repletion, etc.). This possibility is now being investigated.

*Summary.* 1. A microbiological method to assess the biological value of proteins is described. It implies the digestion of proteins with pancreatin and subsequent testing of the growth-promoting activity of the hydrolysate for *S. faecalis*, in a medium devoid of the 10 essential amino acids. The nutritive value of the protein digest is determined in a single assay by the amino acid liberated in a limiting concentration. 2. The results obtained with casein, egg albumin, gelatin, gluten, and zein are presented and their agreement with the biological values obtained by the rat growth method is suggested. 3. The growth promoting activity (biological valued) of casein and egg albumin is high and about equal; gelatin and gluten, in comparison, are much less active, 29 and 15%, respectively, and zein still less (2%). 4. At the limited growth level, casein is deficient in: lysine, arginine, isoleucine, and threonine. Egg albumin under comparable conditions is limited in lysine, arginine, histidine, threonine, isoleucine, leucine, and valine. Gelatin is low in tryptophane, isoleucine, leucine, methionine, and threonine. Gluten and zein are short in lysine. 5. The suitability of the method described for studying imbalances of amino acids is proposed.

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## Induction of Diabetes Insipidus in Adrenalectomized Dogs with Cortisone.* (20182)

W. W. SWINGLE, E. J. FEDOR, MAX BEN, ROBERT MAXWELL, CARLETON BAKER, AND GEORGE BARLOW.

*From the Biological Laboratory, Princeton University, Princeton, N. J.*

Large daily doses of DCA, when administered to intact dogs over a sufficiently prolonged interval, induce a syndrome of polyuria and polydipsia apparently identical with that of diabetes insipidus. According to Ragan *et al.*(1), pituitrin is relatively ineffective in controlling the polyuria and fluid restriction does not cause dehydration. Mulinos *et al.*(2) observed that withdrawal of salts from the diet reduced the severity of the polyuria and that large doses of pitressin in oil led to reduction of water intake and urine volume.

Recently it has been shown that daily injections of cortisone given to normal dogs for periods ranging from 2-3 weeks also bring about a marked polyuria and polydipsia(3,4). Sirek and Best(4) report that 10 I.U. of posterior pituitary hormone administered to one dog for one day restored almost normal conditions so far as thirst, polyuria and urine specific gravity were concerned.

*Material and methods.* Four mongrel male dogs were used in the present experiments; they had been adrenalectomized for periods varying from 8 months to 2 years. The animals were kept in metabolism cages and fed a special diet consisting of Warner's dog food,

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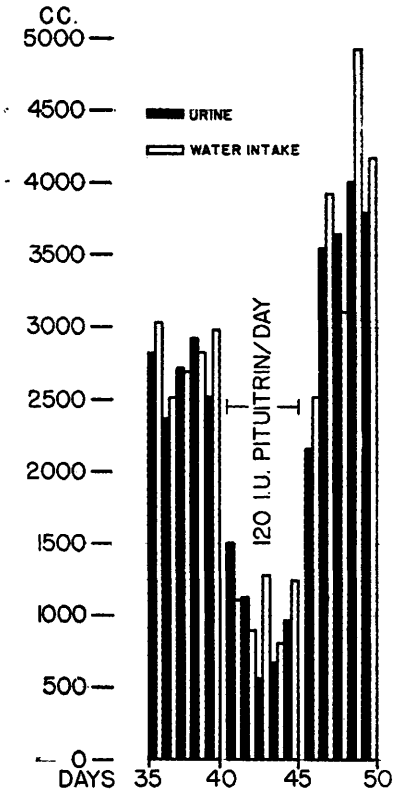


FIG. 1. Control of polyuria and polydipsia with pituitrin. Experiment started on 36th day of cortisone treatment.

horse meat and tripe. All necessary vitamins were added plus one teaspoonful of yeast. The daily ration (435 g, wet weight) contained 0.93 g of Na and 0.94 g of K. Water to the extent of 145 cc was added for mixing the food into a coarse mush. The water drunk and the urine voided during each 24-hour interval were measured and due allowance made for any evaporation and also for the water included in the food. The various methods employed for determining urine electrolytes are: Na(5), Cl(6), K(7). A fore-period of 5 or 6 days was allowed each dog when first placed in the metabolism cage; during this time the animals were fed the low Na and K ration and received the usual maintenance dose of 0.5 mg/dog/day of DCA. The control values for the various urine constituents were determined during this interval. The figures in Table I for water intake and output and urine electrolytes each represent weekly averages of the 7 daily 24-hour specimens. A microcrystalline suspension of cortisone-acetate (Merck's cortone) was administered once daily, intramuscularly, in doses of 10 mg/kg/day.

*Results. Polyuria and polydipsia.* Marked polyuria and polydipsia occurred within the first 10-14 days of cortisone therapy. Representative data, obtained from 2 of the 4 dogs studied, are given in Table I. Within the first 21 days the amount of urine, voided each 24 hours, was 5-10 times that eliminated during the same time interval of the control period. The maximum quantity of urine for any single 24-hour period passed by each of the 2 dogs listed, was as follows: Dog 1, 1790 cc; Dog 2, 2795 cc. Following withdrawal of cortisone at the end of the 6th-7th week, a sharp decline in water intake and urine output took place (Table I). The animals were not permitted to develop severe symptoms of adrenal insufficiency; when the blood pressure had declined to 80-90 mm Hg and the serum Na to values between 130-135 meq/1, the experiment was terminated. It usually required 20-30 days after discontinuing the cortisone before symptoms appeared. Apparently this length of time is required for complete absorption of the considerable amount of microcrystalline material injected.

*Electrolytes of the urine.* Daily urinary excretion of Na and Cl increased under the stimulus of the large doses of cortisone during the period of profound polyuria (Table I). However, in this same interval renal elimination of water increased 5-10 fold per 24 hours; hence, the dogs were excreting a highly dilute urine insofar as Na and Cl were concerned. When cortisone was withdrawn, an increase in the daily excretion of both of these electrolytes took place. The renal excretion of K was studied in but 2 animals (No. 1 and 2, Table I). In both there was an increased K elimination during the first 4 weeks after cortisone administration, but this was not sustained throughout the entire period of treatment. During the last 2-3 weeks, previous to termination of the experiment, urinary excretion of K declined to a value approximately that of the control fore-period.

*Control of the diabetes insipidus by posterior pituitary extract.* An attempt was made

TABLE I. Electrolyte Excretion of 2 Adrenalectomized Dogs Exhibiting a Diabetes Insipidus-Like Syndrome Due to Injecting 10 mg Cortisone per kg/day.

| Wt, kg | Exp. period, wk | Urine electrolytes, avg meq/24 hr sample | | | Avg urine vol, cc/day | Avg water intake, cc/day |
|---|---|---|---|---|---|---|
| | | Na | Cl | K | | |
| 10.7 | 1* | 31.8 | 26.2 | 13.5 | 146 | 226 |
| | 1† | 37.6 | 41.2 | 29.0 | 341 | 364 |
| | 2 | 46.1 | 47.5 | 19.7 | 934 | 965 |
| | 3 | 49.4 | 52.6 | 17.6 | 866 | 997 |
| | 4 | 46.7 | 56.4 | 17.1 | 1386 | 1565 |
| | 5 | 48.3 | 52.3 | 13.4 | 1199 | 1350 |
| | 6 | 40.3 | 45.8 | 15.7 | 923 | 1045 |
| | 7 | 41.6 | 49.3 | 13.7 | 1139 | 1257 |
| | 1‡ | 56.2 | 65.3 | × | 683 | 705 |
| | 2 | 48.7 | 55.2 | × | 516 | 591 |
| | 3 | 52.7 | 54.5 | × | 708 | 797 |
| 12.7 | 1* | 30.9 | 31.7 | 17.5 | 195 | 403 |
| | 1† | 49.4 | 62.8 | 29.9 | 1049 | 1025 |
| | 2 | 55.5 | 64.5 | 19.6 | 2036 | 2141 |
| | 3 | 43.5 | 46.3 | 19.2 | 1206 | 1305 |
| | 4 | 66.8 | 67.7 | 20.0 | 1301 | 1367 |
| | 5 | 63.8 | 68.6 | 15.6 | 1215 | 1275 |
| | 6 | 78.6 | 79.1 | 14.5 | 1202 | 1226 |
| | 1‡ | 80.4 | 85.0 | × | 956 | 1140 |
| | 2 | 92.8 | 90.9 | × | 585 | 693 |
| | 3 | 81.1 | 78.7 | × | 507 | 675 |

* DCA fore-period. † Cortisone started.
‡ Cortisone discontinued. × = not determined.

to control the polyuria and polydipsia of 2 animals by pituitrin. The pertinent data obtained from study of one dog are shown in Fig. 1 and they reveal the striking effect of posterior pituitary extract on the syndrome. The average daily urine volume for 5 days preceding pituitrin injection was 2672 cc; whereas, it promptly declined to an average of 971 cc during the 5-day subcutaneous administration of 120 I.U. of the pituitrin in divided doses of 40 I.U. each. After withdrawal of the extract the average daily urine volume rose to 3432 cc. Smaller daily doses of 5-20 I.U. of pituitrin, twice each day, had little or no effect upon the polyuria and polydipsia of Dog 3 (not listed in Table I). However, 40 I.U., 3 times daily, exerted a noticeable effect.

*Discussion.* Britton and co-workers(8,9) were the first to advance the theory that a normal water balance is maintained owing to a physiological antagonism between the diuretic action of adrenal cortical hormones and the antidiuretic action of posterior pituitary hormone. Recent work by Gaunt, Birnie, and Eversole(10) and others have tended to substantiate the main points of the theory. Thus, when large doses of cortisone (or DCA(1,2)) are administered, they greatly enhance urine volume by 1) augmenting thirst due to Na retention; 2) inhibiting tubular reabsorption of water; and 3) probably by reducing greatly the amount of antidiuretic hormone released by the posterior pituitary. Hence, the diabetes insipidus is due to an upset in the physiological balance between the 2 antagonists controlling water balance. Since cortisone in massive doses acts as a diuretic, it would presumably overwhelm the antidiuretic function of the posterior pituitary and eventuate in a marked polyuria and polydipsia. Factors other than those mentioned are also probably involved, but since they are unknown, it is futile to speculate concerning them.

*Summary.* High cortisone dosage in adrenalectomized dogs induces marked polyuria and polydipsia. The 24-hour urine volume may exceed control values by 5-10 fold. The daily renal excretion of Na, Cl, and K is increased, but considering the quantity of water eliminated, the urine is very dilute. Pituitrin is effective in controlling the polyuria and polydipsia only when administered in large amounts. The normal water balance was not re-established until long after cortisone injections were discontinued.

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