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A new model for the estimation of time of death from vitreous potassium levels corrected for age and temperature

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ABSTRACT

Analysis of potassium concentration in the vitreous fluid of the eye is frequently used by forensic pathologists to estimate the postmortem interval (PMI), particularly when other methods commonly used in the early phase of an investigation can no longer be applied. The postmortem rise in vitreous potassium has been recognized for several decades and is readily explained by a diffusion of potassium from surrounding cells into the vitreous fluid. However, there is no consensus regarding the mathematical equation that best describes this increase. The existing models assume a linear increase, but different slopes and starting points have been proposed. In this study, vitreous potassium levels, and a number of factors that may influence these levels, were examined in 462 cases with known postmortem intervals that ranged from 2 h to 17 days. We found that the postmortem rise in potassium followed a non-linear curve and that decedent age and ambient temperature influenced the variability by 16% and 5%, respectively. A long duration of agony and a high alcohol level at the time of death contributed less than 1% variability, and evaluation of additional possible factors revealed no detectable impact on the rise of vitreous potassium. Two equations were subsequently generated, one that represents the best fit of the potassium concentrations alone, and a second that represents potassium concentrations with correction for decedent age and/or ambient temperature. The former was associated with narrow confidence intervals in the early postmortem phase, but the intervals gradually increased with longer PMIs. For the latter equation, the confidence intervals were reduced at all PMIs. Therefore, the model that best describes the observed postmortem rise in vitreous potassium levels includes potassium concentration, decedent age, and ambient temperature. Furthermore, the precision of these equations, particularly for long PMIs, is expected to gradually improve by adjusting the constants as more reference data are added over time. A web application that facilitates this calculation process and allows for such future modifications has been developed.

1. Introduction

1.1. General background

In forensic medicine, the determination of the postmortem interval (PMI) is commonly requested by the police. Therefore, the development of a straightforward and reliable method for estimating PMI has been an active area of research for a long time. Despite the development of new technologies, the most commonly used method for determining PMI is still the rectal body temperature. However, this method can only be applied before the decedent's body has equilibrated with the temperature of the ambient air, which typically occurs within 24 h of death. Other methods for determining PMI include an evaluation of skeletal muscle responses upon mechanical or electrical stimulation and an evaluation of pupil reactions in response to chemical stimulation. However, even these methods can only be applied during the early postmortem period. Thus, for longer PMIs, the forensic pathologist must rely on other methods, most of which often are considered inaccurate.

1.2. Vitreous analyses

One method that may be applicable to a wide range of postmortem intervals is the analysis of vitreous potassium concentration in the eyeball. Under physiological conditions, intracellular levels of potassium are high (~150 mmol/L) in all

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Table 1
Previously published equations for estimating PMI from vitreous potassium concentrations.

Authors (year)	Equation (h)	*n*	Max PMI (h)	Comments
Adelson et al. (1963) [4]	PMI = 5.88 [K^+]–31.53	209	21	–
Sturner and Gantner (1963) [3]	PMI = 7.14 [K^+]–39.1	125	104	–
Hanson et al. (1966) [5]	PMI = 5.88 [K^+]–47.1	203	310	–
Coe (1969) [29]	PMI = 6.15 [K^+]–38.1	145	100	A separate equation was provided for a PMI < 6 h.
Stephens and Richards (1987) [10]	PMI = 4.20 [K^+]–26.65	1427	35	Outliers, drownings, SIDS, electrolyte imbalances, and temperature extremes were excluded.
Madea et al. (1989) [11]	PMI = 5.26 [K^+]–30.9	107	130	Cases involving elevated urea and prolonged agony were excluded.
James et al. (1997) [14]	PMI = 4.32 [K^+]–18.35	100	80	Also included hypoxanthine.
Munoz et al. (2001) [15]	PMI = 3.92 [K^+]–19.04	133	40	Only non-hospital cases were examined, there was a change in variables.
Zhou et al. (2007) [18]	PMI = 5.88 [K^+]–32.71	62	27	–
Jashnani et al. (2010) [19]	PMI = 1.076 [K^+]–2.81	120	50	Mostly included cases involving sepsis or tuberculosis.
Bortolotti et al. (2011) [30]	PMI = 5.77[K^+]–13.28	164	110	–
Mihailovic et al. (2012) [43]	PMI = 2.749 [K^+]–11.98	32	30	Repetitive sampling.
Siddamsetty et al. (2013) [37]	PMI = 4.701 [K^+]–29.06	210	170	–
Present study	$PMI = \frac{\ln((M-C_0/(M-[K^+]))}{L_0+m_A A+m_T T}$	462	409	No cases were excluded. The proposed equation includes temperature and decedent age.

cells of the body [1]. Upon death, active membrane transport, as well as selective membrane permeability, is disrupted, thereby leading to the gradual leakage of potassium out of the cells and into the surrounding extracellular fluid. The vitreous body contains very few cells and its potassium level is similar to any other extracellular fluid [2]. After death, a rise in vitreous potassium due to leakage from surrounding retinal and choroidal cells takes place. Since the 1960s, this rise in potassium levels has been used to estimate the PMI [3–20]. However, despite extensive research regarding this method, there is still no agreement regarding the most accurate equation to describe this rise and to estimate the PMI (see Table 1 and Fig. 1).

The equations that have previously been proposed for estimating PMI are based on an approximation of the diffusion that occurs from the surrounding cells into the vitreous according to a linear model. This may be a reasonable model for a limited period following death. However, it is not appropriate for longer postmortem intervals when vitreous potassium concentration may be the most important means to estimate PMI. Therefore, for the present study, it is hypothesized that the rise in potassium

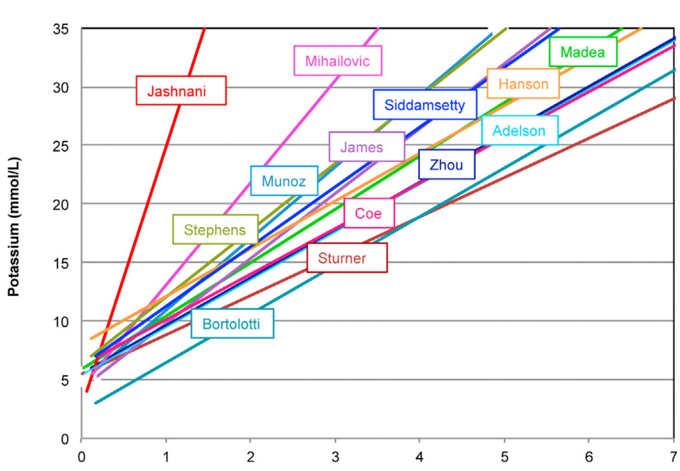

Fig. 1. Regression lines for the calculations of PMI performed in different studies.

displays a non-linear, asymptotic curve and approaches a maximum that is a function of the sum of the potassium ions present in the intra- and extracellular compartments, and their relative volumes. It is further hypothesized that this curve can be right- or left-shifted, and up- and down-shifted, and can have different slope characteristics due to influence of various factors. Regarding the latter, several factors have been proposed:

- ambient temperature [8,12,21–23]
- duration of the terminal episode [4,11]
- renal failure with increased K^+ levels, reflected by elevated urea concentration [11]
- alcohol level at the time of death [11,24]
- sampling method and instrumentation used for analysis [18,25,26].

In this study, we have focused specifically on the changes in postmortem vitreous potassium levels. There are other reports that have evaluated other biochemical parameters, including hypoxanthine [12,14] but the drawback is that such analyses are more time consuming, and typically require more sophisticated analytical procedures, and thus, the results cannot be immediately reported to the police. Moreover, since hypoxanthine also may represent a marker of hypoxia [27,28], the antemortem state of the deceased and circumstances surrounding their death need to be clarified before interpreting the results.

1.3. Objectives of this study

For this study, the potassium levels of more than 3000 vitreous samples that were consistently collected over a three-year period at the Department of Forensic Medicine in Stockholm, Sweden, were compiled. Using these data, the aim of this study was to generate a more appropriate equation for calculating the PMI from vitreous potassium values by taking into account factors that may affect the postmortem diffusion process in the vitreous. Many possible influencing factors were evaluated, but in particular, ambient temperature, decedent age, the presence of alcohol, and agony were investigated to determine whether these factors influence the increase in potassium concentration in the vitreous.