The Relationship of Alcohol Concentration in Epidural or Acute Sub-dural Hematoma Compared with Vitreous Humor and Femoral Blood

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The study of the relationship between the epidural hematoma or subdural hematoma alcohol concentration (SDHAC) compared with femoral blood (BAC) and vitreous humor alcohol concentration (VHAC). The specimens of 25 corpses (total 888 corpses) were carried out which revealed EDH or SDH, no treatment and the autopsy performed within 24 hours after death in 2006 at the Forensic Medicine Department, Maharaj Nakorn Chiang Mai hospital, Chiang Mai University. All specimens were frozen at -20°C until they were processed with Gas Chromatography Head space (GC-HS).The result showed that the relationship of SDHAC: BAC was better than VHAC:BAC. And SDHAC may have more reliable concentration at time of injury than BAC in the absorption phase prior to equilibrium. However, sub-dural hematoma should be one of the best specimens, as femoral blood and vitreous humor, for alcohol analysis in a corpse who died at the scene, no treatment and no sign of putrefaction.

Keywords: *Epidural hematoma, Subdural hematoma, Vitreous humor, Femoral blood, GC-HS, Corpse, Putrefaction, Forensic Medicine, Maharaj Nakorn Chiang Mai*

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The predominant effect of alcohol is in the brain. High alcohol concentration causes central nervous system depression, judgment disorder, decision ability, impairment of perception and reaction to events. This impairment develops prior to the onset of more overt symptom of alcohol intoxication, such as difficulty in walking or maintaining balance. But, low alcohol concentration depresses inhibitory processes of brain or stimulant effect[1].

Alcohol causes many problems due to its effect especially in traffic injury. The National Highway Traffic Safety Administration (NHTSA) estimated that alcohol was involved in 38% of fatal automobile crashes and 7% of crashes in 2000[1]. The International Center of Alcohol Police showed the difference of the limitation of blood alcohol concentration (BAC) in each country for example; no blood alcohol concentration in drivers in Russia and Romania, 40-50 mg/dl in Europe, 80-100 mg/dl in USA and below 50 mg/dl in Thailand[2,3].

There are 2 common methods which were used to measure alcohol concentration in drivers: the breath analyzer and gas chromatography-headspace (GC-HS).

BAC analysis with GC-HS is the method that directly corroborates to the law. However, the pre-analytical factors and standardize sampling protocol should be considered. There are many factors that affect laboratory results such as a blood sample taken by an untrained person, skin cleaning with organic solvent (*e.g.* ether, isopropanol, ethanol) and patients have been treated more than 9 hours[4].

Alcohol analyses are available from body fluids and tissue in postmortem. The most appropriate blood sample of alcohol analysis is obtained from the femoral vein, external iliac vein or cubital vein.

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Otherwise, vitreous humor and urine could also be taken. Vitreous humor might be useful to confirm the BAC in the case of putrefaction. Vitreous humor is useful to corroborate postmortem BAC and distinguish from ante mortem, because it was isolated from bacteria. However, there was evidence that BAC can be produced up to 150 mg/dl in postmortem and putrefaction[4].

In equilibrium phase, BAC:VHAC ratio is less than 1 because water composition in blood is less than vitreous[4]. Many studies have suggested that BAC: VHAC ratio was more than 0.95 in early absorption phase. In contrast, the ratio is equal or less than 0.8 in the late absorptive and elimination phase[4].

Urine alcohol concentration (UAC) is also a good indicator of ethanol ingestion because urine normally contains little or no substrate for conversion by bacteria, unless in a patient who has diabetes mellitus. UAC is not reflected to BAC, which existed at the time of death, because urination and urine accumulation may have occurred before death.

Epidural hematoma is caused by tearing of the branch of the middle meningeal artery and associated with fracture of the skull. The common site is parieto-temporal area. Neurological defects occur between 1.5 hours - 7 days, but mostly were apparent after 4 hours[5-7].

Acute sub-dural hematoma arises by tearing of communicating vein or arterial origins which is not common. Neurological defects may develop a few hours after injury. So, SDHAC and EDHAC probably have the same concentration as BAC at the time of injury.

Material and Method

Selection of cases

All 25 corpses, selected from autopsy cases at the department of Forensic Medicine, Maharaj Nakorn Chiang Mai Hospital, Chiang Mai University in 2006, were performed within 24 hours after death. The inclusion criteria were the corpses which revealed our epidural hematoma or acute sub-dural hematoma, no head operation treatment and no putrefaction.

Sample

The specimens of femoral vessels, vitreous humor, epidural or sub-dural hematoma were collected about 3 ml in 5 ml glass tube which contained sodium fluoride and potassium oxalate as preservative and anticoagulant by a doctor and autopsy assistant. They were collected at -20°C in a refrigerator until processed with GC-HS.

Analytical procedures

The internal standard preparations used methyl ethyl ketone (MEK) 1 ml in a 1,000 ml volumetric flask and dilute to be 1000 ml with distilled water. The solution was divided to 10 ml vials and 0°C in a refrigerator.

200 μL of specimen was mixed with 12% tritron®X-100 solution 100 μL, distilled water 200 μL, 200 mg of sodium chloride and internal standard solution 100 μL in 10 mL vial. The vial was sealed immediately with a Teflon cap and Aluminium crimper.

Restek® ethanol standard solution or college of American Pathologist ethyl alcohol was used as the standard controlled solution with clinical standard solution® instead of blood sample and blood as negative control.

The gas contents were determined on a Hewlett Packard, HP 6890 gas chromatography equipped with a flame ionization detector. Separation was achieved on a capillary column (HP-INNOWax 30 m x 0.5 μm). The column temperature was raised after injection from oven temperature 65°C (5.5 min) raise 15°C/min to 130°C (1 min). The temperature of injection port and of the detector was 180°C and 200°C, respectively. The flow rate of helium which carried gas was adjusted to 1 ml/min. Head space auto sampler (Hewlett Packard, HP 7694) with 10 ml sample vial oven temperature 60°C, loop temperature 90°C, transfered line temperature 100°C, GC cycle time 14 min, vial equilibration time 5 min, mix power (shaking) high, pressurization time 0.15 min, loop fill time 0.15 min, Loop equilibrate time 0.15 min, injection time 0.50 min. The process was run with the HP Chemstation Plus family Rev.A.06.03 program internal stand type. The quality control sample and blank sample were tested at the end of the process.

Results

A total of 25 cases, 20 male and 5 female cases were observed. Age range was 17-58 years in males and 18-45 years in females. The most common manner of death was a traffic accident (16 cases) and 3 physical assaults, 2 gunshot injuries and 4 unknown. There were 7 corpses of epidural hematoma, 24 corpses of sub-dural hematoma and 6 corpses of both hematoma but 2 of them had an alcohol level from clots.

There were 18 corpses with SDHAC. Nine (9) corpses died at the scene of injury and SDHAC were detected in all. Eight (8) corpses showed BAC and VHAC. There was only one case of EDHAC.

SDHAC was less than BAC in the cases that had a history of survival time = 0 (died at scene), but it

varied when compared with VHAC. And ratio range of BAC: VHAC was 0.963-1.206 (Table 1).

The relation value of SDHAC to BAC compared with VHAC to BAC(R value) was 0.996 and 0.986 respectively.

There was the same result of survival rate at less than 9 hours (Fig. 1, 2).

Conclusion

SDHAC was lower than BAC in the corpses which died at the scene or survival time = 0 hour. Anyway, the relations between SDHAC and BAC have been better than VHAC and BAC. There was a small amount of EDHAC. So, it was difficult to get any conclusion.

Discussion

There are significant differences of alcohol concentration in blood samples which were obtained at the same time, from the same corpse because of differences in red cells and plasma composition. Water composition in serum and plasma contain approximately 10-15% more than whole blood. Alcohol concentration in serum:blood ratio is 1.15[4].

SDHAC have been studied and compared with BAC[4]. It was enclosed at different levels[4]. It probably concerned delayed bleeding that may evolve over a period of time as the result of continuous or intermittent bleeding, because the specimens were collected at 1.5 to 26 hours of survival time which affected the concentration level[4].

In the present study, the authors got the corpses which died at the scene of injury or the survival time was recorded as 0 hour but in fact, there was a short time to produce the small amount of hematoma. The alcohol concentrations from these clots probably are more reliability than in the previous study.

Consideration to the alcohol concentration of all cases that died at the scene of injury, BAC: VHAC ratios were ranged from 0.963-1.206. The previous study suggested that a blood: vitreous ratio greater than 0.95 which indicated that death occurred before equilibrium or early 30 minutes of absorption phase. In

Table 1. The study results from 25 corpses

Age/sex	Manner	Treatment	Survival time	EDHAC	SDHAC	BAC	VHAC
54 /M	traffic injury	yes	unknown	0	0	0	0
25/F	traffic injury	yes	43	-	0	0	0
19/M	unknown	yes	>48	-	23	0	-
45/M	unknown	yes	10	-	252	69	135
25/M	traffic injury	yes	0	99	112	132	137
58/M	unknown	unknown	unknown	-	16	-	24
U /M	physical assault	no	0	-	100	108	-
23/M	traffic injury	yes	15	0	0	0	0
19/M	gunshot injury	yes	18	-	28	0	-
25/M	gunshot injury	unknown	unknown	0	0	0	0
27/M	traffic injury	yes	7	17	42	17	43
35/F	traffic injury	yes	1	-	0	0	-
18/M	traffic injury	yes	2	-	205	240	264
20/M	traffic injury	no	5	78	-	81	-
20/M	traffic injury	no	0	-	261	276	269
19/M	traffic injury	no	0	-	122	144	136
18/F	traffic injury	no	0	-	245	259	245
U /M	traffic injury	no	0	-	247	265	232
17/M	traffic injury	no	0	-	159	-	156
45/F	unknown	yes	43	-	95	5	0
50/M	traffic injury	yes	26	-	49	0	-
48/M	traffic injury	yes	28	0	0	0	0
53/M	physical assault	no	0	-	220	256	213
30/F	physical assault	no	0	-	27	35	29
26/M	traffic injury	yes	20	-	79	0	3

* EDHAC: Epidural hematoma alcohol concentration, SDHAC: Subdural Hematoma alcohol concentration, BAC: Femoral blood alcohol concentration, VHAC: Vitreous Humour alcohol concentration (mg/dl) M = male, F = female, U: unknown age

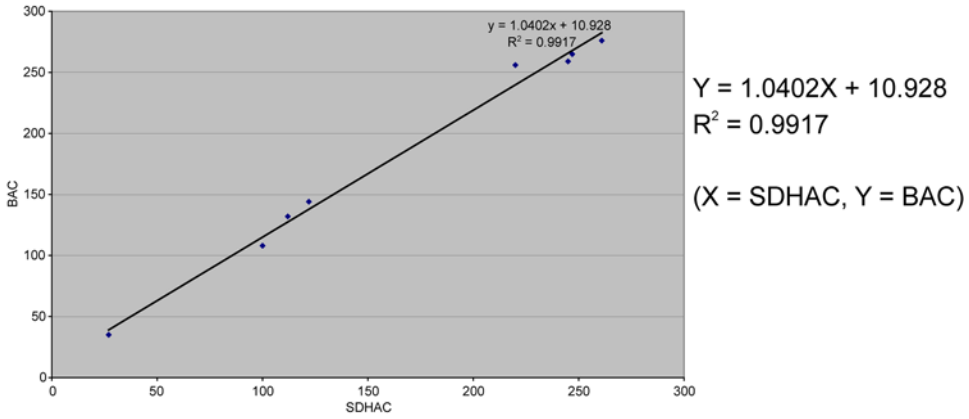

Fig. 1 Show the relationship between SDHAC and BAC of cases who died at scene of injury (Survival time after injury = 0 hour) (R = 0.996)

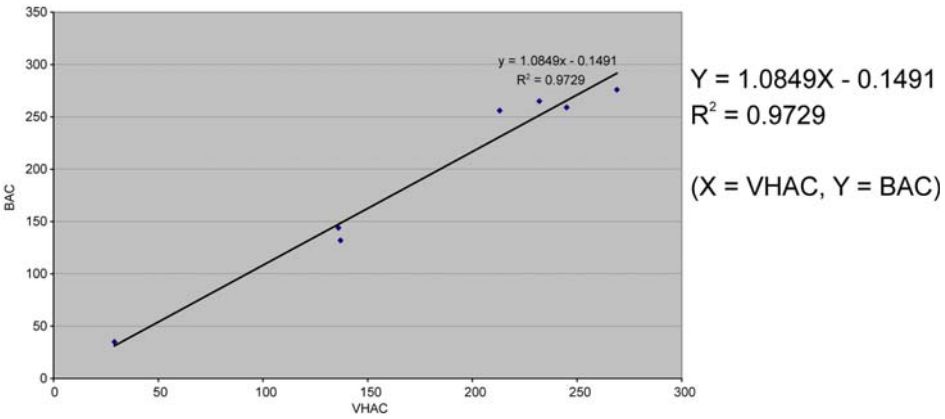

Fig. 2 Show the relationship between VHAC and BAC of cases who died at scene (Survival time after injury = 0 hour) (R = 0.986)

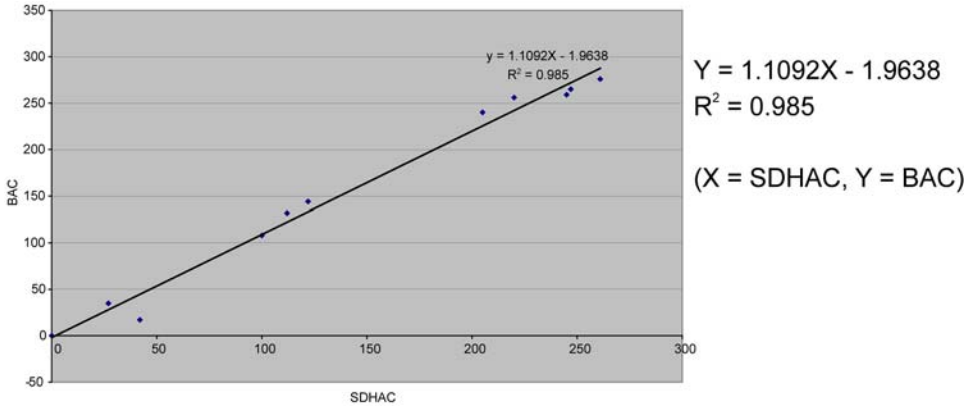

Fig. 3 Show the relationship between SDHAC and BAC of cases who died at 9 hours after injury = (R = 0.992)

late absorption and elimination phase, BAC must be lower than VHAC[4,8].

Alcohol is absorbed from gastrointestinal tract and increases its concentration in the absorption phase until the circulation system stopped or died. SDHAC, however, occurred immediately after head injury. So, it may be the most reliable concentration at time of injury. In the present study, the authors suggested that SDHAC, VHAC and BAC should be performed in every case where death at the scene of injury, with no treatment and no putrefaction. EDHAC would be very interesting to study in the future.

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ความสัมพันธ์ระหว่างระดับแอลกอฮอล์ในก้อนเลือดบริเวณเหนือหรือใต้ต่อเยื่อหุ้มสมองชั้นดูราและในเลือด เปรียบเทียบกับระดับแอลกอฮอล์ในน้ำลูกตาและในเลือด

สุภชัย บุญยัง, ไพฑูรย์ ณรงค์ชัย, อนงพันธ์ จันทร์กุย

ความสัมพันธ์ระหว่างระดับแอลกอฮอล์ในก้อนเลือดบริเวณเหนือหรือใต้ต่อเยื่อหุ้มสมองชั้นดูราและเลือด เปรียบเทียบกับระดับแอลกอฮอล์ในน้ำลูกตาและในเลือดของผู้เสียชีวิตโดยผิดธรรมชาติ ทำการศึกษาในปี พ.ศ. 2549 จำนวน 880 ศพ พบศพที่มีก้อนเลือดบริเวณเหนือต่อหรือใต้ต่อเยื่อหุ้มสมองชั้นดูรา และเสียชีวิตทันทีหรือไม่ได้รับการรักษา จำนวน 25 ศพ โดยทำการชันสูตรพลิกศพ ณ โรงพยาบาลมหาราชนครเชียงใหม่ ภายในเวลา 24 ชั่วโมง หลังการเสียชีวิต และตรวจหาระดับแอลกอฮอล์โดยเครื่อง Gas Chromatography Head space (GC-HS) จากผลการศึกษาพบว่าระดับแอลกอฮอล์ในก้อนเลือดใต้ต่อเยื่อหุ้มสมองชั้นดูราที่เกิดขึ้นอย่างเฉียบพลันมีความสัมพันธ์กับระดับแอลกอฮอล์ในเลือดที่เก็บจากเส้นเลือด femoral และมีความสัมพันธ์ดีกว่าระดับแอลกอฮอล์ในน้ำลูกตากับในเลือด และน่าจะมีค่าใกล้เคียงมากกว่าระดับแอลกอฮอล์ในเลือดจากเส้นเลือด femoral โดยเฉพาะขณะที่เกิดเหตุก่อนแอลกอฮอล์จะเข้าสู่ภาวะสมดุล (absorption phase prior to equilibrium) จากการศึกษาพบว่าแอลกอฮอล์ในก้อนเลือดใต้ต่อเยื่อหุ้มสมองชั้นดูรา สามารถใช้เป็นค่าอ้างอิงได้เช่นเดียวกับแอลกอฮอล์ในเลือดจากเส้นเลือด femoral และน้ำในลูกตา ในผู้ที่มีประวัติเสียชีวิตในที่เกิดเหตุและไม่อยู่ในสภาพเน่าหรือผู้ตายไม่ได้รับการรักษา