Postmortem Levels of Human Vitreous Humor Sodium, Potassium, Magnesium and Glucose in Different Causes of Death.

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ABSTRACT

Vitreous humor (VH) is a highly valuable sterile accessible sample in postmortem chemistry. Relatively few studies concerned the VH postmortem analysis in differentiating between causes of death. The current prospective cross-sectional study aimed at estimating VH postmortem levels of Sodium (Na), Potassium (K), Magnesium (Mg) and Glucose as differentiating variables between (traumatic, pathological and toxicological) causes of death. In the current work, VH samples were obtained from adult autopsied cases (n = 70) with postmortem interval less than 24 hours and after exclusion of ocular diseases or trauma. The traumatic group had significantly higher VH levels of (Na, K and Mg), in line with significantly lower VH glucose compared to Pathological group. The VH glucose levels were significantly higher in toxicological group compared to traumatic group. Toxicological group had significantly higher VH Na and VH Mg levels as well as significant lower VH glucose levels compared to pathological group. The VH glucose achieved the highest accuracy (84.6 %) in the receiver operating characteristic (ROC) curve analysis for differentiating between traumatic and pathological causes of deaths; followed by VH K (82.6 %). Also, the VH glucose had the highest accuracy (76.2 %) in discriminating pathological from toxicological deaths followed by VH Na (70.8 %). Moreover, VH glucose achieved accuracy of (74.2 %) in differentiating between Traumatic and Toxicological deaths. To conclude, postmortem VH electrolytes and glucose levels showed variable patterns in different death causes. Hence, VH analysis could be used as adjuvant tool in suspecting cause of death; particularly the VH glucose level.

Introduction ·

KEYWORDS

Vitreous.

Glucose,

Electrolytes,

ROC analysis.

Postmortem chemistry,

Vitreous humour (VH) is a clear sterile collagen mesh of connective tissue which is imbedded in an extracellular fluid matrix and located in the posterior chamber of the ocular bulb (Saleh and Makhloph, 2024). The VH volume is about 4.5 mL and is composed of (99%) water, electrolytes (0.9%) and proteins as well as polysaccharides (0.1%) (Tram and Swindle-Reilly, 2018).

⁽²⁾ Clinical Pathology Department, Faculty of Medicine Mansoura University Physiologically, VH has several functions including light transmistion, shock absorbant for mentaining eye shape and protection. Moreover, the VH acts as storage for nutrient needs of the retina as well as a deposit for retinal metabolic waste. The composition and sterility of VH is maintained by the selective blood-retinal barrier (BRB) (O'Leary and Campbell, 2023). During life; active transport across BRB is responsiple for slightly higher VH electrolyte levels as well as lower VH glucose compared to antemortem serum levels (Kokavec et al., 2016).

Postmortem chemistry studies chemical structures, reactions, and parameters of a dead organism. Biochemical analyses of body fluids including blood, VH,

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cerebrospinal fluid and urine are widely used in forensic cases (Omran et al., 2022). The VH is a valuable sample in postmortem analysis which is retained for a longer period before putrefactive degeneration because of both BRB and the restricted vascularization of VH. Hence, VH analysis could give idea about the antemortem biochemical status prior to death (Pigaiani et al., 2020).

Regarding body electrolytes of (Na) postmortem importance, sodium represents the most abundant positive extracellular fluid ion. The diagnosis of hypoor hypernatremia is made possible by postmortem vitreous Na concentrations. which show anomalies in the antemortem serum Na concentrations (Li et al., 2018). Because of the Na/K-pump, potassium (K) is located mainly intracellular during life. Following death, cellular hypoxia and a loss of ion-selective membrane permeability led to increase postmortem VH potassium levels. Many factors affect VH K levels especially postmortem interval (PMI) and causes of death (Chowdhuri et al., 2023; Saleh and Makhloph, 2024). Magnesium (Mg) is the second most prevalent cation in intracellular fluid and the fourth most abundant cation in the body (Fiorentini et al., 2021) The Mg acts as cofactor for enzymatic reactions that control the cell cycle, energy metabolism, signaling pathways, and channels (Tse et al., 2018). Postmortem VH Mg is known to be higher in children and to decrease with age until stabilizing at about age 10 years (Pigaiani et al., 2020). Glucose has a central role in metabolism and acts as precursor of other carbohydrates. In the first six hours following death, glycolysis causes hourly fall in VH glucose concentrations by roughly 35-70% (Zilg et al., 2009). Hence, low postmortem VH glucose concentrations could not be the only pathognomonic sign for fatal hypoglycemia. On the other hand, high VH glucose concentration exceeding 10 mmol/L

(>180 mg/dL) indicates that a hyperglycemic state contributed to the death (Nowak et al., 2020).

Causes of death could be classified according to their eliciting factor into four major categories including, traumatic, pathological and toxicological deaths as well as deaths due to undetermined causes (Schmitt and Mazoori, 2017). The aim of this study was to investigate the pattern of the postmortem levels of VH (Na, K, Mg and glucose) in different causes of death.

Material and Methods:

Study design and subjects:

The present study was a prospective descriptive cross-sectional comparative study that was conducted on adult cadavers of both sexes, with postmortem interval of less than 24 h, who were admitted for autopsy at the Department of Forensic Medicine, Ministry of Egypt in the period between Justice, September 2021 and March 2024. Severely decomposed bodies (Tse et al., 2018; Lutz et al., 2020), suspected ocular diseases or ocular trauma (El Sawaf et al., 2019), and children (Swift et al., 1974) were all excluded from current study to avoid contamination, invalid sampling and age dependent electrolyte changes, respectively. On applying inclusion and exclusion criteria, total of (70) autopsied cases were enrolled in the current work. In all included autopsied cases, age, sex, residence and the cause of death were documented. The documented cause of death (traumatic, pathological, toxicological and undetermined) was determined by history, medical reports and approved by medicolegal autopsy.

Ethical consideration: All the autopsied cases in the current work (n=70)were medicolegal autopsies which were conducted by order of the Public Prosecution. Even the pathological deaths were cases of which death in custody underwent medicolegal autopsy to evaluate the torture possibility. A written Approval of the Assistant Minister of Justice for the Experts and Forensic Medicine Sector as well as the Director of the Forensic Medicine Department was obtained by authors for VH samples collection for research purpose. The current study was authorized by the Institutional Research Board (IRB) Mansoura University (no: MS.21.10.1695).

Methods

Vitrous humor sample collection, preservation and preparation:

The VH samples were collected by aspirating (2 to 3ml) from each eye through the sclera near the center of the globe using a (10 ml) syringe to get total amount about (5 ml) from each case (Butler et al., 2020). No significant differences were reported in the concentration of postmortem VH electrolytes between right and left sides (Zilg et al., 2021), therefore; the collected VH from both eyes was mixed, placed into a separate polypropylene plane tube for each case and preserved at -20^{0} C until chemical analysis was performed (Tse et al., 2018). Unclear VH samples on aspiration were excluded to avoid contamination. After sampling, 2 ml of normal saline was injected into the posterior chamber of each eye for cosmetic reconstruction of the eyeball (Chandrakanth et al., 2013).

Immediately before analysis, samples were left at room temperature for 30 min, then vortexed for 5 minutes, and finally centrifuged twice for 5 minutes each one (12,000 rpm, 13,500 \times g). A 300 μ L sample of the supernatant fluid was taken for analysis (Ave et al., 2021)

Vitreous humor electrolytes and glucose analysis:

The Sodium (Na) and Potassium (K) levels in VH were measured by ion selective electrodes on (Cornley AFT-C Electrolyte Analyzer Düsseldorf -Germany) utilizing (Cornley calibration standard solution (potassium chloride, sodium chloride, sodium acetate) (CAL A Conc Na 140 mmol/l ,k 4 mmol/l - CAL B NA 110 mmol/l, K 8 mmol/l -Düsseldorf (Caretechion gmbh Germany, EM230605B02) kits. The Magnesium (Mg) level in VH was estimated by xylidyl blue method on (COBAS c111 Chemistry Analyzer, Roche Diagnostics. Switzerland, Basel. N: 1498-10) using Magnesium Gen.2 (N. 08900019190, Roche Diagnostics, Germany) kits. Glucose level in VH was assayed by enzymatic reference method with hexokinase on (COBAS c111 Chemistry Analyzer, Roche Diagnostics, Basel, Switzerland, N: 1498-10) using (N.044044830190. GLUH2 Roche Diagnostics, Germany) kits. All measurements were performed following protocols of manufacturers.

Statistical analysis

Data were analyzed using IBM SPSS (Statistical package for social science) (Chicago, USA) version. The data normality was tested with Shapiro-Wilk test. Mean ± Standard deviation (SD) was used for parametric variables. Median and range (minimum – mximum) was for non parametric variables. For comparison between different groups; One-way ANOVA test followed by Tukey-HSD post Hoc test were used for

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parametric data, while as; Kruskal Wallis test followed by Bonferroni post hoc test were used for non- parametric data. Pearson correlation was used to test the correlation between two parametric variables. Spearman's correlation was used to test the correlation between two variables when one of them was non-parametric. Receiver operating characteristic (ROC) analysis was applied to obtain area under curve (AUC) and cut of value of variables with statistically significant difference between groups. Interpretartion of AUC values (0.5 to 0.6= Unsatisfactory ,0.6 to 0.7 = satisfactory, 0.7 to 0.8 = good, 0.8 to 0.9is very good, and 0.9 to 1 =excellent) (Lezhenko and Zakharchenko, 2023). In all tests, *P*-values < 0.05 applied were statistically significant.

Results

As shown in (Table 1), most cases in the current study (45.7%) were in (21-30) age group, followed by (22.6%) of cases were in (31-40) age group. The age group (41-50) represented (11.4%); while as both (18-20) and (51-60) age groups represented (10%). As regard sex distribution, males were (81.4%) and females were (18.6%). As regard residence, (51.4%) of cases were urban, while as (48.6%) were rural.

According to the categorial causes of death the included cases (n=70) were subdivided to: Traumatic group (n=25), Pathological group (n=12) and Toxicological group (n=33). Traumatic group (n=25) included deaths due to falling from height (n=5), hanging (n=4), stabbing (n=4), firearm injuries (n=3), road traffic accident (n=3), freshwater drowning (n=2), torture with blunt trauma (n=3) and burn (n=1). Pathological group (n=12) included deaths due to acute renal failure (n=5), cardiac diseases (n=2), hepatic diseases (n=4) and diabetes mellitus hyperglycemic coma (n=1). Most of the included toxicological cases were fatalities due to phosphide toxicity (n=18), followed by addiction overdose fatalities (n=12) and finally deaths due to unknown ingestion (n=3) (Table 2). No reported cases of undetermined causes of death in the current work.

 Table (1): Frequency distribution of demographic data of the autopsied cases in the current study (n=70).

Domographic Venichles	Study cases $(n = 70)$			
Demographic Variables	n	%		
Age groups				
18- 20 years	7	10		
21 - 30 years	32	45.7		
31- 40 years	16	22.6		
41- 50 years	8	11.4		
51- 60 years	7	10		
Sex				
Male	57	81.4		
Female	13	18.6		
Residence				
Urban	36	51.4		
Rural	34	48.6		

SD: standard deviation, n: number,%: percentage

	Study cases (n= 70)			
Categorial and underlying specific causes of Death	n	%		
Traumatic	25	35.7		
• FFH	5	7.1		
• Hanging	4	5.7		
• Stabbing	4	5.7		
• Firearm	3	4.3		
• RTA	3	4.3		
• Fresh water drowning	2	2.9		
• Torture (Blunt injury)	3	4.3		
• Burn	1	1.4		
Pathological	12	17.1		
• ARF	5	7.1		
Hepatic diseases	4	5.7		
Cardiac diseases	2	2.9		
• DM (hyperglycemia)	1	1.4		
Toxicological	33	47.1		
• Phosphide toxicity (AlP or ZnP)	18	25.7		
Addiction overdose	12	17.1		
Unknown ingestion	3	4.3		

Table (2): Frequency distribution of categorial causes of death (Traumatic, Pathological and Toxicological) of the autopsied cases in the current study (n=70).

n: number, %: percentage, FFH: Fall from height, RTA: road traffic accident, ARF : Acute renal failure, DM: Diabetes mellitus, AlP: Aluminum phosphide, ZnP: Zinc phosphide

As shown in (Table 3); Traumatic group had significantly higher postmortem VH levels of (Na, K and Mg) in line with significantly lower VH glucose compared to Pathological group. In addition, VH glucose levels were significantly higher in Toxicological group compared to Traumatic group. Moreover, Toxicological group had significantly higher VH Na and VH Mg levels as well as significant lower VH glucose levels compared to pathological group.

Variables	Traumatic [n=25]	Pathological [n=12]	Toxicological [n=33]	Test of Significance
VH Na (mEq/L) Mean ± SD	132.94 ± 11.26	117.68 ± 27.87	140.88 ± 13.23	F= 9.330 P < 0.001* P1= 0.023* P2= 0.156 P3 < 0.001*
VH K (mEq/L) Mean ± SD	15.66 ± 4.79	10.30 ± 1.99	13.50 ± 4.90	F = 5.808 $P = 0.005*$ $P1= 0.003*$ $P2= 0.176$ $P3=0.096$
VH Mg (mg/dL) Median (Range)	2.6 (0.6 - 6.5)	2.1 (1.5-2.5)	2.5 (0.8 – 3.7)	KW= 7.381 P= 0.025* P1= 0.018* P2= 0.614 P3=0.011*
VH Glucose (mg/dL) Median (Range)	6 (2 – 348)	23 (4 – 59)	11 (3 – 106)	KW= 14.149 P= 0.001* P1= 0.004* P2= 0.003* P3=0.045*

Table (3): Comparing the postmortem	VH Levels of Na,	K, Mg and	Glucose between	different
causes of deaths in the curren	it study (n=70).			

F= One-way ANOVA test, KW: Kruskal Wallis test, VH: vitreous humour Na: sodium, K: potassium, Mg: magnesium N: number SD: standard deviation mg/dL: Milligrams / decilitre, mEq/L: Milliequivalents / litre, *: Statistically significant (p < 0.05)

P1: Comparison between traumatic and pathological, P2: Comparison between traumatic and toxicological, P3: Comparison between pathological and toxicological

As shown in (Table 4), there was significant moderate negative correlation between VH K levels and the age of the study population (rs = -0.327, P= 0.006^{*}). In addition, there was a significant week

negative correlation between VH K levels and VH glucose levels (rs= -0.286, P= 0.016 *). No other statistically significant correlations were reported among the tested variables

		Age (year)	VH Na (mEq/L)	VHK (mEq/L)	VH Mg (mg/dL)	VH Glucose (mg/dL)
	rs		-0.057	-0.327	-0.234	0.201
Age (Year)	Р		0.639	0.006*	0.051	0.095
VH Na	rs	-0.057		-0.061	0.183	-0.039
(mEq/L)	Р	0.639		0.617	0.129	0.748
VH K	rs	-0.327	-0.061		0.162	-0.286
(mEq/L)	Р	0.006*	0.617		0.179	0.016*
VH Mg	rs	-0.234	0.183	0.162		0.015
(mg/dL)	Р	0.051	0.129	0.179		0.900
VH Glucose	rs	0.201	-0.039	-0.286	0.015	
(mg/dL)	Р	0.095	0.748	0.016*	0.900	

Table (4): The correlations between the measured postmortem	No. W.
the age of studied population $(n=70)$.	

rs: Spearman's correlation except in correlation between (Na, K, and between each of them and age

rs: Pearsons correlation),* : Statistically significant (p < 0.05)

Na: Sodium, K: potassium, Mg: Magnesium, mg/dL : Milligrams / deciliter, mEq/L: Milliequivalents / liter

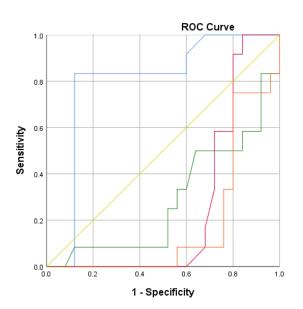
As illustrated in (Table 5); variables enrolled in ROC analysis are those which showed statistically significant differences between different causes of death (traumatic, pathological and toxicological). The cut-off value of each variable, which had the maximum sensitivity, specificity and accuracy differentiate between groups, was to determined. The VH glucose had the largest AUC (0.797) and achieved the highest accuracy (84.6 %) in differentiating between Traumatic and Pathological causes of deaths; followed by VHK, VH Mg and VH Na with accuracies (82.6 %, 76.8 % and 72.8 %) respectively, as shown in figure (1). Moreover, VH glucose achieved accuracy of (74.2 %) in differentiating between Traumatic and Toxicological deaths, as shown in figure (2). Also, figure (3) illustrated that VH glucose had the highest accuracy (76.2 %) in discriminating Pathological from Toxicological deaths followed by, VH Na (70.8 %) and VH Mg (64.2 %).

Diagnostic criteria	Differentiating Traumatic from pathological				Differentiating Traumatic from Toxicological	Differentiating Pathological from toxicologic		-
	VH Na	VHK	VH Mg	VH glucose	VH glucose	VH Na	VH Mg	VH glucose
AUC	0.712	0.817	0.743	0.797	0.732	0.821	0.750	0.697
Crut off restruct	>136.2	> 12.3	>2.45	< 17.5	> 7.5	> 134.5	> 2.15 For	< 20.5
Cut off point	For Tr	For Tr	For Tr	For Tr	For Tx	For Tx	Tx	For Tx
Sensitivity	91.7 %	91.7 %	91.7 %	83.3 %	81.8 %	75.8 %	75.8 %	78.8 %
Specificity	52 %	76 %	68 %	88 %	60 %	66.7 %	58.3 %	75 %
NPV	58.4 %	78.8 %	72.2 %	86.2 %	64.2 %	63.6 %	60.2 %	74.2 %
PPV	86.4 %	86.2 %	86.4 %	80.2 %	78.4 %	77.2 %	78.4 %	72.6%
Accuracy	72.8 %	82.6 %	76.8 %	84.6 %	74.2%	70.8 %	64.2 %	76.2 %
Р	0.039*	0.002*	0.018*	0.004*	0.003*	0.001*	0.011*	0.045*

Table (5): The ROC analysis of (VH Na, K, Mg and glucose) levels to differentiate between causes of death in the current study.

AUC: area under the curve. NPV: Negative predictive value PPV: Positive predictive value

*: significant p value (< 0.05). VH: vitreous humor Na: sodium K: potassium Mg: magnesium, n: number, Tr: Traumatic, Tx: toxicological





Reference Line

Fig. (1): The ROC curve showing sensitivity, specificity and area under the curve for (VH Na, K, Mg and glucose levels) differentiating between traumatic deaths (n= 25) and pathological deaths (n= 12) in the current study.

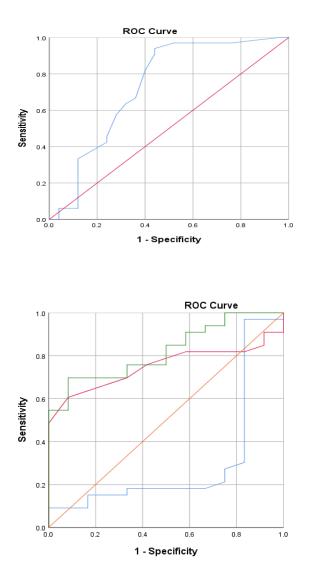


Fig. (2): The ROC curve showing sensitivity, specificity and area under the curve for glucose levels differentiating between traumatic deaths (n=25) and toxicological deaths (n=33) in the current study



Fig. (3): The ROC curve showing sensitivity, specificity and area under the curve for (VH Na, Mg and glucose levels) differentiating between toxicological deaths (n=33) and pathological deaths (n=12) in the current study.

Discussion

The VH is a sterile ideal sample for postmortem analysis (Pigaiani et al., 2020). Relatively few studies concerned the VH chemical analysis in discriminating causes of death (Tse et al., 2018; Kurup et al.,2023). The current study aimed to investigate the pattern of VH postmortem levels of Na, K, Mg and glucose in different causes of death.

In accordance with (Lee et al., 2022; Mahajan et al., 2023; Mezzini et al., 2023) the

traumatic deaths showed current hyponatremia (132.94 \pm 11.26 mEq/L) in relation to antemortem physiological Na serum levels (135 - 145)mEq/L) and antemortem normal VH Na levels (145.4-148.0 mEq/L) (Pigaiani et al., 2020). In massive head trauma, hyponatremia is due to pituitary dysfunction with subsequent ADH (antidiuretic increased hormone) secretion in а condition known as Inappropriate Antidiuretic Hormone Secretion (SIADH) syndrome (Mahajan et al., 2023). On the other hand; Zilg et al. (2016) reported postmortem VH hypernatremia in severe head trauma deaths (n=22).Tudor and Thompson assumed hypernatremia in head (2019)injuries to impaired ADH secreation due to massive brain oedema: in a condition simulating central neurogenic diabetes insipidus, which manifested by water diuresis and sodium retention. Drowning in fresh water is one of the most common causes of postmortem hyponatremia. **B**vard and Summersides (2011) as well as Zilg et al. (2016) reported VH hyponatremia in fresh water drowning with levels (mean= $129.8 \pm$ 17 mM and median 129 mmol/L): respectively. Accordingly, and with comparable results, the present VH Na levels of the two cases of fresh water drowning in the Traumatic group were (128 mEq/L and 131 mEq/L).

In the current Toxicological group, the VH Na was within normal serum and vitreous Na concentrations (140.88 \pm 13.23 mEq/L). Also, Dorooshi et al. (2021) did not report significant changes in antemortem serum Na levels (141.05 \pm 4.29 mEq/L) in 24 cases that died from aluminum phosphide toxicity. Contradictory to our results; some toxicological cases showed hyponatremia as 3,4-Methylenedioxymethamphetamine

(MDMA) (ecstasy) use toxicity (Faria et al., 2020), water intoxication and beer potomania (Windpessl et al., 2017). Besides; hypernatremia was recorded in children abused with salt intoxication as a punishment or due to a Munchhausen syndrome by proxy (Silahlı et al., 2022).

The present VH Na levels were significantly higher in Toxicological and Traumatic groups compared to Pathological group. The current postmortem VH hyponatremia (117.68 ± 27.87 mEq/L) in the pathological group, could be explained by antemortem vasopressin release due to portal hypertension (Goudsmit et al.. 2021). mineralocorticoid deficiency and the use of

diuretics in deaths due to renal diseases (Canaud et al., 2019). Hyponatremia in deaths due cardiac diseases; could be assumed to antemortem fluid over load and renal impairment (Verbrugge et al., 2020). In diabetic deaths; antemortem polyuria and renal dysfunction lead to hyponatremia (Eshetu et al., 2023). In partial agreement with us, Zilg et al. (2016) reported VH hyponatremia community -acquired in pneumonia deaths (n= 13), while as VH hypernatremia was found in deaths due to chronic diseases as cardiac diseases and diabetes (n=17). The difference from our results could be attributed to antemortem dehydration that can accompany the end stage of chronic diseases.

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Our results showed postmortem VH hyperkalemia in traumatic, pathological and toxicological groups (15.66 \pm 4.79 mEq/L; $10.30 \pm 1.99 \text{ mEq/L}$ and $13.50 \pm 4.90 \text{ mEq}$ respectively); in relation to the antemortem normal K serum (3.5 to 5.2 mEq/L) and antemortem normal viterous levels K (4.4–7.8 mEq/L) (Pigaiani et al., 2020). The present postmortem VH hyperkalemia is due to systemic extracellular shift of K due to Na/K pump stoppage (Abdelaal et al., 2023) as well as the local diffusion of K from retinal cells to VH after death (Haryo et al., 2023). The postmortem rise of VH K levels continues with time after death until the equilibration occurs with the plasma K levels (El Sawaf et al., 2019). Interestingly, the Traumatic deaths in the current work had significantly higher postmortem VH K levels compared to the Pathological deaths. In consistent with our results; Zilg et al. (2015) found no obvious differences in VH K levels between various causes of death, such as intoxications, drownings, instant traumatic death, massive skeletal trauma with delayed death, kidney failure and diabetic coma. This discripancy may be due to difference in postmortem intervals of VH sampling as well as different sample size of the study populations.

In the current work; the median values of postmortem VH Mg in traumatic, pathological and toxicological groups were: (2.6 mg/dL, 2.1 mg/dL and 2.5 mg/dL, respectively), which were within ranges of the normal antemortem viterous Mg level (1.48-3.23 mg/dL) and to some extent slightly higher in traumatic and toxicological groups than the normal antemortem total serum Mg (1.82 to 2.31 mg/dL) (Kokavec et al., 2016; Pigaiani et al., 2020). Similar to the current findings and with comparable values; Tse et al. (2018) reported that mean value of postmortem VH Mg level at (1.03 mmol/L) which was equevilant to (2.51 mg/dL) with a PMI ranging from (5–75 h) in a retrospective study on total 40 cases (20 cases diabetics and 20 non-diabetic adult deaths). Kokavec et al. (2016) indicated no considerable change in postmortem VH Mg levels compared to antemortem levels. Also; Zhu et al. (2005) did not found significant postmortem timedependent rise in serum Mg autopsy cases (total, n = 360; 5–48 h postmortem). On the other hand; Li et al. (2009) found mild postmortem time-dependent higher Mg level in pericardial fluid of serial autopsy cases of adults within 48 hour postmortem. The reported increased postmortem Mg levels, could be explained by the extracellular release of Mg though the altered memebranes of the hemolysed anoxic erythrocytes into the plasma (Mihailovic et al., 2014). The controversial evalution of postmortem Mg levels, either hypermagnesmia or within normal ranges, could be related to PMI variations in different studies as anoxic rapture of erythrocytes occurs at (48–72 hour) not immediately after death (Nicák et al., 1999)

Moreover, in the present results both Traumatic and Toxicological groups had significantly higher postmortem VH Mg levels in comparison to Pathological group. Accordingly; significant higher postmortem Mg levels were measured in pericardial fluid (Li et al., 2009) and cardiac blood (Zhu et al., in cases of salt water drowning 2005) compared to other causes of death. Besides; deaths due to hyperthermia, intoxaction and sharp instrument injury showed a higher precardial Mg levels compared to blunt injury, asphyxiation, freshwater drowning, fire fitalitity, hypothermia, acute cardiac death, pneumonia and spontaneous cerebral hemorrhage (PMI 5-48 hour) (Li et al., 2009). Additionally; asphyxiation and fatal methamphetamine intoxication had higher cardaic Mg levels compared to blunt injury, sharp injury, freshwater drowning, fire fatalities, delayed death from traumas and acute myocardial infarction groups (PMI 48 hour) (Zhu et al., 2005).

The physiological antemortem blood glucose level is ranged between (63 to 99 mg/dL) (Pigaiani et al., 2020); while as the VH glucose normal antemortem level represents about 85% of blood values and ranged between (2.08-2.5 mmol/L equivalent to 36 to 50 mg/dl) (Kokavec et al., 2016). Current findings agreed with (Zilg et al., 2009) who reported a drop in the postmortem VH glucose concentrations compared to the normal antemortem values in blood and VH; as the median values of the present postmortem VH glucose in Traumatic, Pathological and toxicological groups were: (6 mg/dL, 23 mg/dL and 11 mg/dL),respectively. The postmortem drop of VH glucose could be explained by postmortem glycolysis (Pigaiani et al., 2020). In addition, hypoglycemia in traumatic cases could be due to depletion of glucose stores that follow initial induced hyperglycemia stress (Quintana-Pajaro et al., 2023).

In present work, the Pathological group had significantly higher VH glucose levels than both Traumatic and Toxicological groups. The Toxicological group had a significantly higher VH glucose level than Traumatic group. Similarly but with incomparable values, Osuna et al. (2001) demonstrated significantly higher VH glucose levels (116.2± 27.3 mg/dL) in pathological compared to traumatic deaths deaths (36.0±8.9 mg/dL). The difference in postmortem VH glucose levels, particularly in diabetic cases, assumed to slow gradual drop of VH glucose in the diabetic cadaver (Belsey and Flanagan, 2016).

Regarding the correlations between the investigated postmortem VH variables and the age of the study population, current results showed a significant moderate negative correlation between VH K levels and age. The decline of VH K level with increased age in adults could be explianed by the decreased number of retinal cells with age (Harman et al., 2020). Contradictory to our findings; Zilg et al. (2015) reported increased VH K level from infancy to old age in 462 autopsied cases with postmortem intervals (2 h - 17 days). On the other hand; (Chandrakanth et al., 2013; Hassan and El-Dakroory, 2013; Kurup et al., 2023) demonestrated that age and sex had no bearing on the postmortem VH K levels. These differences can be assumed to different PMI and different age groups in each study. In accordance with Chandrakanth et al. (2013), no statistically significant correlation, between postmortem VH Na levels and age, was reported in the present result. Similarly; Tse et al. (2018) declared absent correlation between postmortem VH Mg level and age, PMI, other VH electrolytes (Na, K and chloride) or sex. the present Moreover. results showed significant week negative correlation between VH K levels and VH glucose levels. In partial agreement; Murad et al., (2021) reported high

VH glucose levels with low K levels <10 mmol/L in early deaths, while as no significant decrease in VH glucose levels when the potassium concentration increased to >10 mmol/L with increased PMI (2-3 days). The fact that explained by sudden steeper drop in VH glucose immediately after death. This difference may be due to different PMI of VH sampling.

For further analysis of the current results, ROC curve analysis was performed for VH variables that showed significant differences between different causes of death. The VH glucose achieved the highest accuracy (84.6 %) in differentiating between Traumatic and Pathological causes of deaths; followed by VHK, VH Mg and VH Na with accuracies (82.6 %, 76.8 % and 72.8 %) respectively. The VH glucose had the highest %) accuracy (76.2)in discriminating Toxicological Pathological from deaths followed by VH Na (70.8 %) and VH Mg (64.2 %). Moreover, VH glucose achieved accuracy of (74.2 %) in differentiating between Traumatic and Toxicological death. Few studies concerned the ROC analysis of VH components for differentiation between causes of death. Osuna et al. (2001) reported that VH glucose achieved (AUC 0.814) in differentiating between diabetic and nondiabetic deaths. while as the best discrimination achieved by ROC analysis of sum values of both VH glucose and VH lactate. Also, Tse et al. (2018) found that ROC analysis of VH Mg could discriminate diabetic and non-diabetic deaths with (AUC 0.65, Sensitivity 0.35 and specificity of 0.95). Garland et al. (2016) elucidated that postmortem VH Na achieved cut off (136 mmol/L) with (sensitivity = 0.8; specificity = 0.8) in discriminating drowning in salt water from other causes of death, while as; ROC analysis of sum values of VH Na and VH chloride levels achieved higher accuracy and larger AUC.

To conclude; the present study investigated the postmortem levels of VH electrolytes and glucose in different causes of death declaring that there were differences traumatic. Pathological between and Toxicological groups. In addition, there were negative correlations between VH K and age as well as between VHK and VH glucose. The ROC analysis revealed that VH glucose was the best variable which achieved highest accuracy in differentiating between all groups. Our study was limited by absence of cases with undetermined causes of death as well as the effect of PMI was not investigated. Further investigation involving effects of PMI and including cases with unexplained death is highly recommended. In addition, the ROC curve analysis of sum values of VH variables is advisable.

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Conflict of Interest:

The authors declare no conflicts of interest as well as the current work did not receive any organization or financial support.

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عمرو محد حسن¹ و عبد العزيز غانم¹ و هند محمود أبو العطا¹ و أحمد حسن² و هبة الله عبد الرحمن¹ ¹: قسم الطب الشرعي والسموم الإكلينيكية - كلية طب- جامعة المنصورة- مصر ²: قسم الباثولوجيا الاكلينيكية كلية طب- جامعة المنصورة-مصر

الجسم الزجاجي للعين هو عينة معقمة يسهل الوصول إليها ولهذا فهو ذو قيمة عالية في التحاليل الكيميائية ما بعد الوفاة. عدد قليل نسبيا من الدر اسات المعنية بتحليل الجسم الزجاجي بعد الوفاة في التمييز بين أسباب الوفاة. تهدف الدراسة المستقبلية المقطعية الحالية إلى تقدير مستويات الصوديوم ، والبوتاسيوم ، والمغنيسيوم ، والجلوكوز في الجسم الزجاجي بعد الوفاة كمتغيرات تمييزية بين أسباب الوفاة (الاصابية ، والمرضية، والسمية). في العمل الحالي، تم الحصول على عينات الجسم الزجاجي من حالات تشريح البالغين (العدد = 70) مع فاصل زمني بعد الوفاة أقل من 24 ساعة وبعد استبعاد امرض العين أوالاصابات في العين . كان لدى مجموعة الوفاة الناتجة عن الاصابات مستويات أعلى من الصوديوم ، والبوتاسيوم ، والمغنيسيوم ، وذلك تمشيا مع انخفاض ملحوظ في الجلوكوز مقارنة بمجموعة الوفيات المرضية. كانت مستويات الجلوكوز أعلى بشكل ملحوظ في مجموعة الوفيات السمية مقارنة بمجموعة الوفيات الاصابية. كان لدى مجموعة الوفيات السمية مستويات أعلى من الصوديوم والمغنيسيوم بالإضافة إلى مستويات أقل من الجلوكوز مقارنة بمجموعة الوفيات المرضية. حقق نسب الجلوكوزفي الجسم الزجاجي أعلى دقة (84.6٪) في تحليل منحني خاصية تشغيل المستقبل (ROC) للتمييز بين الأسباب الاصابية والمرضية للوفيات؛ يليه البوتاسيوم بنسبة (82.6%). كما كان الجلوكوز أعلى دقة (76.2%) في التمييز بين الوفيات المرضية والسمية يليه الصوديم (70.8%). علاوة على ذلك، حقق الجلوكوز دقة قدرها (74.2%) في التمييز بين الوفيات الاصابية والسمية. في الختام، كانت مستويات إلكتروليتات الجسم الزجاجي ومستويات الجلوكوز بعد الوفاة مختلفة وفقًا لأسباب الوفاة المختلفة و من ثم يمكن استخدامها كأدوات مساعدة في الاشتباه في سبب الوفاة وخاصبة مستوى الجلوكوزفي الجسم الزجاجي .