H67 Tau and Neurofilament Light Proteins in Cerebrospinal Fluid as Biomarkers of the Time of Death

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Learning Overview: The goal of this presentation is to present an exploratory, cross-sectional study aimed at identifying two new biomarkers of the time of death by means of thanatochemistry.

Impact on the Forensic Science Community: This presentation will impact the forensic science community by raising awareness of how thanatochemistry may help in more precise and reliable estimation of the Postmortem Interval (PMI), thanks to the most recent technological developments in this field.

Background: One of the most important and challenging tasks in forensic medicine is the accurate determination of the PMI. Different methods have been proposed, but they all lack precision and reliability. Recent developments in biochemical techniques may provide the opportunity to assist in more precise estimation of the time of death. The study of postmortem chemical changes in closed compartment body fluids such as vitreous humur and Cerebrospinal Fluid (CSF) has shown promise. In the present study, two major proteins of the central nervous system (Tau and Neurofilament Light (NFL) proteins) were investigated in CSF as potential biomarkers of the time of death.

Objectives: The main objective of the study was to assess the correlation between the concentrations of Tau and NFL in CSF and the PMI. The secondary objectives were: (1) to determine the inter-individual variability of the concentrations of Tau and NFL for a same PMI; (2) to determine the kinetics of these concentrations over time; and (3) to determine the variability of these concentrations according to the sampling site (lumbar vs. sub-occipital).

Materials and Methods: This study was reviewed and approved by the Ethics Committee of the University Hospital of Montpellier, France. Postmortem CSF samples were collected from 100 adult cadavers whose time of death was precisely known, at the mortuary of the University Hospital of Montpellier, France. Individuals with neurological disorders and head trauma were excluded from the study. CSF samples were removed by cisternal and lumbar punctures at different time intervals. Two mL of CSF were obtained at each tap in clean, sterile polypropylene tubes, using a 18G lumbar puncture needle. The cadavers were divided into four groups according to the PMI (n=25 in each group). The samples were taken 0h–6h (group A), 6h–12h (group B), 12h–18h (group C), and 18h–24h (group D) after death. Additionally, CSF samples were collected every 3h from ten cadavers during the first 15h postmortem. All cadavers were kept at room temperature (+20°C±2°C) during sample collection. CSF samples were transferred in ice to the laboratory where they were centrifuged for 10min (+4°C, 1,000g). The clear supernatant was divided into aliquots, then stored at -80°C until analysis. The rectal and tympanic temperatures at the time of CSF collection were measured using a probe thermometer, and *rigor* and *livor mortis* were assessed.

Concentrations of total Tau and NFL in CSF were measured by conventional and ultrasensitive digital immunoassays, respectively. Total protein concentration was determined using a bicinchoninic acid protein assay.

The correlation coefficient between the concentrations of Tau and NFL in CSF and the PMI was calculated in each case. The inter-individual variability was assessed by measuring the Standard Deviation (SD) of the mean concentrations of Tau and NFL in each group. Linear regression analysis (adjusted for confounders) was used in assessing whether concentrations of Tau and NFL were dependent on the PMI. Paired Student's *t*-test was used to assess the variability of Tau and NFL concentrations depending on the site of CSF collection.

Results pending.

Conclusion: Thanks to this exploratory study, it will be possible to know if Tau and NFL proteins can be considered as potential CSF biomarkers of the time of death and if further research is needed to confirm these preliminary results.

Tau Protein, Neurofilament Light, Biomarkers