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Original Research Article

Evaluation of Cells in Cerebro Spinal Fluid for Postmortem Interval

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Abstract

Background: The post-mortem interval, often known as the time since death, is the period of time between when a person passes away and when the body is examined by a forensic expert.

Objectives: To estimate the total count and differential count of cells in the cerebrospinal fluid with cytological differences.

Methods: This prospective study was conducted in the Department of Forensic Medicine in a medical college for a period of 6 months. This study analyzed the CSF from the cerebellopontine cistern for cell count and morphology in all medicolegal autopsies with known time since death by Neubaur chamber and light microscopy. The study sample consisted of 40 CSF samples taken from medicolegal autopsies. Ethical clearance was obtained from the Institutional Ethical committee (IEC) before starting the study. All cases subjected for medico-legal autopsy whose time of death is known were included in the study.

Results: Most common cause is natural causes comprising 36%, followed by poisoning in 28%. Hanging and trauma were other common causes. Out of 40 patients (27 were males and 13 were females) so the study was female preponderance. The most common age group was 21-40 years in 48.71% followed by 41-60 years in 28%. The mean value of total cell count in CSF in PMI of 0-6 hours is 250 with a standard error of 28.86. The mean value of total cell count in PMI 6 – 12 hours is 388.81 with a standard error of 37.36. The mean differential cell count in cases with PMI 0 – 6 hours shows that mean neutrophils count is 23.33, mean lymphocyte count is 70 and the mean eosinophil count id 6.66. There were no degenerated cells in this period The mean differential cell count in cases with PMI 6 – 12 hours shows that the mean neutrophil count is 12.72; the mean lymphocyte count is 75.45. The mean degenerated cells in this period are 11.81.

Conclusion: According to the study, the post-mortem increase in CSF cell count is a typical physiological occurrence. Up to 12 hours after the injury, the CSF cell count increases, but the first 6 hours show no degenerative alterations hours. Neutrophils, lymphocytes, plasma cells, eosinophils, and histiocytes are the distinguishable cells.

Keywords: Cerebro-Spinal Fluid (CSF), Post Mortem Interval, Pleocytosis, Differential Count, Autopsy.

Introduction

The post-mortem interval, often known as the time since death, is the period of time between when a person passes away and when the body is examined by a forensic expert. [1] Accurate calculation of the post-mortem window has significant repercussions in both criminal and civil trials. Fixing the incident's time (homicide, suicide, accident), tracking the travels of the accused and victim, and confirming the veracity of eyewitness testimony are all benefits. [2]

Thanatochemistry is the study of the chemical composition of the dead. Following a person's death, it details changes in the biochemical content of their various bodily fluids. It contains the numerical estimate of ions, metabolites, and electrolytes in bodily fluids, which are typically housed in closed containers. Blood may be easily biochemically analyzed thanks to the simple venepuncture technique. But because blood circulates, a variety of things could have an impact on its biological makeup. Moreover, it decomposes more quickly than other compartmental fluids. [3,4]

These compartmental fluids have the drawback that grading based on physical post-mortem alterations is proven to be more accurate in the first 60 hours following death than scoring based on biochemical changes. [5] Thus, various methods similar post-mortem proteomic investigations and post-mortem cytological alterations in cerebrospinal fluid. In this study, the total count and differential count of cells in the cerebrospinal fluid is estimated.

Materials and Methods

This prospective study was conducted in the Department of Forensic Medicine in a medical college for a period of 6 months. This study analyzed the CSF from the cerebellopontine cistern for cell count and morphology in all medicolegal autopsies with known time since death by Neubaur chamber and light microscopy. The study sample consisted of 40 CSF samples taken from medicolegal autopsies. Ethical clearance was obtained from the Institutional Ethical committee (IEC) before starting the study. All cases subjected for medico-legal autopsy whose time of death is known were included in the study.

Cases having past history of head injury, previous history of neurosurgery, Head and neck tumors, cranial infections like meningitis, encephalitis, Blood stained CSF aspirate, Postmortem interval more than 36 hours were excluded.

Methodology:

The cases of head injury and previous history of neurosurgery indicates a breach in blood CSF barrier that may affect the cell count in the CSF in living patients which will give false positive elevation in the CSF pleocytosis. Infections like meningitis result in the increase in neutrophils and lymphocytes depending upon the causative organism of the meningitis which result in false positive value in the total and differential count. The resulting CSF is admixed with 47 when the cisternal puncture is performed deeper or in an incorrect place so that it pierces any of the spinal veins blood. As a result, the CSF cell count readings become inconsistent. Blood-stained aspirates ought to be thrown away

in order to obtain a true positive for CSF pleocytosis.

The CSF pleocytosis and morphological studies that were conducted to determine the postmortem interval reveal that the cells become indistinguishable after 24 hours. In order to investigate the cases with positive connection, cases with postmortem intervals more than 36 hours were disregarded.

Statistical Analysis:

The statistical analysis was performed using SPSS for windows version 22.0 software (Mac, and Linux). The findings were present in number and percentage analyzed by frequency, percent, and Chi‑squared test. Chi‑squared test was used to find the association among variables. The critical value of *P* indicating the probability of significant difference was taken as <0.05 for comparison.

Results

Table 1: Causes of cases in the study.

Cause of Death	Number of cases	Percentage
Natural cause	14	35.89%
Hanging	7	17.94 %
Snake bite	2	5.12 %
Poisoning	11	28.20 %
Electrocution	2	2.56%
Trauma	4	10.25%
Total	N=40	

As per table 1 total number of cases in post-mortem interval is 40 in the present study. Most common cause is natural causes comprising 36%, followed by poisoning in 28%. Hanging and trauma were other common causes. Out of 40 patients (27 were males and 13 were females) so the study was female preponderance. The most common age group was 21-40 years in 48.71% followed by 41-60 years in 28%.

Table 2: Postmortem Interval (PMI) in the study cases

PMI (hours)	No. of cases	Mean TC	Std. Deviation	Std error
0-6	3	250	50	28.86
6-12	11	381.81	125.045	37.76
12-18	16	390	142.98	45.24
18-24	3	316.67	57.73	33.33
>24	7	262.5	75	37.5

As per table 2 the mean value of total cell count in CSF in PMI of 0-6 hours is 250 with a standard error of 28.86. The mean value of total cell count in PMI 6 – 12 hours is 388.81 with a standard error of 37.36. The mean value of total cell count in CSF in PMI of 12 to 18 hours is 390 with a standard error 45.24. The mean total count in CSF in PMI 18 – 24 hours is 316.67 with a standard error of 33.33. The mean total count in CSF in PMI >24 hours is 262.5 with a standard error 37.5.

Table 3: Differential Cell count with PMI

PMI (hour)	Neutrophils	Lymphocytes	Plasma cell	Histiocytes	Eosinophils	Degenerated cells
0-6	23	70	-	-	6.6	-
6-12	12.7	75.4	-	-	-	11.81
12-18	17	30	1.67	2	4.67	44
18-24	20	50	6.67	6.67	3.33	10
>24	7.14	30	5.71	2.85	2.85	57.14

As per table 3 the mean differential cell count in cases with PMI 0 – 6 hours shows that mean neutrophils count is 23.33, mean lymphocyte count is 70 and the mean eosinophil count id 6.66. There were no degenerated cells in this period The mean differential cell count in cases with PMI 6 – 12 hours shows that the mean neutrophil count is 12.72; the mean lymphocyte count is 75.45. The mean degenerated cells in this period are 11.81. The mean differential cell count in cases with PMI 12 – 18 hours shows that the mean neutrophil count is 17, the mean lymphocyte count is30. The mean plasma cell count is 1.67, mean histiocyte is 2 and the mean eosinophil is 4.67. The mean degenerated cell in this period is 44. The mean differential cell count in cases with PMI 18 – 24 hours shows that the mean neutrophil count is 20; the mean lymphocyte count is 50. The mean plasma cell count is 6.67, mean histiocyte is 6.67 and the mean eosinophil 3.33. The mean degenerated cell in this period is 10. The mean differential cell count in cases with PMI >24 hours shows that the mean neutrophil count is 7.14, the mean lymphocyte count is 30. The mean plasma cell count is 5.71, mean histiocyte is 2.85 and the mean eosinophil is 2.85. The mean degenerated cell in this period is 57.14.

Table 4: Degenerative changes with PMI

PMI (hours)	Cytoplasmic vacuolation	Nuclear vacuolation	Nuclear fragmentation	Degene ration
0-6				
6-12	9%		18%	17%
12 - 18	13.33%	6.67%	40%	46.67%
18-24		33.33%	66.67%	66.67%
>24	14.28%	14.28%	28.57%	14.28%

As per table 4 The cells were studied microscopically to detect the degenerative changes. The cytological study of cells in CSF in cases with PMI 0 – 6 hours did not show any degenerative changes. The cytological study of cells in CSF with PMI 6 – 12 hours showed that 54 % of the cells did not show any degenerative changes while 9% of cells were completely degenerated beyond recognition. 9% of the cells showed cytoplasmic vacuolation, 18% of the cells showed nuclear fragmentation and 17% of the cells showed nuclear degeneration.

Discussion

The cerebrospinal fluid in a healthy person is a transparent liquid with no or very few cells (1–5/cu millimeter). Few research have been done to determine the relationship between the PMI and the increase in CSF cell count. [5,6,7] There are variations in white blood cell counts across studies. Neutrophils and lymphocytes are the most prevalent cell types. Eosinophils and histiocytes are two more cells that are visible. Only very rarely do plasma cells appear, which is consistent with earlier research. [8,9,10] The finding in this study supports those in other investigations and demonstrates that the CSF cell count increases after death.

The differential count indicates 30 – 40% of neutrophils and 60 – 70% of lymphocytes in the first 6 hours. In the next 6 hours the neutrophil count was 10-20% and lymphocytes were 80 -90%. Infrequently plasma cells, eosinophils and histiocytes were present. After 12 hours, in most cases the cell count becomes impossible due to degenerative changes and the differential count does not have significant correlation.

When the PMI is the same duration, it is discovered that the cause of death has no relationship to the rise in cell count. The false positive results were excluded by the exclusion criteria. The reason for this post-

mortem rise in CSF has not been investigated because the goal of this work is to determine a correlation between the cell count in CSF and the PMI. [11,12] It is still unknown whether cells actively enter the CSF immediately after death or whether post-mortem diffusion of cells results from a breach of the blood-to-cSF barrier. Due to breakdown of the CSF brain barrier, viral and inflammatory disorders of the meninges and brain result in an increase in CSF cell count in living patients. [13,14]

Although time since death is affected by various factors and results in various outcome in the physical features of the body, biochemical composition of the body fluids, it could be estimated in a range when more than one method is used together. [15] The CSF cell count could be used corroboratively to estimate the PMI range along with other methods of estimating PMI. [14]

Conclusion:

According to the study, the post-mortem increase in CSF cell count is a typical physiological occurrence. Up to 12 hours after the injury, the CSF cell count increases, but the first 6 hours show no degenerative alterations hours. Neutrophils, lymphocytes, plasma cells, eosinophils, and histiocytes are the distinguishable cells. At 12 hours, neutrophils are no longer recognisable, and in 20 hours, lymphocytes have degraded. The differential count is unreliable beyond 12 hours. The lymphocyte count is higher than the neutrophil count in the first six hours. The cell count rises during the following six hours, with lymphocytes continuing to outnumber neutrophils.

This research suggests that CSF pleocytosis, a rise in total count, relative counts of several types of white blood cells, including neutrophils, lymphocytes, and plasma cells, as well as morphological analyses of each cell for the degenerative alterations, in addition to other measures already in use, can aid in determining the post-mortem interval. Hence, in circumstances when the period since death is unknown, CSF cell count and morphological analysis could be used as a corroboration method with other studies.

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