showing maximal optical activity[2] generously placed at our disposal by Professor J. H. Andrews of the University of Pennsylvania. When analyzed by the Sullivan-Lugg method, the urinary cystine showed a purity of 99.7% as compared with a purity of 99.5% for the protein cystine, values within the range of accuracy of the Sullivan-Lugg procedure.[5]

0.5 gm. samples of each of these cystines were dissolved in 50 cc. of N hydrochloric acid and the rotations determined in 2 dcm. tubes at 30° using sodium light. With the urinary cystine an $[\alpha]_D^{30°}$ of —201.0° was obtained; with the protein cystine, a rotation under the same conditions of —202.5°. As a further check on our determinations, the optical activities of the 2 cystines were determined independently by Professor Andrews. Values of —214.0° and —215.0° at 25° respectively were obtained.

If our values are corrected to a temperature of 20°, using the factor of Toennies and Lavine,[3] the specific rotations are —221.6° and —223.1° respectively, values which compare favorably with the classical values of Fischer and Suzuki,[6] —223.6° for stone cystine and —221.9° for cystine from hair.

These data offer no evidence that cystine which crystallized spontaneously from cystinuric urine and which had had minimal opportunity for racemization, differed significantly in its specific rotation from the maximal values usually given for the rotation of *l*-cystine from protein hydrolysates.

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Charcoal Adsorption as a Method for the Preparation of a Concentrated Liver Extract.

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It has been noted that in concentrating liver extracts for the treatment of pernicious anemia to very small volumes a considerable degree of potency was lost in the precipitate which formed. In the search for a satisfactory method of obtaining a concentrated liver extract which could be administered intramuscularly and which

[5] Lugg, J. W. H., *Biochem. J.*, 1933, **27**, 668.

[6] Fischer, E., and Suzuki, V., *Z. physiol. Chem.*, 1905, **45**, 405.

was as potent as a dilute preparation derived from 100 gm. of liver, the following procedure was developed.

Fresh liver is ground and suspended in water in the proportion of 5 lb. of liver to 5 liters of water. This suspension is stirred for 15 minutes and then heated to 80°C. in a water bath. The solution is filtered off, and the residue is washed with hot water and filtered again. The combined filtrates are concentrated to about one-fourth of their volume and a 25% neutral lead acetate solution is added until precipitation no longer occurs. The precipitate is filtered off and enough baryta-water is added to the filtrate to give complete precipitation. The precipitate is filtered off and sulfuric acid is added to the filtrate to precipitate the lead and the barium. This suspension is centrifuged, and the clear yellow filtrate is poured off and brought to pH 5.0 with sodium hydroxide. Norite charcoal (about 25 gm. for every initial 5 lb. of liver) is then added to the solution and the mixture is allowed to stand for an hour with frequent shaking. The charcoal is filtered off, washed, suspended in 50% alcohol (400 cc. per 25 gm. charcoal) adjusted to pH 5.0, and heated to 65-70°C. on a steam bath. The alcoholic solution is removed and the precipitate is again treated with 50% alcohol. The combined alcoholic filtrates are concentrated under reduced pressure and reduced in volume until 3 cc. of the solution represent 100 gm. of fresh liver. This solution is then filtered, boiled in a water bath for a few minutes, cooled in the ice box over night, filtered again, bottled, and sterilized. Final pH = 5.

The clear yellow extract was given intramuscularly in a 3 cc. dose to a patient with pernicious anemia whose red blood cell count was 0.80 millions per cu. mm. before transfusion and 1.43 millions per cu. mm. after transfusion the day treatment began. The reticulocyte peak was 52.4% in 5 days, and in 7 days the red blood cell count was 1.94 millions per cu. mm. and the hemoglobin was 35%. After 3 more injections at weekly intervals the red blood cell count was 3.91 millions per cu. mm. and the hemoglobin was 74%.

Further studies are now being made on the properties of this concentrated extract, and it is being tested clinically for immediate response to treatment and also for maintenance of a normal red blood cell count.

Summary. A method for the preparation of liver extract based on the property of the hematopoietically active principle of becoming adsorbed by charcoal from an acid solution is described. This allows the concentration of the fluid to a small volume without much loss of potency.