faster and to a much greater extent than thiamine in man and in dogs. In rats, it was disclosed also that absorption of B₁ is rather limited to the upper intestine while that of DCET was efficient even in the ileum and colon.

Study on Human Gastric Intrinsic Factor Using Radioactive Vitamin B₁₂

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Purification of human intrinsic factor (IF) is of prime importance in the study of vitamin B₁₂ (B₁₂) metabolism. It is generally accepted that the cause of pernicious anemia (PA) is the decreased secretion or lack of IF in the gastric juice. Pepsin-inactivated and neutralized normal human gastric juice (NHGJ) was dialyzed, lyophilized, added ⁵⁷⁷Co or ⁶⁰⁶⁰Co B₁₂ at room temperature for 30 minutes. This mixture was filtered through Sephadex G-100 gel. Bound B₁₂ peak was named Fraction I, pooled, concentrated with Carbowax-6000 and chromatographed through DEAE-cellulose. A single peak of radioactivity was obtained and named Fraction II. Fraction II was re-concentrated with Carbowax-6000 or by lyophilization and finally filtered through Sephadex G-50, producing Fraction III in similar meanings. Fraction II was found active at 1.5 mg in a total gastrectomy patient by Schilling test. The ultracentrifugation of Fraction III revealed that it consisted of a single protein peak and its sedimentation coefficient was 10.1 S, and the molecular weight was about 155,000. In order to study if Fraction III and PA gastric juice (PAGJ) have IF activity or not, in vitro assay was performed using everted sacs of guinea pig intestine. The incubation media consisted of either 1000 pg B₁₂ alone, 1000 pg B₁₂ + NHGJ, 1000 pg B₁₂ + Fraction III or 1000 pg B₁₂ + PAGJ. Average B₁₂—uptake by the sac was 59.4 pg, 202.7 pg, 211.5 pg or 13.6 pg, respectively. This shows Fraction III is sufficiently active as IF. On the other hand, the B₁₂ uptake by the sac from PAGJ and B₁₂ containing medium was, on the average, 13.6 pg, rather lower than when B₁₂ alone was added to the medium. This decrease in B₁₂ uptake by the sac from JAGJ may be accounted for by the possible presence of non-IF B₁₂ binding substances, although PAGJ contains no more proteins than NHGJ. Non-IF B₁₂ binding substances may inhibit the uptake of B₁₂ by the sac. Taking this fact into consideration, we proceeded to the subsequent experiment. After being chromatographed through Sephadex G-100 and DEAE-cellulose, PAGJ was separately pooled for radioactive B₁₂ peak and for protein peak which appeared after the B₁₂ peak. Each pool was studied for IF activity by the everted sacs of guinea pig intestine. Inhibitory effect of each pool was shown by depressed B₁₂ uptake by the sac, but the protein pool was more inhibitory as compared with radioactive B₁₂ pool. This suggests the possible presence in PAGJ of B₁₂ utilization-inhibitory substances. We were not convinced of the presence in PA serum of what is called antibody against IF as long as immunologically obtained data are concerned, however.