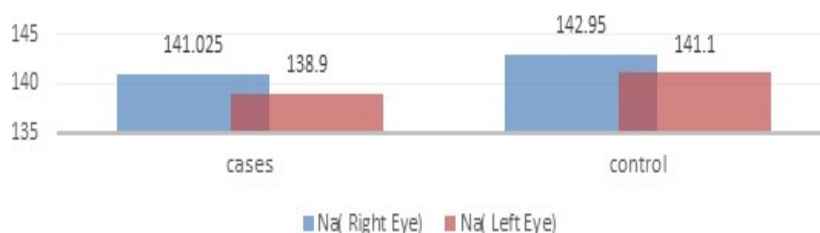
OBSERVATIONS AND RESULTS

The mean values of sodium, potassium, chloride and sodium/potassium ratio in cases and controls were compared by using Student's t test. Also the mean values of sodium/potassium ratio in left eye and right eye were compared by using Student's t test. P value < 0.05 was considered as significant. Out of 40 patients in control group, there were 35 males and 5 females. The minimum and maximum ages noted were 7 years and 80 years respectively. Where as in Cases, out of 40, there were 25 males and 15 females. The minimum and maximum ages noted were 5 years and 83 years respectively. (Graph 1).



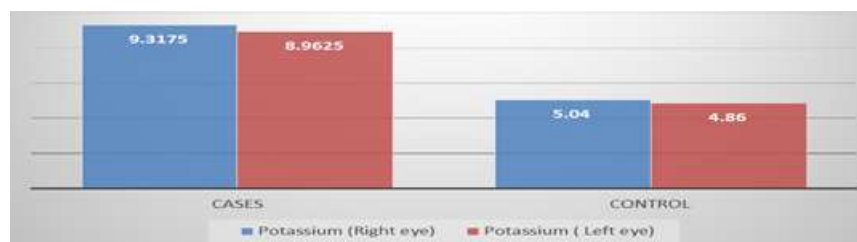
Graph 1: Gender wise distribution of cases and controls

Graph 2 shows, from the statistical analysis, Vitreous sodium level in cases and control, there was no significant difference in sodium level between right and left eye when withdrawn at the same time.



Graph 2: Sodium levels in cases and controls

In graph 3, we got significant difference in vitreous potassium levels between right and left eye of cases and no significant difference in control group when withdrawn at the same time.



Graph 3: Potassium levels in cases and controls

Graph 4, is showing Vitreous chloride levels in cases and control, there was no significant difference in chloride level between right and left eye in cases and in control group when samples were withdrawn at the same time.



Graph 4: Chloride levels in cases and controls

It was observed that potassium level and PMI of vitreous were associated significantly in the given table. (Table 1)

Table 1: vitreous potassium levels in right and left eyes of cases and control

Indicator	Cases Mean	Cases S.D.	Control Mean	Control S.D.	P value	Significance
Rt. K	9.3175	1.876561	5.04	0.55	0.0001	S
Lt.K	8.9625	1.604431	4.86	0.54	0.0001	S

Whereas there was no significant association found between vitreous levels of sodium and chloride with PMI. As PMI increases, level of vitreous sodium and chloride were not found to be significantly changed as shown below table.(Table 2)

Table 2: vitreous Sodium and Chloride levels in right and left eyes of cases and control

Indicator	Cases Mean	Cases S.D.	Control Mean	Control S.D.	P value	Significance
Rt. Na	141.025	8.26946	142.95	8.649737	0.156060479	NS
Lt. Na	138.9	7.2175	141.1	8.359855	0.105784812	NS
Rt.Cl	98	5.782378	97.45	4.125903	0.312933511	NS
Lt. Cl	94.95	5.615683	94.65	4.110961	0.39296334	NS

From the given two regression graphs, it is observed that PMI and vitreous potassium level of cases were correlated significantly. The regression co-efficient for left and right eye were 0.711 and 0.6047. This means that when there is increase in vitreous potassium level by 1 mEq/L in left and right eye, will indicate increase in PMI by 0.711 hours and 0.6047 hours respectively.

Regression equation of PMI on K (Left Eye) is $PMI = 0.711 K (Left) - 1.213$

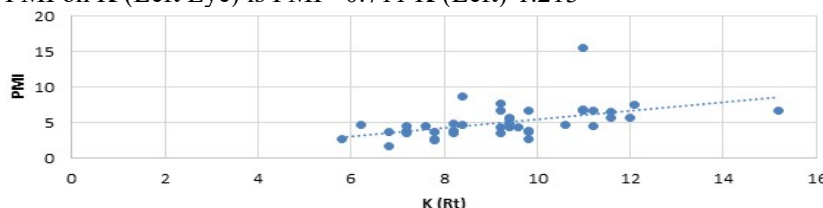


Figure 1: PMI and K (Rt eye)

Regression equation of PMI on K (Right Eye) is $PMI = 0.6047 K (Right) - 0.4714$

From the statistical analysis, shown in the regression graphs mentioned below, It was found that vitreous sodium level and PMI were not co-related significantly.

Regression equation of PMI on Na (Rt) $PMI = - (0.0445) Na - 1.1345$

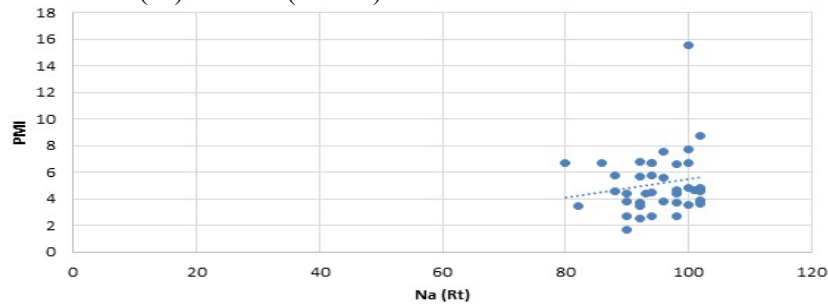


Figure 2: PMI and Na (Rt eye)

Regression equation of PMI on Na (Lt) is $PMI = -0.0606 Na - 3.2724$

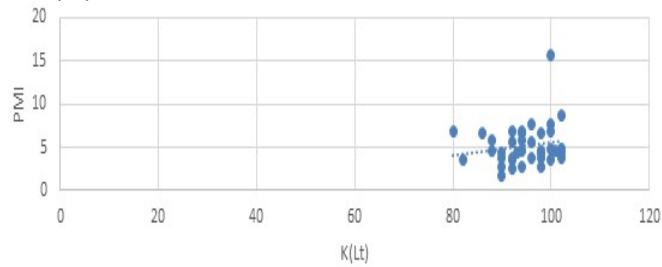


Figure 3: PMI and K (Lt eye)

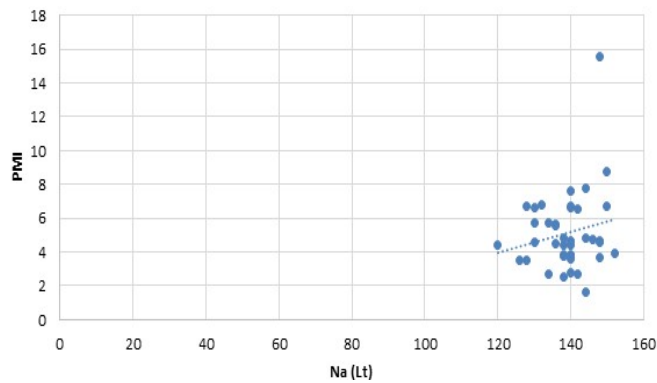


Figure 4: PMI and Na (Lt eye)

From the statistical analysis shown in the below mentioned graphs, it was found that vitreous chloride level and PMI, were not cor-related significantly.

Regression equation of PMI on Cl (Rt) is $PMI = 0.0803 Cl (Rt) - 2.7244$

Regression equation of PMI on Cl (Rt) is $PMI = 0.0803 Cl (Rt) - 2.7244$

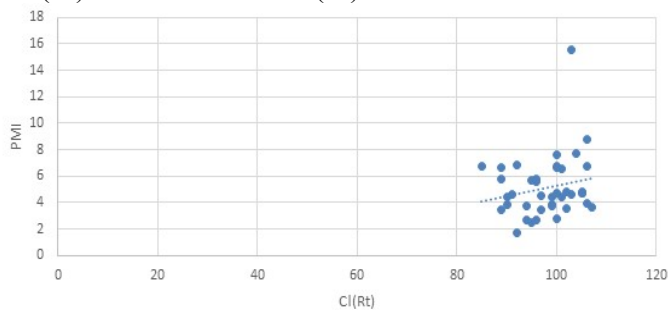


Figure 5: PMI and Cl (Rt eye)

Regression equation of PMI on Cl (Lt) is $PMI = 0.0682Cl(Lt) - 1.333$

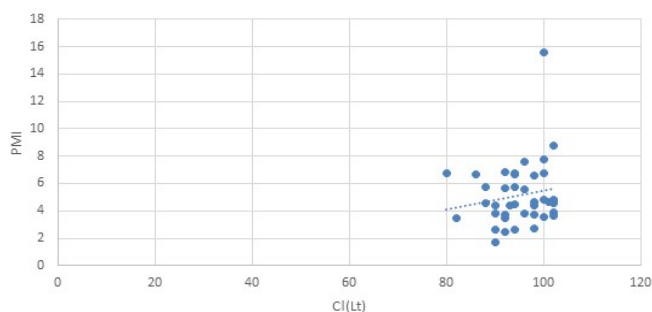


Figure 6: PMI and Cl (Lt eye)

DISCUSSION

In the present study, vitreous humor was preferred as it was easy to obtain in sufficient quantity and without contamination. Vitreous potassium acts as a valuable marker in the determination of post-mortem interval. The linear relationship of increase in vitreous potassium concentration to lengthening post-mortem interval is arithmetic rather than logarithmic.

After death, post-mortem vitreous potassium levels change because of cellular hypoxia which induces the depletion of ATP and loss of selective membrane permeability for ion, after which intracellular potassium diffuses with the passive diffusion into the vitreous body leading to increase in vitreous potassium levels. In this regard, our results are in full agreement with the results of other studies done by Jaffe¹ Coe² Gregora *et al.*³ Catherman *et al.*⁴. The autolysis of vascular choroid and retinal cells is thought to be responsible for this rise. This has been shown by Sterner *et al.*⁵. The concentration of sodium and chloride fall slowly after death, while potassium concentration rises slowly as per the study done by Weichen *et al.*⁶. These changes are reported to be in proportion with the post-mortem interval. However, it was also found that the co-relation between vitreous electrolytes and time since death was not statistically significant, was studied by Weichen *et al.*⁶. The results of study conducted by Balasooriya *et al.*⁷. and Tao *et al.*⁸. where there was a slight negative cor-relation between vitreous sodium and chloride concentration and PMI. There was no significant co-relation found between vitreous sodium and chloride concentration and PMI in the studies conducted by Mulla⁹, Yogiraj *et al.*¹⁰ Jashnani *et al.*¹¹. The difference in the results in relation to sodium and chloride concentration in vitreous humor and PMI between the present study and other studies likely to be due to smaller sample size in the other studies, shorter range of PMI in the other studies and the difference in the analytical methods used for the estimation of sodium and chloride concentration.

Thus, in this study we found significant linear relationship between K and PMI.

CONCLUSION

From the present study, it is concluded that there is linear relationship between vitreous potassium concentration and time since death. Among the other electrolytes, vitreous sodium and chloride have no role in determining post-mortem interval.

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