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"BOUND" CHLORINE IN CASEIN AND IN TISSUE PROTEINS

BY

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Lindahl recently analysed casein, and found that it contained about one per cent chlorine in bound form. In the analytical method used by him the casein was treated with a boiling mixture of chromic and sulphuric acids, and the gases evolved were passed through a known quantity of silver nitrate containing As_2O_3. The consumption of silver nitrate, assumed to be due to the clorine in the casein, was determined by the Volhard method. As the presence of chlorine in casein does not agree with the generally accepted opinion on the composition of this protein it seemed necessary to check *Lindahl*'s results. At the suggestion of Professor *E. Jorpes* I undertook to control the analytical procedure. Dr. *Ragnar Berg* who performed the analyses for *Lindahl* kindly demonstrated his technique in our laboratory, thereby greatly facilitating our work.

Casein, prepared by isoelectric precipitation with acetic acid, was analysed for chlorine by the Carius method. No trace of chlorine could be detected. When the analysis was performed with the technique used by *Lindahl,* a precipitate insoluble in nitric acid was actually formed, and the Volhard titration showed a consumption of silver nitrate corresponding to 0.25 per cent chlorine in the casein.. It was, however, observed that the precipitate differed from silver chloride in that it did not darken when exposed to sunlight. When fused with

Na_2CO_3 no chloride was obtained in the alkaline filtrate whereas a similar quantity of silver chloride gave a quantitative precipitate with $AgNO_3$ on acidification with HNO_3. Evidently the precipitate formed in the trap with silver nitrate was not silver chloride. Of the different silver salts that might be formed the formiate, carbonate and acetate are readily soluble in dilute nitric acid. The oxalate darkens easily in sunlight. The silver cyanide, however, is practically insoluble in dilute and only slowly soluble in concentrated nitric acid; moreover, it does not darken when exposed to sunlight. Consequently, the precipitate was assumed to be silver cyanide. This assumption was confirmed by analyses of the sample. A qualitative test for the cyanide ion with ammonium polysulphide and ferric chloride was positive. On ignition of 31,265 mg the silver residue weighed 25,065 mg or 80.17 per cent. The calculated amount of silver in silver cyanide is 80.57 per cent. In another sample the ignition residue was dissolved in nitric acid precipitated with hydrochloric acid and the precipitate weighed. The silver content of the chloride corresponded to 80.2 per cent of the original sample.

Evidently this source of error in the chlorine determination escaped the attention of *Lindahl*. His conclusions about the protein-bound chlorine in the body are consequently erroneous.