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A Correlation Study Of Time Since Death By Vitreous Potassium With Postmortem Changes

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ABSTRACT

The interval between the death and the time of the examination of the body is known as postmortem interval. The exact time of death cannot be fixed by any method but only approximate range of time since death can be given because there are considerable biological variation in the individual cases[1]. The present study was carried out in the Morgue and the Department of Forensic Medicine and Toxicology, of Shri V. N. Government Medical College, Yavatmal from Maharashtra. The vitreous humor was sampled from about 201 cases subjected to autopsy. The main objectives of the study are to investigate

1. The utility of vitreous Na+ and K+ level in estimating postmortem interval (PMI).
2. Correlation of postmortem changes with changes in vitreous potassium.

The results of the present study lead to the following conclusions:

1. There is linear relationship between vitreous potassium concentration and postmortem interval.
2. The accuracy of PMI calculated using death changes is less significant and less accurate.

Keywords: Vitreous Potassium, Postmortem interval, Postmortem changes

INTRODUCTION

The interval between the death and the time of the examination of the body is known as postmortem interval. The exact time of death cannot be fixed by any method but only approximate range of time since death can be given because there are considerable biological variation in the individual cases[1].

In the early period i.e., during second half of 18th century the Forensic experts made use of postmortem changes like postmortem cooling, postmortem lividity and rigor mortis to estimate PMI[2]. Unfortunately, the accuracy was low, the margin of error remained large and unpredictable. Therefore the second half of the 19th century onwards, chemical tests were tried to determine PMI by estimating the electrolyte concentration of various body fluids like blood, CSF, synovial fluid, pericardial fluid and vitreous humor[3].

To obviate the problems associated with cerebrospinal fluid, there has been increasing used of vitreous humor for chemical analysis. The eye is isolated and well protected anatomically so vitreous humor is usually preserved despite of serious trauma to the head and is much less subjected to contamination or putrefactive changes than either blood or cerebrospinal fluid. Most important it has been found that the chemical changes for many substances occur much more slowly in the vitreous humor than in the blood or cerebrospinal fluid[4].

Thus in the present study an effort is made to derive a formula to estimate near accurate PMI in this Central India region from vitreous potassium. An effort will be made to know the correlation of postmortem changes with changes in vitreous potassium to determine time since death.

MATERIAL AND CASES

The study was carried out in the Morgue and the Department of Forensic Medicine and Toxicology, of Shri V. N. Government Medical College, Yavatmal from Maharashtra.

The vitreous humor was sampled from about 201 cases subjected to autopsy at Mortuary of this Department.

Inclusion criteria: Case with known time of death, only the clear vitreous humor samples taken.

Exclusion criteria: Cases, where time of death on enquiry from different sources are differ by more than ±30 min, vitreous humor samples either turbid or having any particulate matter or blood is discarded

The vitreous samples was drawn from the eyes of deceased at the beginning of postmortem examination of dead body to the mortuary. The sample was collected and needs to be immediately centrifuged. Liquid paraffin gel or normal saline was injected in the posterior chamber of eye for cosmetic purposes.

The sample was analyzed for sodium and potassium, the analysis for these constituents will be carried out using Flame Photometer Mediflame 129 (Systronics).

RELEVANT AUTOPSY FINDINGS

After sample removal detail postmortem examination carried out including findings with details of livor mortis, rigor mortis, external and internal sign of decomposition.

The statistical analyses for the data were carried out using the Graph Pad Prism Version 5.01 software.

RESULT & OBSERVATION

A total 201 samples in which 145 subjects were males and 56 were females ranging from age group of 09 to 75 years with an average value of 35.52 years used in present study.

In this study the minimum value of known postmortem interval was 1 hours and the maximum of known postmortem interval was 26 hours with an average value of 10.99 hours.

VITREOUS HUMOR BIOCHEMICAL CONCENTRATIONS

Table 1.

PMI in hours	No. Of cases	Range of K^+	Range of Na^+	MEAN ± SD K^+	MEAN ± SD Na^+	MEAN ± SD Na/K Ratio
0 - 6	65	6.1 – 7.4	91.2 - 118	6.49 ± 0.44	114.10±3.80	17.63±1.05
6.1 - 12.0	44	6.4 - 7.2	120 - 130	6.82 ± 0.19	126.81±2.87	18.5 ±0.35
12.1 - 18.0	70	6.3 - 8.2	126 -140	7.44 ± 0.28	135.05±2.98	18.16±0.42
18.1- 24.0	21	7.7- 8.2	138 - 144	7.90 ± 0.11	140.19±2.08	17.73±0.28
24.1 - 36.0	01	8	144	-----	-----	-----
Total (0-36 hrs)	201	6.1 – 8.2	91.2- 144	7.05 ± 0.58	127.1±10.31	18.04±0.76

The observed linear regression analyses correlation of the vitreous potassium and sodium with PMI

Vitreous potassium and PMI

The linear regression correlation of vitreous potassium and PMI The linear rise of vitreous potassium against increasing PMI is represented in Fig. 1 (n: 201; r: 0.8911; P<0.0001).

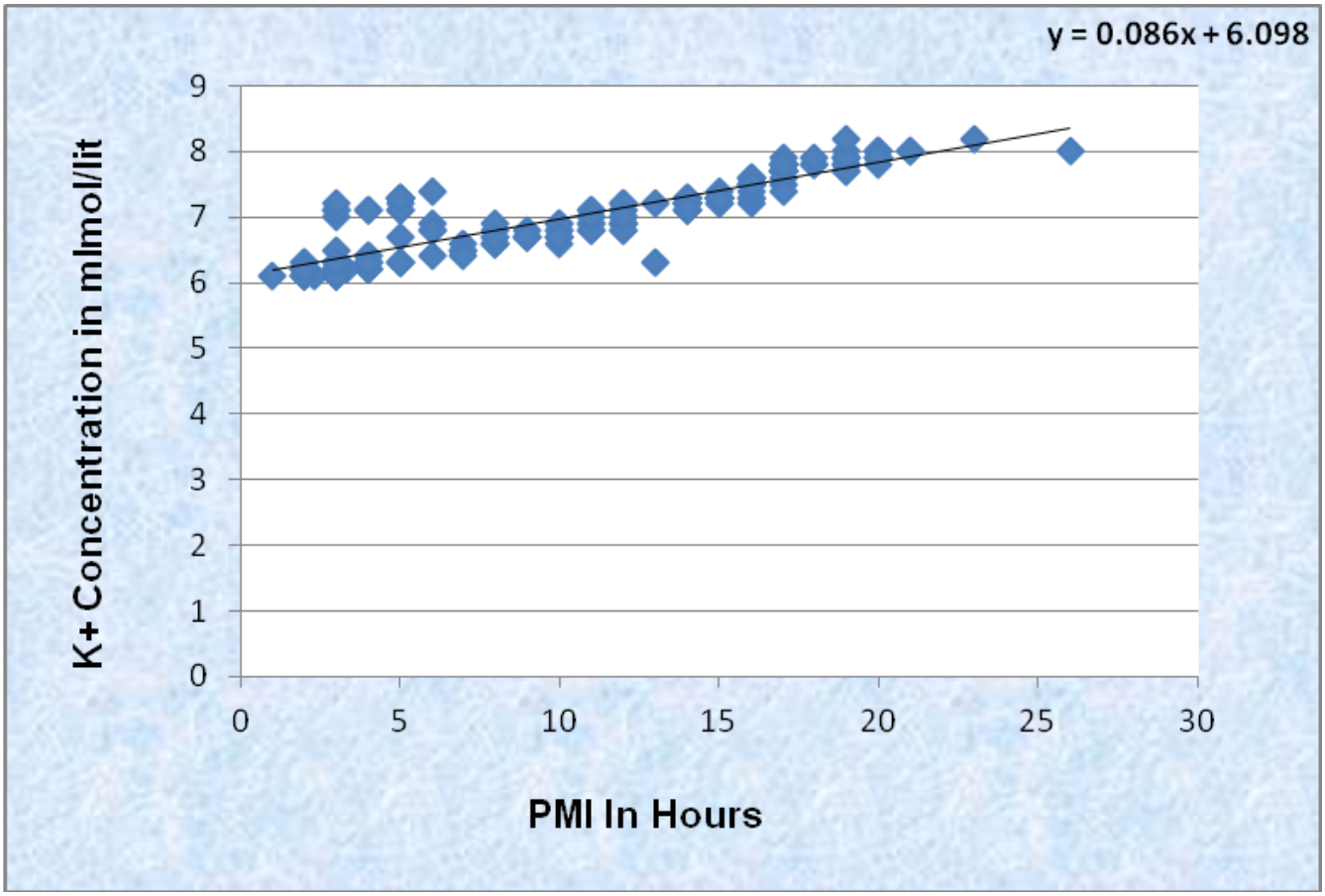


Fig. 1. The regression plot of vitreous humor potassium concentration plotted against the postmortem interval (PMI) in hours

Correlation between Postmortem Interval and Vitreous Humor Potassium For Proposed Formulae

Postmortem Interval (hrs) x variable	**N**	**201**
	Mean	**10.99**
	±S.D.	**±6.06**
Potassium (mmol/L) y variable	N	201
	Mean	7.05
	±S.D.	± 0.58

The vitreous potassium concentrations were used as the dependent variable to calculate the estimated PMI. The resulting linear regression equation in the form of y = ax + b

Where,

'y' is vitreous potassium concentration;

'x' is actual PMI in hours;

'a' is the slope of regression line and a = 0.086, 'b' is the intercept of the regression (Y-intercept when X=0.0) and b = 6.098

Thus the resulting linear regression equation is y = 0.086x + 6.098 ie..

x = (y – 6.098)/ 0.086 or x = 11.63y – 70.90

Thus Postmortem interval estimated by resulting linear regression formula.

PMI = 11.63 (K^+) – 70.90

95% confidence limit of x = Mean ± 2SD of x

= Mean ± 2 x 6.06

= Mean ± 12.12

SD of x

Regression coefficient = r

SD of y

= 0.8911(6.06/ 0.58)

= 9.30

This means that an increase of potassium values of 1mmol/L will indicate an increase of 9.30 hrs in the postmortem interval and 95% confidence limit for all cases will be ± 12.12hrs.

Thus average rate of increase of vitreous potassium was calculated as 0.1075mmol/L per hours.

Comparison of Actual PMI and Estimated PMI by Derived Formula

Statistical analysis By using Paired t test

Comparison	P value	Significance
Actual PMI with Estimated PMI	0.6444	NS

NS - Not significant.

As the potassium based formula derived from the data of the present study to estimate PMI is: PMI = 11.63 (K^+) – 70.90

No significant difference(NS) find between actual PMI and estimated PMI from derived potassium based formula (P 0.6444)

APPEARANCE OF EARLY AND LATE DEATH CHANGES

Table 2.

Group. No.	Rigor Mortis	PM lividity (NF/F)	Ext.s/o decom.	Int.s/o decom.	Duration (Hours)	No. Of cases
1	Not appeared.	In patches (NF)	Absent	Absent	00 to 03	03
2	Appeared	In patches (NF)	Absent	Absent	01 to 04	31
3	Appeared	Well formed (NF)	Absent	Absent	03 to 08	37
4	Well marked	Well formed (FX)	Absent	Absent	08 to 18	107
5	Well marked	Well formed (FX)	Absent	Absent	18 to 24	21
6	Start disapp.	Well formed (FX)	Absent	Absent	18 to 36	01
7	Passed off	Dispersed	Present	Absent	24 to 48	01
	Total					**201**

FX - Fixed, NF - Not Fixed

. On the basis of different combinations of early and late death changes present in the subjects under study they were divided into 7 subgroups viz. group 1 to group 7 and the post mortem interval has been assessed. Thus the Group 1 consists of 3 cases with duration of autopsy PMI 00 - 03 hours having postmortem lividity in patches and not fixed and rigor mortis not appeared. Group 2 includes 31 cases with duration of autopsy PMI 01-04 hours having postmortem lividity in patches and not fixed but rigor mortis appeared. Group 3 having duration of autopsy PMI 03-08 hours includes 37 cases with postmortem lividity well formed but not fixed and rigor mortis appeared. Group 4 consists of 107 cases with duration of autopsy PMI 08-18 hours having postmortem lividity well formed, fixed and rigor mortis well marked. In group 5 include 21 cases having duration of autopsy PMI 18- 24 hours with postmortem lividity well formed, fixed and rigor mortis well marked. While 1 case in group 6 having duration of autopsy PMI 18- 36 hours with postmortem lividity well formed, fixed and rigor mortis start disappearing and 1 case in group 7 having duration of autopsy PMI 24- 48 hours with dispersed postmortem lividity, rigor mortis pass off and external sign of decomposition present.

Comparison Of Actual PMI To The PMI On Autopsy Findings And PMI Calculated Using The Derived Formula

For each individual case actual PMI is known and as per the death changes observed on postmortem examination the Autopsy based PMI in corresponding cases has been calculated from table no.2. Also by applying the formula derived in present study using $[K^+]$ concentration has been proposed to estimate PMI. Thus PMI calculated using proposed formula i.e. PMI = 11.63 (K^+) – 70.90

Table 3. The comparison of actual PMI to the PMI on autopsy findings and PMI calculated using the derived formula for PMI = 11.63 (K^+) –70.90

Group. No.	Autopsy PMI (Duration in Hours)	No. Of Cases	Range Of Actual PMI	Mean±SD Of Actual PMI	Range Of K^+	Range Of PMI By Present Formula PMI = 11.63(K^+) – 70.90	Mean±SD Of PMI By Derived Formula PMI = 11.63(K^+) – 70.90
1	00 to 03	03	1-2	1.67± 0.57	6.1-6.2	0.043-1.206	0.43± 0.67
2	01 to 04	31	2-3	2.58± 0.50	6.1-7.2	0.043-12.836	2.37±3.8
3	03 to 08	37	4-8	5.16± 1.19	6.2-7.4	1.206-15.162	6.89± 4.91
4	08 to 18	107	8-18	13.78±2.78	6.3-7.9	2.369-20.977	13.19± 4.17
5	18 to 24	21	19-23	19.76±0.99	7.7-8.2	18.651-24.667	21.25± 1.51
6	18 to 36	01	19	--	7.8	19.81	--
7	24 to 48	01	26	--	8	22.14	--
	Total	201					

From Table No 2 and 3, mean±SD of actual PMI for group 1 is 1.67± 0.57 hours and mean±SD of PMI by present study derived formula is 0.43± 0.67 hours with autopsy PMI ranges from 00 to 03 hours. For group 2 mean±SD of actual PMI is 2.58± 0.50 hours and mean±SD of PMI by derived formula is 2.37±3.8hours with autopsy PMI ranges from 01 to 04 hours. For group 3 mean±SD of actual PMI is 5.16± 1.1 hours and mean±SD of PMI by derived formula is 6.89± 4.91hours with autopsy PMI ranges from 03 to 08 hours. In group 4 mean±SD of actual PMI is 13.78±2.78 hours and mean±SD of PMI by derived formula is 13.19± 4.17 hours with autopsy PMI ranges from 08 to 18 hours. In group 5 mean±SD of actual PMI is 19.76±0.99 hours and mean±SD of PMI by derived formula is 21.25± 1.51hours with autopsy PMI ranges from 18 to 24 hours. While group 6 with autopsy PMI ranges from 18 to 36 hours and group 7 with autopsy PMI ranges from24 to 48 hours involving only 1 case having actual PMI 19 hours and 26 hours and PMI by derived formula 19.81 hours and 22.14 hours respectively.

DISCUSSION

During lifetime, potassium is almost intracellular. High intracellular concentration of potassium is maintained by Na^+-K^+ pump. After death this Na^+-K^+ pump does not operate, therefore K^+ is leaked out of cell, leading to high postmortem levels. It is postulated that normal antemortem entry route of potassium into vitreous humor is through ciliary body. After death, autolysis of the vascular choroids and retinal cells are responsible for its rise[3].

The levels of sodium in vitreous remained constant for prolonged postmortem intervals. Later, their level decreases at a rate of 1mmol/L per hour[3].

The normal vitreous concentration of potassium is 2.6 - 4.2mmol/L[2] and sodium is 118 - 154 mmol/L.[5]

Vitreous Potassium

The present study of potassium in the vitreous humor and sodium/ potassium ratio shows that there is considerable rise in the levels of potassium with increasing postmortem interval and this is one of the most accurate methods of estimating time since death.

It is observed from Table no. 1 that with the increase in the postmortem interval, the levels of potassium in the vitreous humor go on increasing from normal. The Postmortem interval ranges from 1 – 26 hours with mean postmortem interval 10.99 hours. Vitreous potassium concentration ranges from 6.1 – 8.2 with mean±SD 7.05 ± 0.58. This observation is authenticated by many workers including Garg et al (2004)[2], Yogiraj et al (2008)[3] , Balasoorriya et al (1984)[7], Dalbir Sing et al (2001)[8], Prasad et al (2003)[9].

Comparison of Actual PMI and estimated PMI by Derived formula

Postmortem interval estimated by resulting linear regression formula.

PMI = 11.63 (K^+) – 70.90.

By using Paired t test

As the potassium based formula derived from the data of the present study to estimate PMI is: PMI = 11.63 (K+) – 70.90

No significant difference found between actual PMI and estimated PMI from derived potassium based formula (P 0.6444).

Thus average rate of increase of vitreous potassium was calculated as 0.1075mmol/L per hours.

Dalbir Sing et al (2001)[8] observed that mean rate of increase of potassium concentration was 0.29 mEq/| hr.

Hanson et al (1966)[6] found that the slope during the period when potassium levels were increasing was 0.17 mEq/l per hr with an intercept of 8 mEq/l. The 95% confidence limit was over ± 20 hours.

Prasad et al (2003)[9] found that the average rate of increase of vitreous potassium was calculated as 0.21 mEq/L.

From Table No 2 and 3, It is observed that the accuracy of PMI calculated using death changes is less significant and less accurate, while the mean of actual PMI are correlated with the mean of PMI by present study derived formula.

It is unfortunate that the length of time required to attain a particular degree or type of postmortem changes cannot be categorically furnished because the time of onset and the rates of change are usually governed by unpredictable endogenous and exogenous factors[10].

Hence, death changes are fairly useful to calculate the postmortem interval/time since death as there are many factor which modify the early and late death changes and its variability is such that it is not much useful for accurate estimation of time since death.

As there is no significant difference finds between actual PMI and estimated PMI from derived potassium based formula (P 0.6444).

CONCLUSION

The results of the present study lead to the following conclusions:

1. There is linear relationship between vitreous potassium concentration and postmortem interval.
2. The Vitreous Potassium based formula for the estimation of Postmortem interval is PMI = 11.63 (K^+) – 70.90.
3. Thus average rate of increase of vitreous potassium was calculated as 0.1075mmol/L per hours.
4. The accuracy of PMI calculated using death changes is less significant and less accurate.

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