Title
Time-related change evaluation of the cerebrospinal fluid using postmortem CT

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Title: Time-related change evaluation of the cerebrospinal fluid using postmortem CT

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Key words: postmortem CT, cerebrospinal fluid, postmortem interval, postmortem
change, forensic radiology
Highlights

- We retrospectively evaluated the cerebrospinal fluid (CSF) CT density to compare the postmortem intervals.
- Postmortem 3rd day was the earliest increased CSF density compared to the day 1 and day 1.5, statistically.
- Postmortem 7th day showed increased CSF density compared to the early postmortem days (up to the 3rd day).
- There was no significant correlation between CSF density and estimated postmortem interval.
**Time-related change evaluation of the cerebrospinal fluid using postmortem CT**

Abstract

Purpose:
We retrospectively evaluated the cerebro-spinal fluid (CSF) CT density at the lateral ventricle to compare the postmortem intervals in cadavers.

Materials and Methods:
The number of cadavers enrolled in this study was 189 (male 120, female 69). According to the estimated postmortem time, the cadavers were divided into 13 groups (postmortem day 0.5, 1, 1.5, 2, 2.5, 3, 4, 5, 7, 10, 14, 21, 30), and were also re-grouped into 3 groups according to the postmortem time-width: group A (postmortem day 0.5 – 2.5), group B (day 3 – 7), and group C (day 10 – 30). Comparisons between the CSF density and estimated postmortem time were also analyzed.

Results:
The CSF density was around 20 HU up to day 2.5, and it increased gradually after day 3. Day 3 and 4 presented higher CSF density than day 1 and 1.5 (p < 0.05). Day 7 presented higher CSF density than day 3 (p < 0.05).

According to the postmortem time-width, the CSF density increased with postmortem time (p < 0.05).

The simple linear regression equations presented negative correlation between CSF density and estimated postmortem time, and $R^2$ was 0.119.

Conclusion:
The CSF density increased, but not linearly, according to the postmortem time, and the 3rd postmortem day was the earliest time allowing the difference to be detected. The CSF density needs further evaluation to enable estimation of the postmortem time.
CSF CT value along the postmortem period
The short postmortem period showed around 20 HU in CSF density, and it is gradually increased along with postmortem intervals. In statistical evaluation, postmortem day 3 was the earliest increased CSF density compared to day 1 and 1.5.
CSF CT value compared with the postmortem time-width

The CSF density shows an increase along with postmortem time-width, which is statistically significant.

Postmortem period:
Group A: up to day 2.5
Group B: day 3 to 7
Group C: beyond day 7
Background
One of the recent topics attracting interest in the field of legal medicine is postmortem CT imaging [1-3], and one of the reasons for this attraction is its objectivity [4, 5]. Postmortem CT images have the advantage of being able to provide objective information when required in cases such as personal identification [6, 7], and injury [8-12]. In addition, postmortem CT images have been used to investigate the cause of death [13, 14]. Thali et al. have reported on postmortem CT images combined with 3D surface recording, so called Virtopsy, and the postmortem image has been established as one of the important documents in the regional medical / social system [1, 15].

With increasing use and experience of postmortem CT examination, unknown findings have been detected on postmortem CT images [16, 17], so that it is important to evaluate specific postmortem change as it relates to pathological condition [18]. In addition, some postmortem CT image changes presented similarly to specific pathological lesions, so that it is important to differentiate disease-oriented pathological change from postmortem change [18, 19]. To address these issues and facilitate the use of postmortem CT in investigating the cause of death, it will be necessary to understand both the normal postmortem changes, and how to differentiate them from changes related to other causes.

Repeated studies on CT imaging have been reported which demonstrate the time-related postmortem change in brain, especially the lateral ventricle density in early postmortem period [20, 21]. But these reports covered only the short postmortem period, and longer postmortem interval evaluation is necessary in order to understand the time-related change in practical forensic radiology investigation.

In this study, we retrospectively evaluated the cerebrospinal fluid (CSF) CT density at the lateral ventricle to compare the postmortem intervals in cadavers.
Materials and Methods
This study was approved by the ethics committees of our institutions. Postmortem CT images obtained prior to full autopsy from January 2012 to November 2015 were utilized in the study. The following exclusion criteria were used: pediatric (younger than 20), small ventricle size (less than region-of-interest (ROI) size), cause of death was hypothermia or intra-cranial bleeding, containing intra-cranial gas, part of brain missing, and ambiguous postmortem interval time.

Imaging protocol
All cadavers were examined by postmortem CT prior to the full autopsy. The whole body scan was evaluated using a 64-slice multidetector CT scanner (Aquilion CX, Toshiba, Japan). The parameters from neck to head part were as follows: 120 kV, 300 mA, 1.0 s/rotation, pitch factor 0.641, configuration 0.5×32, reconstruction 0.5 mm, MPR (multiplanar reconstruction) image reconstruction 5 mm in axial, sagittal, and coronal sections.

Imaging analysis
All data sets were transferred to an image server in Digital Imaging and Communication in Medicine (so called DICOM) format and the 3 ROIs, at the lateral ventricle excluding the choroid plexus or artifact, were positioned to measure the CSF density using OsiriX (App v.3.9.4). The CSF density was calculated from the average of the 3 measured ROIs and the standard deviation (SD) was also calculated (Fig 1).

Estimated postmortem time
Postmortem time was evaluated according to the results of the full autopsy and the information on the cadaver’s circumstances. When the postmortem time was estimated with time width, the maximum period was used to estimate postmortem time.

Statistical analysis
To evaluate the time-related CSF density change course, the full number of cadavers was divided into groups according to the postmortem time: such as day 0.5, 1, 1.5, 2, 2.5, 3, 4, 5, 7, and >7. In addition, the cadavers were divided into 3 groups using the concept of postmortem time-width: up to day 2.5 (group A), day 3 to 7 (group B), and beyond day 7 (group C). We used StatView (SAS Institute Inc., North
Carolina, USA, Version 5.0) software with a Bonferroni/Dunn study. Differences with p < 0.05 were considered statistically significant. The collected data were also analyzed with a Spearman’s rank correlation test for the comparison between the CSF density and estimated postmortem time.
Results

The cadavers evaluated by postmortem CT prior to full autopsy numbered 391. According to the exclusion criteria, 202 cadavers were excluded: less than 20 year-old (n = 25), ambiguousness of lateral ventricles (n = 5), hypothermia (n = 10), intra-cranial bleeding (n = 26), intra-cranial gas (n = 109), lost information of head part (n = 6), and undefined postmortem-time (n = 21). Eventually, 189 cadavers (male 120, female 69) were enrolled in this study.

The causes of death were as follows: drowning (n = 31), sudden cardiac death (n = 23), asphyxia (n = 18), carbon monoxide poisoning (n = 10), multi-organ failure (n = 7), blood loss (n = 5), sepsis (n = 4), burn (n = 4), drug poisoning (n = 4), pulmonary artery embolization (n = 2), crash syndrome (n = 2), electrocution (n = 2), peritonitis (n = 1), ketoacidosis (n = 1), cardiac tamponade (n = 1), cancer (n = 1), hanging (n = 1), brain infarction (n = 1), malnutrition (n = 1), renal failure (n = 1), and unknown (n = 69).

Postmortem times were as follows: day 0.5 (n = 12), day 1 (n = 17), day 1.5 (n = 24), day 2 (n = 80), day 2.5 (n = 4), day 3 (n = 22), day 4 (n = 7), day 5 (n = 2), day 7 (n = 9), day 10 (n = 4), day 14 (n = 4), day 21 (n = 1), and day 30 (n = 3). The 3 groups of the postmortem time-width were group A day 0.5 – 2.5 (mean 1.7 days), group B day 3 – 7 (4.2 days), and group C day 10 – 30 (17.3 days). On examining basic case profiles, no significant differences were found among the postmortem times, nor among the groups A, B, and C.

CSF CT value

The measurements of CSF density are shown in Table 1. The CSF density was around 20 HU up to day 2.5, and it increased gradually after day 3. Day 5, 10, 14, 21, and 30 were excluded from statistical evaluation because of insufficient numbers. Using Bonferroni-Dunn test, day 3 and day 4 presented higher CSF density than day 1, and day1.5 (p < 0.05), and day 7 presented higher CSF density than day 0.5, 1, 1.5, 2, 2.5, and day 3 (p < 0.05) (Fig 2) (Table 2).

According to the postmortem time-width, the mean CT values of CSF density were 20.0 ± 3.3 in group A, 22.0 ± 4.2 in group B, and 26.5 ± 5.9 in group C (Fig 3, Table 3). There were statistical differences among the three groups, thus the CSF density was increasing with the postmortem intervals.
The simple linear regression equations presented negative correlation between CSF density and estimated postmortem time, and $R^2$ was 0.119. The linear equation formula for postmortem time was $Y = 19.804 + 0.324 \times X$. 
Discussion

Postmortem CT findings have been reported showing changes with the postmortem intervals in vascular diameters [22, 23], lung appearance [24, 25], and pleural effusion [18, 26]. In this study, the CSF density at the lateral ventricles showed an increase with postmortem time, and this was concordant with previous reports [20, 21]. In addition, according to postmortem time-width, the CSF density exhibited a statistically significant increase along with postmortem time. This is the first report of a statistical increase in CSF.

The mechanisms of increasing CSF density are still under evaluation. Hasegawa et al. [21] reported the possibility of decomposition of CSF protein and/or the ependymal layer, CSF condensation while a swelling brain exerts pressure on the ventricles leading to shrinkage, and inflow of protein-like substances from the periventricular brain tissues to the CSF. The increasing X-ray absorption leads to the increasing CT value, so that the CSF must be increasing its density along with postmortem time. Further study will be needed to evaluate the pathological change of the ependymal layer and/or periventricular brain tissue affect increasing CT density with the postmortem time.

Koopmanschap et al. [20] reported that the correlation between density and postmortem interval showed a significant correlation and the maximum postmortem interval can be estimated by a linear correlation in early postmortem period. Hasegawa et al. [21] also reported a linear equation formula relating postmortem time and CSF CT density. They used cadavers with confirmed time of death and examined within the early postmortem time. From our results, there was no significant linear correlation between CSF density and estimated postmortem time. It was thought that the postmortem times covered too wide a time range, and in addition, environmental factors (temperature, humidity, and wind) might affect the postmortem change. Furthermore, the standard deviation of CSF density was large and there was overlap between postmortem times, which made it difficult to estimate the postmortem time using CSF CT density.

In this study, the difference in CSF density over the postmortem interval was within 10 Hounsfield Units (HU), and it would seem difficult to differentiate it without ROI measurement. To put it another way, the CSF density showed no big difference over postmortem time. If we selected a suitable threshold, it could be
used to indicate a physiological condition (such as intra-ventricle bleeding) based on the CSF density. In this investigation, the CSF density reached a maximum of 34 HU, so that a value of more than 35 HU may present the possibility of a pathological condition. On the other hand, blood leakage (transudation) to the sub-arachnoid space has been reported in postmortem change [27]. If this phenomenon may affect the lateral ventricle, the CSF density evaluation would be useless. It will be necessary to evaluate if and how blood leakage (transudation) occurs during the postmortem time into the lateral ventricle.

We used exclusion criteria such as pediatrics, small ventricle size, hypothermic death, intra-cranial bleeding, intra-cranial gas, incomplete brain, and unknown postmortem interval time. A small ventricle would make it difficult to measure CSF density using ROI. Hypothermia has been reported to affect the postmortem change [18, 24, 28, 29]. Intra-cranial bleeding and gas would affect the CSF density directly, and make it difficult to evaluate the normal postmortem CSF density change. Either brain incompleteness or unknown postmortem interval time would make it impossible to evaluate the CSF density change according to postmortem time. In the normal postmortem change, putrefaction might also affect the cadaver and produce putrefaction gas [30, 31]. In this study, CSF density could be evaluated for long postmortem time in 12 cases (beyond day 10). A cadaver which was not a hypothermic death might be placed low temperature conditions so that it would retain its postmortem condition without deterioration. Therefore, to observe the true CSF density change according to the postmortem interval, it would be necessary to control the cadaver’s environmental conditions, especially the temperature.

Limitations
Several limitations may be listed concerning this study. First, we used the same CT equipment with the same protocol throughout, but if different CT equipment and / or a different protocol was used, the CSF density might result in a different CT number. Second, the 3 ROI measurements were taken by one operator to minimize the operator dependency. But if automatic ROI setting was used for the measurements, more objective data could be obtained for evaluating the CSF density change with the postmortem time. Third, we used the postmortem time from the evaluation of the autopsy findings, which might contain some error range.
Conclusion
The CSF density was increasing, but not linearly, according to the postmortem time. Compared with the early postmortem time, the 3rd postmortem day was the earliest time to show any increase, and the 7th postmortem day was the first to show a statistically significant difference. Because much overlap of CSF density was evident, it was difficult to estimate the postmortem time based on CSF density.
References


Acknowledgments

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<td>mean ±SD (HU)</td>
<td>20.0 ± 3.3</td>
<td>22.0 ± 4.2</td>
<td>26.5 ± 5.9</td>
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Fig. 1 Measurement of CSF CT density
The CSF densities at the lateral ventricles were measured using 3 ROIs, which excluded the choroid plexus or visible beam hardening artifact. The pixel standard deviation was also measured.
(a) 40 year-old female, death by pulmonary artery thromboembolism. 2 days postmortem (group A).
(b) 30 year-old male, death by cardiac infarction. 5 days postmortem (group B).
(c) 70 year-old female, death by starvation. 10 days postmortem (group C)

Fig. 2 CSF CT value along the postmortem time
The short postmortem period showed around 20 HU in CSF density, and it is gradually increased along with postmortem intervals. In statistical evaluation, postmortem day 3 was the earliest increased CSF density compared to day 1 and 1.5. Day 7 presented statistically increased CSF density compared to the short postmortem period (up to 3 days).

Fig. 3 CSF CT value compared with the postmortem time-width
The CSF density shows an increase along with postmortem time-width, which is statistically significant.
Postmortem period:
Group A: up to day 2.5
Group B: day 3 to 7
Group C: beyond day 7
Figure 2

Bonferroni-Dunn
* p < 0.05
Figure 3

CT value (HU)

Bonferroni-Dunn
* p < 0.05

Group A
Group B
Group C