ORIGINAL ARTICLE

Role of Vitreous Potassium Level in Postmortem Interval Estimation

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ABSTRACT

Background: A consistent rise in the level of vitreous potassium, commencing shortly after death and continuing for 125 hours has been known for 40 years and recommended for the estimation of the time since death.

Aim: To find out the Potassium level in vitreous humor samples taken from the medicolegal autopsy cases and to determine its correlation with the known postmortem interval.

Methods: In this study 102 vitreous humor samples were collected from medicolegal autopsy cases from the mortuary of Forensic Medicine and Toxicology Department, King Edward Medical University, Lahore. Analysis was carried out at the Department of Pathology, KEMU, Lahore by Autoanalyzer using ion selective electrode technique.

Results: A highly significant linear correlation was observed between the vitreous potassium concentrations with the postmortem interval (r=0.428, P<0.001). The estimated PMI in hours was obtained by using the Sturner's equation but it either over estimated or under estimated. The Ordinary Regression equation was more useful.

Conclusion: It is established that the postmortem vitreous potassium value can play an important role in estimating the postmortem interval. The Ordinary Regression equation based upon our own data estimated comparatively more accurate PMI than the well-known Sturner's equation.

Keywords: Postmortem interval (PMI), vitreous humor, potassium**,** Sturner's equation,

INTRODUCTION

From the knowledge of the Forensic Medicine, medical man has to help the investigating agencies in answering the critical questions such as cause of death and time since death[1]. The postmortem interval or time since death is extremely important in the investigation of death cases especially where homicide, suicide or sudden death is suspected[2]. The postmortem interval is the time interval between death and the time of postmortem examination[1,3]. It helps the police to efficiently conduct the investigation[4]. Our aim should be to limit the margin of error[5]. A poor opinion is often worse than no opinion at al[6].

Postmortem phenomena takes over the corpse sooner the vital systems fail and become manifested by definite physical changes in the soft parts of the body including the fluids, which show chemical changes[1]. There are many endogenous and exogenous factors that affect the rates of these postmortem changes[7]. Both the rate method and concurrent method are used for the estimation of postmortem interval. In the rate method cooling of the body, eye changes, postmortem staining, rigor mortis, putrefaction, mummification, and adipocere formation are seen. These findings may be lost due to mishandling of the dead body at the scene of crime or during transportation and refrigeration.[1] In concurrent method stomach and bowel contents, urinary bladder contents, circumstantial evidence like wrist watch time (if it is stopped during RTA), relation to wadu or prayer time are included. Entomology may also be helpful in estimation of postmortem interval.Experienced researchers avoid making a definite statement of the postmortem interval based on a single observation[8].

After death so many chemical changes begin to take place in the body fluids (whole blood, cerebrospinal fluid, aqueous and vitreous humor of eye) and progress in an orderly manner until the body is completely disintegrated[9,10]. Vitreous humor is one of the most widely used body fluids in the postmortem interval estimation[11]. After death, a return to equilibrium occurs at a steady rate. The cell membrane becomes a semi permeable membrane and potassium immediately begins to diffuse from inside retinal cells out into the vitreous at a steady rate resulting in potassium level increase over time for upto 120-125 hours after death[6,12,13]. Many equations were developed to estimate the postmortem interval by inserting the vitreous potassium value[10]. Sturner in a study of 91 autopsies

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proposed a formula using a linear rise in potassium level in vitreous humor during postmortem interval[14,15].

Time since death in hours = 7.14 (potassium level in mEq/L) – 39.1

MATERIAL AND METHOD

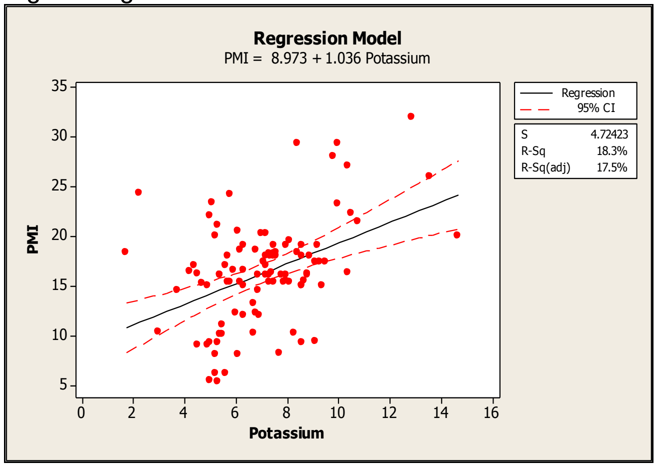
This was a Confirmatory Analytical study. Purposive sampling was carried out. Putrefied, poisoning, with damaged eyes and unknown cases were excluded from this study. Only the cases with exact time of death given by law enforcement agencies were included in the study. Vitreous humor samples were collected from 102 medicolegal autopsy cases at the Forensic Medicine Department, King Edward Medical University, Lahore. These samples were collected at the autopsy table just before conducting autopsy. Vitreous humor was collected from the vitreous chamber of left eye by scleral puncture at the autopsy table by using a 10ml syringe with 20 gauge needle. The tip of the needle was kept in the center of the globe. This technique prevents dislodging of the retina. As much of the fluid sample as can be drawn was obtained as the vitreous humor next to the retina has a different concentration of salutes than in the central part of the eye ball[11,16,17]. On average, approximately 2-3 ml was collected from each subject. For the cosmetic purpose normal saline was introduced through the same needle hole to avoid the collapse of the eye ball. Only clear, transparent liquid free from tissue fragments was used for study. A detailed proforma was filled for each case to collect the relevant information such as name, age, sex, address, police station, cause of death, exact death time, sampling time, and the respective potassium concentration.

The samples were stored at the temperature of □ 20°C as recommended in the literature[18,19]. At the completion of the sampling, the samples were transferred to the Department of Pathology, King Edward Medical University, Lahore for laboratory analysis by using autoanalyzer with ion selective electrode technique. Each sample was centrifuged at the speed of 3000 revolutions per minute for 5 minutes just before analysis.

RESULTS

The collected data was analyzed using the SPSS statistical software version 16. Regression and correlation analysis was used. P- value ≤ 0.05 was considered as statistical significant. Mean, SD, frequencies and percentages were calculated. The results are presented in tables and graphs. Then a scatter plot was utilized to depict the relationship between PMI and K^+ level (scatter plot was made in Mini-tab). The linear regression equation was obtained from K^+ level concentration verses time since death as shown in figure 1.

Fig. 1: Regression model for PMI



In this study 102 samples were included. The mean PMI in hours was found to be 16.35±5.20 with range 5.30 to 32 hours. The mean potassium was 7.11±2.15 with range 1.73 to 14.70mEq/L. A significant correlation was found between PMI and potassium (r=0.428, P<0.001). Data analysis revealed that majority of cases were males 81.37% and females were only 18.63%. It was proposed to divide the studied cases based on the postmortem Interval (PMI) into four groups. From the statistical analysis given in Table 1, it is evident that there is a substantial increase in the vitreous potassium levels with the rise in time since death.

Ordinary Regression Equation was developed by using vitreous potassium level as independent variable is given below:

PMI (Y) = 8.973 +1.036 K^+ *(X)* ---------------------------------

Both the Ordinary Regression and Sturner's Equations were utilized to calculate PMI by using the vitreous potassium values obtained from all the samples. Sturner's equation either overestimated or under estimated the PMI for most of the cases. On the other hand for ordinary regression equation deviations between the actual and estimated PMI were within 6 hours for more than 80% cases. The mean of the error of the calculated PMI (difference between actual PMI and estimated PMI) for both the equations was determined and is shown in Table 2. The mean error margin due to Ordinary Regression was Zero. This is the first basic assumption of regression model that the mean error must be equal to zero, E (e) = 0. However, for the Sturner's equation, the mean error is not zero. Therefore, we can conclude that our Ordinary Regression Model seems to be a good model for our settings.

Post testing of Ordinary Regression model was done by taking four new samples of medicolegal autopsy cases with known PMI by a third party in the mortuary of King Edward Medical University, Lahore. The results obtained from the Sturner's and Ordinary regression equations are shown in Table 3. It is clearly evident from the results that the ordinary regression equation showed estimated PMI with smaller deviations from the actual PMI than the Sturner's equation.

Table 1: Distribution of Cases based on PMI

S No	TSD / PMI	n	%age	Range K^+ (mEq/l)	Mean K^+ (mEq/l)	SD
Group I	0-12 Hours	19	18.63	3 - 9.1	5.89	1.54
Group II	12.1 to 24 Hours	77	75.49	1.73 - 14.7	7.13	1.96
Group III	24.1 to 36 Hours	6	5.88	8.4 - 13.6	10.85	1.99

Statistical Analysis

Comparison	t value	p value	Significance
Group I & Group II	2.958	P<0.005	HS
Group I & Group III	5.6	P<0.005	HS
Group II & Group III	4.421	P<0.005	HS

Table 2: Descriptive Statistics of Errors

Equation	n	Mean
Error due to Ordinary Regression Equation	102	.0025
Error due to Sturner Equation	102	4.6492

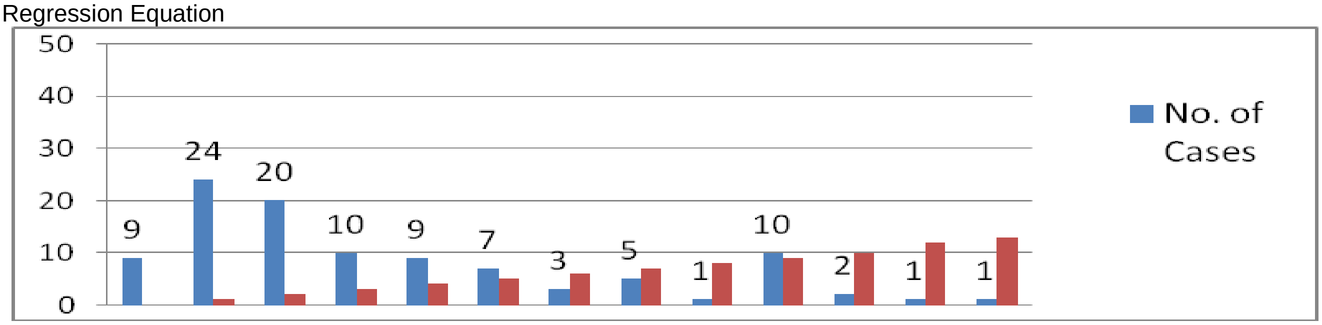
Table 3: Post testing results comparison with the Sturner's equation and Ordinary Regression Equation

K+	Actual PMI	Estimated PMI Comparison	
		Sturner's Equation	Ordinary Regression Equation
6.9	16	10.16	16.12
8.6	16.5	22.30	17.88
7.7	13.5	15.88	16.95
7.6	13	15.16	16.87

Table 4: Deviations (in hrs) between the actual PMI and estimated PMI using the vitreous potassium through the Ordinary Regression Equation

No. of cases	Deviation in Hrs from actual PMI	No. of cases	Deviation in Hrs from actual PMI
9	0	5	7
24	1	1	8
20	2	10	9
10	3	2	10
9	4	1	12
7	5	1	13
3	6		

Fig. 2: Deviations (in hrs) between the actual PMI and estimated PMI using the vitreous potassium through the Ordinary Regression Equation

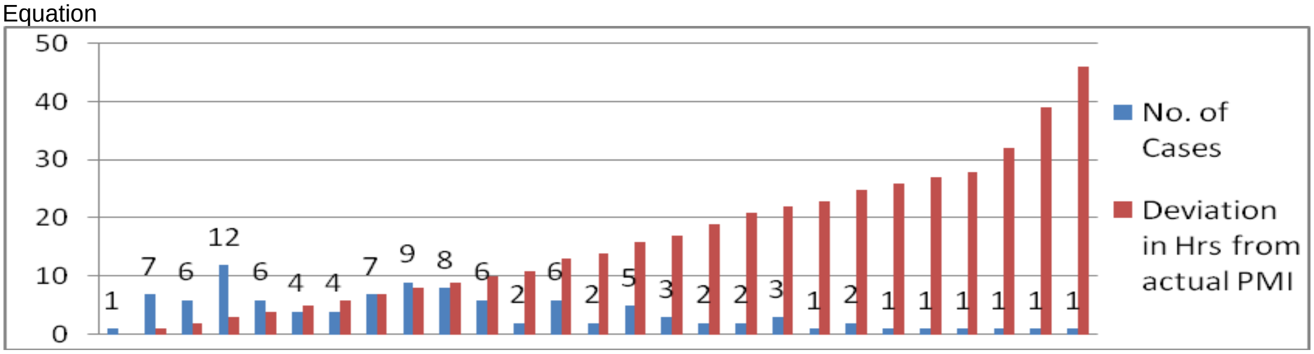


Sturner's equation either overestimated or underestimated the PMI showing no relation with the actual PMI. The deviations from the actual PMI ranged from 27 to 53 hours as shown in figure 3 and table 5.

Table 5: Deviations (in hrs) between the actual PMI and estimated PMI using the vitreous potassium through the Sturner's Equation

No. of cases	Deviation in Hrs from actual PMI	No. of cases	Deviation in Hrs from actual PMI
1	0	5	16
7	1	3	17
6	2	2	19
12	3	2	21
6	4	3	22
4	5	1	23
4	6	2	25
7	7	1	26
9	8	1	27
8	9	1	28
6	10	1	32
2	11	1	39
6	13	1	46
2	14	1	47

Fig. 3: Deviations (in hrs) between the actual PMI and estimated PMI using the vitreous potassium through the Sturner's Equation



DISCUSSION

Many chemical changes start in the body after death in fairly orderly manner. This research has suggested that vitreous potassium is a valuable biochemical marker in postmortem interval estimation. During the postmortem interval, the vitreous potassium represented a fairly linear rise with the rise in postmortem interval. This linear rise of potassium was consistent during the early postmortem interval. The range of vitreous potassium scatter increased during the late postmortem hours especially 20 hours after death. These results are in accordance with previous reports in literature where vitreous potassium showed same pattern[14,15,19,20]. Statistical analysis also highlighted a significant effect of sex on the levels of vitreous potassium. The slope of the regression line for vitreous potassium rise during the postmortem interval in the present study is 0.1768mmol/l per hour. It is essential for the slope of regression line to be relatively steeper because the flatter slopes tend to overestimate the postmortem interval based on the obtained regression line.

Sturner in the year 1963 proposed an equation for postmortem interval estimation. His research based upon sample size of 92 cases[15].

PMI (in hours) = 7.14 × potassium — 39.1

The flatter regression slope (0.14mmol/L per hour) derived from his data led to overestimation of the postmortem interval. Our Ordinary Regression Equation was devised using the vitreous potassium data of 102 samples is given below:

PMI (in hours) = 1.03 × potassium + 8.97

In this equation K^+ is used as independent variable. The Ordinary Regression and Sturner's Equations were utilized to calculate the PMI by using the potassium levels obtained from all the vitreous humor samples in this study.

With the Ordinary Regression Equation, 32% cases had estimated PMI with 2 hours deviations from the actual PMI. Further analysis showed that 82% of the total cases showed deviations of less than 6 hours from the actual PMI (Figure 2, Table 4). The deviations widen as the actual PMI increased. It was observed that rise of vitreous potassium was consistent during the early postmortem hours but during the later hours especially after 20 hours the range of scatter increased.

CONCLUSION

The results of this study showed that the biochemistry of postmortem vitreous humor is significantly important and the vitreous potassium concentration can play an important role in estimating the postmortem interval. It would help the law enforcement agencies and the courts in reaching the right decision. Our analysis has shown that the Ordinary Regression Model developed in this research is comparatively more accurate than other existing formulae for the estimation of PMI. Therefore, we can conclude that this research has resulted in the development of an equation for the estimation of PMI in hours which is more suitable for our setup.

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