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Spectrographic Analyses of Human Spinal Fluid.*

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In connection with other studies on inorganic salt and metal distribution in cells and tissues we have had occasion to examine spectrographically a series of samples of spinal fluids taken from the usual hospital and clinic population. Our attention was directed toward a search for elements that are not usually recorded as being normal constituents of human spinal fluid. These are Pb, Al, Ba, Sr, B, Sn. Samples of spinal fluid were generously provided for us by Drs. A. F. Hartman and W. B. Wendell.

The method used routinely in our examinations was as follows: 2 cc. of spinal fluid were placed on a carefully cleaned glass plate and evaporated to dryness at 100°C. The residue was scraped together, placed on a pure carbon electrode, wetted slightly with a small drop of the original fluid and dried. The loaded carbon was placed in front of the slit of a Bausch and Lomb Medium Quartz Spectrograph and the salt ignited by means of an intermittent arc. About 100 flashes of the arc covering a total time interval of 80 seconds sufficed to produce good pictures of lines throughout the ultra-violet spectrum. The intermittent arc is formed by making and breaking electrical contact between 2 vertical electrodes. The upper one has a motor-driven piston-like motion while the lower one is fixed and capped with the sample.

In our search for Al we used 22 samples of spinal fluid. The line used to detect the presence of aluminum was the 3082.16 Å line and as a reference line we employed the conveniently placed 3096.92 Å line of Mg. Those Al lines of greater intensity than that of the Mg were called "strong" and those of much less intensity were designated as "weak". If the 2 lines, Al and Mg, were of approximately equal intensity the Al was referred to as "medium". It is admitted that this method of estimation is a more or less arbitrary one, but is frequently used where more accurate estimates are not necessary. A rough estimation of the actual amount of Al present in the samples yielding the strong lines is one part to 10^8 parts by weight, of fluid.

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The result of the 22 samples examined for Al are briefly as follows: 5 spinal fluids showed strong lines; 11 were medium, and 4 designated as weak. Thus 20 of the 22 gave a positive Al test and 2 were negative.

As a check on the method and the materials blank runs were frequently made on the electrodes. It so happens that when samples are arced in the fashion outlined above very little of the electrode is consumed and when blank runs of the electrodes are made a great deal of the carbon is used. In spite of this balance in favor of the blank electrodes, we have consistently failed to find Al in them. While this at first would seem to meet the necessary control requirements further consideration shows that a more rigorous control measure would be one in which a "synthetic" spinal fluid was arced. For this purpose we concocted a chemical mixture which approaches as far as possible the same salt content as spinal fluid. The "synthetic" spinal fluid, although made with reagent quality chemicals, when examined spectrographically reveals some Al. The amount is sufficiently great to be comparable with the spinal fluids showing Al lines designated as faint. But we are confident that the Al is in the chemicals used in the mixture rather than in the electrodes. Dee Tourtellotte and Rask,[1] for example, report that a spectrographic analysis of the C. P. reagents in their laboratory showed that 50% of them contained definite traces of Al. That we have not been able to obtain a carbon bearing the "synthetic" spinal fluid free of Al need not throw doubt upon the existence of this metal in human spinal fluid. Both the strong and medium lines are well outside the possible contamination category. Nor do we feel that the instruments used in obtaining the samples contribute anything to the fluid. This has been checked by a number of means.

A series of 18 specimens of spinal fluid was examined for Pb. seven of these were from the Children's Hospital and plumbism was a possible diagnosis. None of this group was finally diagnosed as a case of Pb poisoning; however, one showed evidence of lead deposition in the bones. Of the remaining 11 samples 3 were diagnosed clinically as plumbism.

As in the case of spinal fluids studied for Al the results were divided into 3 categories, strong, medium and weak. Four of the specimens showed strong Pb lines, 2 of these were from the 3 diagnosed clinically as Pb poisoning and 2 from the group of children which were suspected of having plumbism. In 6 of the samples Pb

[1] Tourtellotte, Dee, and Rask, O. S., *Indust. Eng. Chem.*, 1931, **3**, 97.

was found to be present in "medium" quantities and in 5 "faint." Three of the spinal fluids showed no Pb.

Observations for Pb were made on the 2833.07 Å line since it is one of the most sensitive. The amount of Pb was estimated by comparing the intensity of the 2833.07 Å line in a suspect with that of a synthetic solution to which had been added a known amount of $Pb(No_3)_2$. From this comparison it was estimated that the specimens designated as having a strong line contained about 10^{-3} parts of lead by weight.

An examination was made of 18 samples of spinal fluid for Sn. Of the group 5 showed definite evidence of this metal with one specimen catalogued as very strong, one medium and 3 weak. The remaining members of the series, 13 in number, showed no trace of Sn. The occurrence of the sensitive 2839.99 Å line was used as a criterion in the estimations.

In addition to the elements named above the ubiquitous Na, Ca, Mg, Cu, K, and P were always found in relative abundance. Ba, Sr, and B were in all specimens examined. We were hampered in our studies by the fact that the electrodes used contained Fe, Mo, Rb, and Si. These elements have been reported as being found in tissues and one might reasonably expect them in spinal fluid.

While our series of cases is too small to permit us to indulge in statistical treatment it may be said that, within the limits of the sensitivity of the method used, all samples of spinal fluid can be expected to show evidence of Al, Ba, Sr, B; about half of them Pb and a fourth of them Sn.

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Action of Immune Serum on Meningeal H. Influenzae in vitro and in Experimental Infections.

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Freshly isolated meningeal strains of *H. influenzae* differ from most respiratory strains in their "smooth" colony forms and their virulence for rabbits and mice, and these closely correlated proper-

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