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ORIGINAL PAPER

The postmortem distribution of ketone bodies between blood, vitreous humor, spinal fluid, and urine

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Abstract The distribution of the ketone bodies: acetone, acetoacetate, and D-β-hydroxybutyrate, between blood, vitreous humor, spinal fluid, and urine was examined in 105 medico-legal autopsies. The ketone body concentration in the body fluids was determinated by head-space gas chromatography. The correlation between blood and the body fluids could be described with regression lines on the logarithmic-transformed results. The correlation is dependent on the ketone body concentration. The ketone bodies in spinal fluid show the best correlation to blood, followed by vitreous humor, and last urine. The concentration dependence in spinal fluid is mainly due to ketone bodies being metabolized in the brain. The human brain utilizes ketone bodies during normal nutritional state. In vitreous humor, the dependence is mainly due to protein bindings of acetoacetate and β-hydroxybutyrate in blood and the difference in dry matter between blood and vitreous humor.

Keywords Postmortem distribution · Ketone bodies · Blood · Spinal fluid · Vitreous humor · Urine · Metabolism

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Introduction

In forensic medicine, the ketone body level in blood is an important tool in the effort to uncover if the cause of death is ketoacidosis of diabetic or alcoholic origin [1].

Ketone bodies are produced in the liver, mainly from oxidation of fatty acids, and are exported to peripheral tissues for use as an energy source. The ketone bodies are acetoacetate, D-β-hydroxybutyrate, and acetone. Acetone originates from acetoacetate, because acetoacetate, partly spontaneously, decarboxylate to acetone and carbon dioxide. D-β-hydroxybutyrate is henceforward called β-hydroxybutyrate.

At autopsies blood may be difficult to collect, and the use of other body fluids for the analysis of ketone bodies may therefore be needed. Most of the reference values are related to blood. It would be useful to be able to estimate a ketone body level from knowledge of the distribution of ketone bodies between blood and the body fluids.

The few existing studies [2, 3], however, have not been in agreement as to the possible use of other body fluids than blood for the analysis.

We have examined the postmortem distribution of acetoacetate and β-hydroxybutyrate between blood and vitreous humor, spinal fluid and urine, respectively.

Material and methods

Blood, vitreous humor, spinal fluid, and urine were taken at 105 medico-legal autopsies without or only with slight putrefaction, comprising 30 females and 75 males. The causes of death were: poisoning 29, including 6 cases of alcohol poisoning, natural death 26, traumatic injuries 33, alcoholic ketoacidosis 1, diabetic coma 1, others 9, and

unknown 6. Blood samples were obtained from extremity veins. In 53 cases, the ketone bodies were measured in all four body fluids, in the remaining 52 cases the ketone bodies were measured in blood and in two other body fluids. The specimens were taken in polycarbonate tubes containing sodium fluoride and stored at −25 °C until analysis. Measurement of acetone, acetoacetate, and β-hydroxybutyrate in blood, vitreous humor, spinal fluid, and urine was done by head-space gas chromatography [4]. The measurement of ketone bodies is based on enzymatic transfer of β-hydroxybutyrate to acetoacetate and subsequent decarboxylation of acetoacetate into acetone. Acetone is subsequently analyzed by head-space gas chromatography [4]. The procedure is briefly as follows. Two head space vials are prepared for analysis. In the first vial acetoacetate in the sample is decarboxylated to acetone, by heating it 1 h at 100 °C. The sum of acetone and acetoacetate is measured. In the second vial β-hydroxybutyrate, in the sample, is first reduced to acetoacetate by β-hydroxybutyrate dehydrogenase at 37 °C and then the decarboxylation is taking place as described. The sum of acetone, acetoacetate and β-hydroxybutyrate, the total of ketone bodies is then measured. The amount of β-hydroxybutyrate is the difference between the amount of total ketone body and the amount of acetone coming from acetoacetate and acetone in the sample. The head-space gas chromatograph used was a Perkin–Elmer model HS-8500 equipped with a flame ionization detector. Instrument settings were: Column DBWAX, 30-m long, i.d. 0.5 mm, film thickness 1 μ, oven temperature 60 °C, carrier gas nitrogen. The decarboxylation of acetoacetate was done by thermostating the head space vial at 100 °C for 1 h. The relative standard deviation for the acetoacetate analysis was 14.8–5.9%, in the concentration interval 25–100 μmol and for β-hydroxybutyrate 3.1–2.0%, in the interval 75–300 μmol. The method requires a sample of at least 2 ml, which is not always at disposal with reference to vitreous humor. Therefore, it was, some times, necessary to reduce the requirement for material. This was done by using 4-ml head space vials instead of 22 ml vials. In order to make the small 4-ml head space vials fit into the head space auto sampler carousel (Perkin–Elmer HS-101) they were placed in bored aluminum cylinders of the same size as the 22-ml head space vials with which the auto sampler is constructed. The sample size was then reduced to at least 0.2 ml. As acetone is derived from the decarboxylation of acetoacetate, the measured sum of acetone + acetoacetate is just named acetoacetate.

Results

Table 1 shows the summary data for acetoacetate, β-hydroxybutyrate, and total ketone bodies. The table shows that the ketone bodies are present in all the fluids, except in urine, where β-hydroxybutyrate was absent in 11 of the 75 urine samples. The median, the range, and the quartile range are to some extent dependent on the type of ketone body and body fluid. The median of acetoacetate has its minimum value in vitreous humor, whereas the median value of β-hydroxybutyrate has its maximum value in the same fluid. The median of the total ketone bodies was, except for urine, almost the same in the body fluids.

For all samples there was a skewed negative distribution. After logarithmic transformation ($\log_{10}$) the results showed an approximate normal distribution.

To study the correlation of the concentration of acetoacetate, total ketone bodies and β-hydroxybutyrate between blood and the other fluids a regression analysis was performed on the logarithmically transformed results [5]. Acetoacetate, total ketone bodies, or β-hydroxybutyrate concentrations in blood was the response variable (y) and acetoacetate, total ketone bodies or β-hydroxybutyrate in one of the other fluids was the explanatory variables (x). Table 2 shows the summary data of the regression analyses. Ketone bodies in spinal fluid had the best correlation to blood, acetoacetate ($R^2 = 0.94$, $s = 0.1152$), ß-hydroxybuty-

Table 1 Summary data for ketone bodies (μmol/L) in blood, spinal fluid, vitreous humor, and urine

	Acetoacetate				Total ketone bodies				β-hydroxybutyrate			
	Blood	Spinal fluid	Vitreous humor	Urine	Blood	Spinal fluid	Vitreous humor	Urine	Blood	Spinal fluid	Vitreous humor	Urine
Mean	109	134	97	348	285	256	360	712	176	122	263	364
Median	40	39	29	49	101	107	118	67	55	58	73	24
Range	10–1,173	10–1,621	6–975	8–7,986	31–3,831	36–2,653	25–5,753	14–13,719	11–3,001	14–1,533	7–5,015	0–5,733
Q^1[a]	23	22	15	28	66	64	71	43	33	37	48	5
Q^3[a]	85	109	77	139	201	216	245	209	115	104	166	102
N[b]	105	93	96	75	105	93	96	75	105	93	96	75

[a] Q^1 and Q^3 = quartiles

[b] N = number of samples

Table 2 Summary of logarithmic regression analysis

Parameter	Estimate	Standard deviation	*t*-test	Significance-probability
Acetoacetate				
Blood/spinal fluid				
Log(α)	0.26340	0.04111	6.41	<0.001
β_1	0.86269	0.02280	37.82	<0.001
$R^2 = 0.94$ $s = 0.1152$				
Blood/vitreous humor				
Log(α)	0.38446	0.04273	9.00	<0.001
β_1	0.81748	0.02542	32.16	<0.001
$R^2 = 0.92$ $s = 0.1307$				
Blood/urine				
Log(α)	0.58458	0.08428	6.94	<0.001
β_1	0.61760	0.04362	14.16	<0.001
$R^2 = 0.73$ $s = 0.2386$				
Total ketone bodies				
Blood/spinal fluid				
Log(α)	−0.13739	0.05979	−2.30	0.024
β_1	1.07242	0.02751	38.98	<0.001
$R^2 = 0.94$ $s = 0.1127$				
Blood/vitreous humor				
Log(α)	0.21951	0.06940	3.16	0.002
β_1	0.86870	0.03111	27.92	<0.001
$R^2 = 0.89$ $s = 0.1493$				
Blood/urine				
Log(α)	0.86127	0.08305	10.37	<0.001
β_1	0.61617	0.03821	16.13	<0.001
$R^2 = 0.78$ $s = 0.2223$				
β-hydroxybutyrate				
Blood/spinal fluid				
Log(α)	−0.39046	0.08926	−4.37	<0.001
β_1	1.22068	0.04720	25.86	<0.001
$R^2 = 0.88$ $s = 0.1834$				
Blood/vitreous humor				
Log(α)	0.10810	0.10580	1.02	0.309
β_1	0.86504	0.05101	16.96	<0.001
$R^2 = 0.75$ $s = 0.2540$				
Blood/urine				
Log(α)	1.15619	0.09223	12.54	<0.001
β_1	0.46782	0.04917	9.51	<0.001
$R^2 = 0.60$ $s = 0.3394$				

α = the point where the line crosses the y-axis

β = slope of the line

R^2 = coefficient of determination

s = standard deviation of the points about the line

rate ($R^2 = 0.88$, $s = 0.1834$), total ketone bodies ($R^2 = 0.94$, $s = 0.1127$), followed by vitreous humor, acetoacetate ($R^2 = 0.92$, $s = 0.1307$), β-hydroxybutyrate ($R^2 = 0.75$, $s = 0.2540$), total ketone bodies ($R^2 = 0.89$, $s = 0.1493$) and last urine, acetoacetate ($R^2 = 0.73$, $s = 0.2386$), β-hydroxybutyrate ($R^2 = 0.60$, $s = 0.3394$), total ketone bodies ($R^2 = 0.78$, $s = 0.2223$). Figure 1 shows the regression line for the correlation blood/spinal fluid for total ketone body

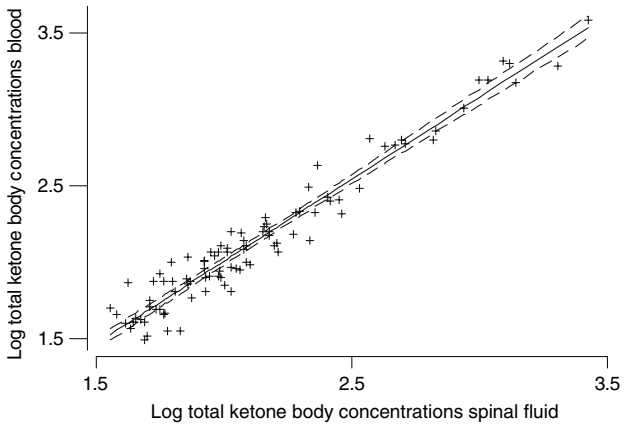

Fig. 1 The relation between the total ketone body concentration in blood and spinal fluid with 95% cofidence intervals

concentrations and the 95% confidence interval. The standard deviations around the correlation lines of β-hydroxybutyrate were almost twice the standard deviations of the other lines (acetoacetate, total ketone bodies). This is because the results of β-hydroxybutyrate appear as the difference between the results of total ketone body analysis and the acetoacetate analysis. β-Hydroxybutyrate concentrations below 30 µmol in vitreous humor show a lack of fits regarding the regression line. The table also shows that the relations are concentration dependent. For acetoacetate the relations decrease for all three fluids, with increasing concentrations, the slope of the lines is less than 1. For β-hydroxybutyrate the relation for vitreous humor is also decreasing with increasing concentration, the slope of the line is <1, but for spinal fluid, it is the opposite. The slope is >1. The slope for the total ketone body shows that the relation for spinal fluid is increasing, while the relations for vitreous humor and for urine are decreasing with increasing concentrations.

The table is to be used in the following way:

The equation of the regression line is $\log(y) = \log(\alpha) + \beta_1 \log(x)$ [3].We have for example a measured concentration of the total ketone body in spinal fluid of 500 µmol. What is the expected total ketone body concentration in blood? It is (Table 2, total ketone body): $\log(y) = -0.13739 + 1.07242 * \log(500) = 2.75704$ equal to $y = 10^{2.75704}$ or 572 µmol. Then the relation y/x, in casu the relation blood/spinal fluid, are 572/500 = 1.14.

In order to get a more general view the relations for increasing concentrations are tabulated in Table 3A–C. The relations in Tables 3A–C are calculated by means of the regression line equations (Table 2) for concentrations from 25 µmol to 1,500 µmol of acetoacetate, total ketone bodies, and β-hydroxybutyrate. The highest concentration of acetoacetate in vitreous humor is 975 µmol, therefore the relation is only calculated to 1,000 µmol. For acetoacetate (Table 3A), the relations decrease, spinal fluid 42%, vitreous humor 50%, urine 82%. For β-hydroxybutyrate (Table 3C), the relation of vitreous humor decreases 38%, urine 88%. However, for spinal fluid it is the opposite. The spinal fluid relation increases 150% when the β-hydroxybutyrate concentration increases from 25 µmol to 1,500 µmol. For the total ketone bodies, Table 3B, the spinal fluid relation increases 33% with the concentration. The influence of β-hydroxybutyrate overshadows the influence of acetoacetate on the total ketone body relation in spinal fluid. Vitreous humor decreased 45% and urine 81%.

Discussion

Only two reports concerning the postmortem distribution of ketone bodies between different body fluids were found. In the first study on 105 medico-legal autopsies, the distribution of ketone bodies between blood, vitreous humor, and pericardial fluid were studied [2]. The study found a strong correlation between blood and vitreous humor ketone body levels, which is in agreement with our results. In the other study [3] consisting of 30 medico-legal autopsies, the concentrations of β-hydroxybutyrate in blood, vitreous humor, spinal fluid, and urine were determined. The study found, in contrast to ours, that the levels of β-hydroxybutyrate in blood and in vitreous humor were of the same size. They found, in agreement with our results, that the levels in spinal fluid were lower than the levels in vitreous humor. The conclusion was that the β-hydroxybutyrate levels in spinal fluid could not be used to diagnose ketoacidosis, which is in disagreement with our conclusion.

Why are the blood/body fluid concentrations correlated? Postmortem changes of the levels of the ketone bodies are not the explanation, because it has been shown [1] that there is no correlation ($r = 0.031$–0.083) between the ketone body concentration and the postmortem interval. The main reason for the changes is more likely to be found in parameters such as transport mechanisms through the barriers of blood-brain, blood-spinal fluid and blood-vitreous humor, protein bindings, dry matter content, and in the metabolism of ketone bodies in the brain and in the eye, respectively.

The blood-brain and blood-spinal fluid barrier is in this context the same barrier and will henceforward be called blood-brain barrier. The blood-brain barrier is composed of endothelial cells of the microvasculature, which possesses some unique features such as tight intercellular junctions and a number of mechanisms whereby the endothelial cells can control solute fluxes across the capillary wall [6].

The ketone bodies acetone, acetoacetate, and β-hydroxybutyrate pass the blood-brain barrier by simple diffusion, and as monocarboxylic acids, acetoacetate and

Table 3 (A) Acetoacetate (μmol/L)—The relation dependence of the acetoacetate concentration, (B) Total ketone bodies (μmol/L)—The relation dependence of the total ketone body concentration, (C) β-hydroxybutyrate (μmol/L)—The relation dependence of β-hydroxybutyrate concentration

Measured	Blood/ spinal fluid	Lower	Upper	Blood/ vitreous humor	Lower	Upper	Blood/ urine	Lower	Upper
(A)									
25	1.2	1.1	1.3	1.4	1.3	1.4	1.1	1.0	1.3
50	1.1	1.0	1.1	1.2	1.1	1.3	0.9	0.8	1.0
100	1.0	0.9	1.0	1.1	1.0	1.1	0.7	0.6	0.8
200	0.9	0.8	1.0	0.9	0.8	1.0	0.5	0.4	0.6
300	0.8	0.8	0.9	0.9	0.8	1.0	0.4	0.4	0.5
400	0.8	0.7	0.9	0.8	0.7	0.9	0.4	0.3	0.5
500	0.8	0.7	0.9	0.8	0.7	0.9	0.4	0.3	0.4
600	0.8	0.7	0.9	0.8	0.7	0.9	0.3	0.3	0.4
700	0.8	0.7	0.9	0.7	0.6	0.9	0.3	0.3	0.4
800	0.7	0.6	0.8	0.7	0.6	0.8	0.3	0.2	0.4
900	0.7	0.6	0.8	0.7	0.6	0.8	0.3	0.2	0.4
1,000	0.7	0.6	0.8	0.7	0.6	0.8	0.3	0.2	0.4
1,100	0.7	0.6	0.8	–	–	–	0.3	0.2	0.4
1,200	0.7	0.6	0.8	–	–	–	0.3	0.2	0.3
1,300	0.7	0.6	0.8	–	–	–	0.3	0.2	0.3
1,400	0.7	0.6	0.8	–	–	–	0.2	0.2	0.3
1,500	0.7	0.6	0.8	–	–	–	0.2	0.2	0.3
Change	−42%			−50%			−82%		
(B)									
25	0.9	0.8	1.0	1.1	1.0	1.2	2.1	1.8	2.5
50	1.0	0.9	1.0	1.0	0.9	1.1	1.6	1.4	1.9
100	1.0	1.0	1.1	0.9	0.8	1.0	1.2	1.1	1.4
200	1.1	1.0	1.1	0.8	0.8	0.9	1.0	0.8	1.1
300	1.1	1.0	1.2	0.8	0.7	0.9	0.8	0.7	0.9
400	1.1	1.0	1.2	0.8	0.7	0.8	0.7	0.6	0.8
500	1.1	1.1	1.3	0.7	0.7	0.8	0.7	0.6	0.8
600	1.2	1.1	1.3	0.7	0.6	0.8	0.6	0.5	0.7
700	1.2	1.1	1.3	0.7	0.6	0.8	0.6	0.5	0.7
800	1.2	1.1	1.3	0.7	0.6	0.8	0.6	0.5	0.7
900	1.2	1.1	1.3	0.7	0.6	0.8	0.5	0.4	0.6
1,000	1.2	1.1	1.4	0.7	0.6	0.8	0.5	0.4	0.6
1,100	1.2	1.1	1.4	0.7	0.6	0.8	0.5	0.4	0.6
1,200	1.2	1.1	1.4	0.7	0.6	0.8	0.5	0.4	0.6
1,300	1.2	1.1	1.4	0.7	0.6	0.8	0.5	0.4	0.6
1,400	1.2	1.1	1.4	0.6	0.6	0.8	0.5	0.4	0.6
1,500	1.2	1.1	1.4	0.6	0.5	0.7	0.4	0.3	0.6
Change	33%			−45%			−81%		
(C)									
25	0.8	0.7	0.9	0.8	0.7	1.0	2.6	2.1	3.2
50	1.0	0.9	1.1	0.8	0.7	0.9	1.8	1.5	2.2
100	1.1	1.0	1.2	0.7	0.6	0.8	1.2	1.0	1.5
200	1.3	1.2	1.5	0.6	0.6	0.7	0.9	0.7	1.1
300	1.4	1.2	1.7	0.6	0.5	0.7	0.7	0.5	0.9

Table 3 continued

Measured	Blood/ spinal fluid	Lower	Upper	Blood/ vitreous humor	Lower	Upper	Blood/ urine	Lower	Upper
400	1.5	1.3	1.8	0.6	0.5	0.7	0.6	0.4	0.8
500	1.6	1.3	2.0	0.6	0.5	0.7	0.5	0.4	0.7
600	1.7	1.3	2.1	0.5	0.4	0.7	0.5	0.4	0.7
700	1.7	1.4	2.2	0.5	0.4	0.7	0.4	0.3	0.6
800	1.8	1.4	2.3	0.5	0.4	0.7	0.4	0.3	0.6
900	1.8	1.4	2.4	0.5	0.4	0.7	0.4	0.3	0.6
1,000	1.9	1.4	2.4	0.5	0.4	0.7	0.4	0.3	0.5
1,100	1.9	1.5	2.5	0.5	0.4	0.6	0.3	0.2	0.5
1,200	2.0	1.5	2.6	0.5	0.4	0.6	0.3	0.2	0.5
1,300	2.0	1.5	2.6	0.5	0.4	0.6	0.3	0.2	0.5
1,400	2.0	1.5	2.7	0.5	0.4	0.6	0.3	0.2	0.5
1,500	2.0	1.5	2.8	0.5	0.4	0.6	0.3	0.2	0.4
Change	150%			−38%			−88%		

Lower = lower limit of the 95% confidence interval

Upper = upper limit of the 95% confidence interval

β-hydroxybutyrate also pass by facilitated diffusion [7–9]. Both types of diffusion are passive, i.e., energy independent and go from high to low concentration. Studies have shown that the blood-eye barrier in many ways is similar to the blood-brain barrier. The barrier consists of tight endothelial cell junctions. The barrier has carrier-mediated transport mechanisms, including monocarboxylic acids transporters [10–12], and small molecules can pass the barrier by diffusion [13, 14].

The uniform structure of the blood/brain barrier and blood/eye barrier results in small molecules having the same rate of diffusion through the two barriers. The better correlation for blood/spinal fluid may be due to a better flow of spinal fluid through the brain surface than that of vitreous humors through the eye. Spinal fluid is renewed about 3.7 times every 24 h, while the renewal of vitreous humor water takes days [15]. Spinal fluid is better reflecting the actual biochemical situation than vitreous humor. Urine has the lowest correlation to blood. Urine differs from spinal fluid and vitreous humor in being formed by an active regulation on its way from the kidney to the bladder; furthermore the urine in the bladder has often been formed over some time [15].

Acetoacetate: The decrease of the blood/vitreous humor and blood/spinal fluid relations for acetoacetate, Table 3A, is most likely due to protein binding and difference in dry matter content. As an acidic compound acetoacetate is to some extent bound to the proteins, mostly albumin, in blood. At low acetoacetate concentrations, the stronger the affinity between acetoacetate and protein, the smaller the fraction that is free. As the acetoacetate levels increase, a point is reached at which the binding capacity of the protein becomes saturated, and more free acetoacetate becomes available for diffusion [16]. Table 3A shows that the protein binding capacity is reached at a concentration of about 800 µmol acetoacetate.

The free acetoacetate and acetone are equally distributed by diffusion over the water phases of blood and vitreous humor. The water content of blood is normally 78% and of vitreous humor 99% giving the relation 0.8. The dry matter content of spinal fluid must be of the same size. Due to hemoconcentration postmortem, the dry matter content of blood increases giving a relation about 0.7 [14]. The equal distribution on water phases, the amount of protein bound acetoacetate and the postmortem hemoconcentration explains why the relation blood/vitreous humor of acetoacetate is decreasing toward a value of 0.7 (Table 3A) with increasing levels of acetoacetate.

β-Hydroxybutyrate: The explanation for the increasing β-hydroxybutyrate relation blood/spinal fluid may be found in the metabolism of the ketone bodies in the brain (Table 3C). The first step in β-hydroxybutyrate metabolism, in the brain, is the conversion to acetoacetate, from there the metabolic pathway is the same for β-hydroxybutyrate and acetoacetate. Although both substances are metabolized, only the β-hydroxybutyrate level in spinal fluid decreases. This is probably because β-hydroxybutyrate converts to acetoacetate and thereby holds the acetoacetate level almost constant and because acetoacetate is transported through the blood/brain barrier 2–3 times faster than β-hydroxybutyrate. The transport of β-hydroxybutyrate across the blood-brain barrier is competitively inhibited by acetoacetate [17–19]. The metabolic reactions are of first order, i.e., the higher the concentration, the higher the reaction

rate of the ketone body metabolism. Maximum rates are rarely if ever achieved, certainly because the substrates never reach levels high enough to saturate the enzymes. The three factors, metabolism, rate of metabolism, and difference in transport rates through the blood-brain barrier lead to a β-hydroxybutyrate relation blood/spinal fluid that increase with increasing β-hydroxybutyrate concentration to a level of 2.0. Our results thus show that the brain utilizes ketone bodies, not only during starvation, but also during normal nutritional state, which is in agreement with others [20, 21].

The β-hydroxybutyrate relation of blood/vitreous humor is decreasing with increasing concentration to a level of 0.5 (Table 3C). If the decrease is only due to hemoconcentration and difference in dry matter content, the level should be about 0.7. The explanation may be that the β-hydroxybutyrate concentration in vitreous humor is higher than expected. The higher concentration of β-hydroxybutyrate may be due to the equilibrium between acetoacetate and β-hydroxybutyrate, in the presence of β-hydroxybutyrate dehydrogenase in the retina, being displaced a little in favor of β-hydroxybutyrate. This could also explain that β-hydroxybutyrate concentrations below 30 μmol in vitreous humor show a lack of fits to the regression line.

In an earlier study [1] the ketone body cut off values for blood in case of keto acidosis were found. The material in the study consisted of blood specimens from 131 deceased persons, divided into three groups: Group 1: controls, 79 cases of non-alcoholic abusers with a known cause of death, group 2: 35 cases of alcohol abusers with known causes of death, and group 3: 17 cases of alcohol abusers without ascertainable cause of death. The limit value between the controls and the group of alcohol abusers with keto acidosis as cause of death was by binary logistic regression found to be 531 μmol or said in another way the value for a 50% probability of belonging to the group of alcohol abusers with keto acidosis as a cause of death. With a probability of 80% the value is 1,213 μmol. The term "ketoalcoholic death" was suggested as a possibility with 50% probability, when the postmortem blood ketone body concentration in an alcoholic with otherwise unknown cause of death exceed 531 μmol. One must be aware that the magnitude of the cut off values are heavily dependent on how the control group is defined.

Table 4 shows the cut off values, with 95% confidence interval, of spinal fluid, vitreous humor, and urine calculated from the blood values by means of the regression line for total ketone bodies. A simple way to calculate an approximate value of total ketone bodies, acetoacetate or β-hydroxybutyrate in blood from a value in spinal fluid, vitreous humor, or urine is to use the calculated relations of total ketone body, acetoacetate and β-hydroxybutyrate concentrations between blood and body fluids in Table 3A–B.

Conclusion

The correlation between ketone body concentrations in blood and in spinal fluid, vitreous humor and urine are dependent on the concentration level. The results from the analyses of ketone bodies in other fluids than blood may only be used if these correlations are known. The concentration dependence is due to parameters such as postmortem hemoconcentration, protein binding, diffusion through the blood-brain or blood-eye barriers and metabolism of the ketone bodies.

The correlation blood/spinal fluid of β-hydroxybutyrate shows that the human brain utilizes ketone bodies, not only during starvation, but also at normal state of nutrition.

Educational message

1. The reader of the article will learn about the metabolism of ketone bodies and the use of ketone bodies in forensic medicine.
2. He or she will experience an argumentation leading to the conclusion that β-hydroxybutyrate is metabolized as an energy source in the brain.
3. The possible use of spinal fluid, vitreous humor, or urine as a substitute for blood in the diagnosis of alcoholic keto acidosis is described.

Table 4 Cut off values (μmol/L) with increasing probability for keto acidosis

Probability of keto acidosis						
	50%	Lower	Upper	80%	Lower	Upper
Blood	531	–	–	1,213	–	–
Spinal fluid	467	431	506	1,008	900	1,130
Vitreous humor	766	666	882	1,982	1,623	2,422
Urine	1,059	759	1,478	4,045	2,508	6,525

Lower = lower limit of the 95% confidence interval

Upper = upper limit of the 95% confidence interval

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