# ELECTROLYTE BALANCE STUDIES IN ADRENALECTOMIZED DOGS WITH PARTICULAR REFERENCE TO THE EXCRETION OF SODIUM

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A characteristic clinical picture develops as the loss of inorganic base from the body progresses in diabetic acidosis, in severe diarrheas (11), in high intestinal obstruction and probably in certain other pathological states. The changes encountered are profound weakness, prostration, dehydration, anorexia, nausea, vomiting, fall in blood pressure, shock at times accompanied by anuria, retention of non-protein nitrogen, decrease in the concentration of the chloride ion in the blood and frequently a fall in the bicarbonate content of the serum. In view of the fact that this clinical picture is also characteristic of that present in severe adrenal insufficiency, the blood of three patients suffering from Addison's disease was studied with the result that a striking decrease in the sodium content of the blood serum was found (1). Marine and Baumann (2), in 1927, showed similarly that the sodium content of the blood of cats decreased following bilateral adrenalectomy, and suggested that sodium salts might have a "specific action" in prolonging the life of these animals. Zwemer (3) has also found, in addition to other abnormalities, a decrease in the sodium concentration of the blood of adrenalectomized cats. More recently, Harrop and Weinstein (4) have shown a slight drop in the total base of adrenalectomized dogs.

It is thus apparent that a decrease in the sodium concentration of the blood is a constant finding in both Addison's disease and in adrenalectomized animals. Since these observations have been limited entirely to the examination of the blood, nothing is known of the proc-

esses involved in bringing about this and other profound physiological changes encountered. The present work was undertaken to correlate, if possible, the described alterations in the blood with the excretion of various ions, particularly sodium, occurring during the development of adrenal insufficiency. For this purpose relatively complete balance studies have been made on dogs before and after adrenalectomy.

## EXPERIMENTAL

Most of the analytical methods employed have been described elsewhere (5–7). Sodium was determined according to the method of Butler and Tuthill (8). The urine, stool and food specimens used for the determination of sodium were ashed in the muffle oven at 470°C. before analysis and potassium was removed from the urine, because of its high concentration.

Adult male dogs weighing between 13 and 15 kilos served as the experimental subjects. The animals were given diets consisting solely of ground, raw, lean beef and received daily rations of 400 or 500 gm. for periods of 2 to 14 weeks before balance studies were begun. On this diet they remained in good health and their general nutrition improved. At the beginning of the observations sufficient meat to last 7 days was ground up and thoroughly mixed. The daily rations were then weighed carefully, wrapped in waxed paper and kept on ice until used. A sample of each lot of meat was dried and analyzed for total inorganic base, Na, K, Ca, Cl and total nitrogen. Water was allowed *ad lib.* The animals ate their food with relish until the 4th day after the second adrenal gland was removed, when their appetites decreased rapidly. On only one occasion was vomiting encountered in this work. Dog 2 vomited undigested meat once on the 5th day following the second adrenalectomy. The vomitus was dried and analyzed and deductions were made from the food intake.

When the experiments were started the animals were catheterized and placed in the metabolism cage, where they remained continuously. The urine was collected under toluol and at the end of each period the dogs were catheterized. The cage was scrubbed and washed with 1500 cc. of distilled water, except in the case of Dog 1, where it was merely rinsed with 500 cc. of distilled water, and these washings were added to the urine of the period. Urine specimens were analyzed for total inorganic base, Na, K, Ca, Cl and total nitrogen.

Stools were collected during each period and were usually formed. Occasionally there were soft stools and slight admixture with urine specimens resulted in these instances. This had little quantitative or qualitative effect upon the results, as the tables show that the stools contained only minute amounts of the electrolytes other than calcium. In the case of Dog 2, a little bloody mucus was passed following the second adrenalectomy. Stools were analyzed similarly to the food and urine specimens.

Blood studies were carried out on 50 to 80 cc. samples of blood removed from the

jugular vein with oiled syringes and delivered under paraffin oil. The analyses were made in duplicate on the serum thus obtained.

Adrenalectomies were performed by Dr. Rudolph Schullinger of the Department of Surgery with ether anesthesia and with rigid surgical asepsis. In no instance was there hemorrhage or surgical shock, and all of the animals ate heartily within a few hours after the operations. Dog 3 had a superficial wound infection after the first adrenalectomy, many weeks before the period of metabolic observation began. Dogs 2 and 3 had some serosanguineous ooze from their wounds following the second operation. Salivation during anesthesia was marked and fluid thus lost was not analyzed. Autopsies were performed on the dogs and careful search revealed no evidence of accessory adrenal tissue. In Dog 2, the small intestine contained a moderate amount of "currant jelly" bloody mucus. Dog 3 showed beginning congestion of the right lower lobe.

## RESULTS

*Dog 1.*—Balance studies were made on this animal for more than 1 month before the removal of either adrenal gland. Fig. 1 and Tables I and II show the results of this study. *Sodium*: The dog appeared to be in slightly positive Na balance (2 m.-eq. per day) during the fore period, and the irregularities observed are probably due, in part, to the fact that in this case the metabolism cage was merely rinsed and not scrubbed at the end of each period. It will be seen that the first adrenalectomy, performed on the 7th day of Period V, did not result in obvious change in Na balance when compared with the other control periods. Following the second adrenalectomy, the dog went into negative sodium balance, losing about 65 m.-eq. in the course of a week, whereas the average positive balance in the fore periods would have been about 14 m.-eq. in the same period of time. *Potassium*: The experiment on this dog was complicated by the attempt to increase the excretion of Na by the administration of large amounts of KCl. During Periods III and IV the animal was fed 5 gm. of KCl daily with its ration of 500 gm. of lean meat. During Period V this was increased to 7.0 gm. and in Period VI the dose was again reduced to 5.0 gm. That this feeding of KCl had no effect upon Na excretion is obvious from the data. Following the second adrenalectomy, the excretion of K decreased in contrast to the increasing Na excretion, but was nevertheless greater than the intake. *Chloride*: The chloride excretion in this animal was naturally complicated by the KCl administration, but it may be seen from the table that, as was true of the Na ion, the excretion actually increased over the levels of the control periods after the second adrenalectomy. *Calcium*: This animal, as well as the other dogs, was in constant negative Ca balance, as might be expected as a result of the meat diet. The effect of adrenalectomy on the calcium balance is not apparent, and if existent is masked by the inaccuracy of stool collections. *Nitrogen*: Approximate nitrogen balance was maintained throughout the experiment, even during Period VIII when the intake was reduced following the second adrenalectomy. *Water*: During the first 4 days of Period VIII, following

TABLE I

*Balance Study on Dog 1*

| Inclusive dates | Period | Urine | | | | | | | Stool | | | | | | | Intake | | | | | | Body wt. | Remarks |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Vol. | K | Na | Ca | T.B.* | Cl | T.N | Dry Wt. | K | Na | Ca | T.B.* | Cl | T.N | K | Na | Ca | T.B.* | Cl | T.N | | |
| *1932* | | *cc.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *gm.* | *gm.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *gm.* | *gm.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *gm.* | *kg.* | |
| | I | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 332 | 43.5 | 7.9 | 1.49 | 62.3 | 5.2 | 18 1 | 3.7 | 0.12 | 0.20 | 3.6 | 8.6 | 0.1 | 0.19 | 43.2 | 9.3 | 1.1 | 65.3 | 4.6 | 16.2 | | |
| Sept. 16–21 | Total | 1990 | 260.7 | 47.3 | 8.94 | 373.6 | 30.9 | 108.5 | 22.0 | 0.71 | 1.21 | 21.7 | 51.5 | 0.5 | 1.13 | 259.2 | 55.8 | 6.6 | 391.8 | 27.6 | 97.2 | | |
| | II | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 313 | 47.0 | 7.7 | 1.69 | 60.2 | 4.8 | 17.9 | 2.9 | 0.06 | 0.08 | 3.2 | 7.3 | 0.1 | 0.19 | 43.4 | 10.6 | 1.4 | 65.0 | 5.4 | 16.7 | | |
| Sept. 22–28 | Total | 2190 | 329.0 | 54.0 | 11.80 | 421.2 | 33.4 | 125.4 | 20.0 | 0.45 | 0.59 | 22.2 | 51.0 | 0.5 | 1.18 | 303.6 | 74.1 | 9.5 | 455.3 | 37.6 | 117.0 | 12.9 | |
| | III | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 334 | 114.0 | 5.8 | 1.47 | 123.2 | 65.4 | 17.6 | 4.4 | 0.23 | 0.10 | 6.6 | 13.7 | 0.1 | 0.23 | 112.9 | 10.3 | 1.5 | 135.3 | 72.7 | 17.3 | | 5.0 gm. of KCl ingested daily with diet |
| Sept. 29–Oct. 5 | Total | 2335 | 798.0 | 40.9 | 10.30 | 862.3 | 457.8 | 123.4 | 31.0 | 1.60 | 0.72 | 46.1 | 95.5 | 0.7 | 1.58 | 790.4 | 72.0 | 10.4 | 946.8 | 508.7 | 121.2 | 12.9 | |
| | IV | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 307 | 107.5 | 9.7 | 0.88 | 124.2 | 73.4 | 16.3 | 3.1 | 0.04 | 0.05 | 4.3 | 9.2 | 0.1 | 0.18 | 110.6 | 11.0 | 1.6 | 131.8 | 72.2 | 16.3 | | 5.0 gm. of KCl ingested daily with diet |
| *Oct. 6–12* | Total | 2150 | 752.3 | 67.9 | 6.13 | 869.1 | 514.2 | 114.1 | 22.0 | 0.31 | 0.37 | 30.1 | 64.4 | 0.3 | 1.25 | 774.4 | 76.8 | 11.1 | 922.4 | 505.3 | 115.8 | 12.9 | |
| | V | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 346 | 122.7 | 11.9 | 0.44 | 140.6 | 94.7 | 15.3 | 3.2 | 0.18 | 0.10 | 4.7 | 9.8 | 0.1 | 0.20 | 134.7 | 12.6 | 1.5 | 156.6 | 100.3 | 16.9 | | 7.0 gm. of KCl ingested daily with diet. 1st adrenalectomy on 7th day of this period |
| Oct. 13–21 | Total | 3110 | 1103.9 | 106.9 | 4.00 | 1265.0 | 852.0 | 137.9 | 29.0 | 1.60 | 0.87 | 42.7 | 87.9 | 0.6 | 1.80 | 1212.4 | 113.4 | 13.4 | 1409.5 | 902.9 | 152.3 | 12.7 | |

| | | | | | | | | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VI | | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 331 | 103.2 | 4.1 | 0.99 | 121.0 | 70.2 | 16.7 | 3.5 | 0.26 | 0.10 | 5.1 | 12.0 | 0.1 | 0.19 | 111.4 | 9.9 | 2.0 | 135.4 | 72.1 | 17.6 | | 5.0 gm. of KCl ingested daily with diet |
| Oct. 22–27 | Total | 1985 | 619.0 | 24.5 | 5.94 | 725.9 | 421.0 | 100.0 | 21.0 | 1.55 | 0.59 | 30.6 | 71.9 | 0.9 | 1.16 | 668.4 | 59.4 | 12.0 | 812.4 | 432.6 | 105.6 | 12.3 | |
| | VII | | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 244 | 42.0 | 10.7 | 1.11 | 59.6 | 9.4 | 14.3 | 2.4 | 0.05 | 0 15 | 4.0 | 7.7 | 0.1 | 0.13 | 39.3 | 11.8 | 1.4 | 62.8 | 5.1 | 15.5 | | |
| Oct. 28–Nov. 3 | Total | 1705 | 294.0 | 75.0 | 7.76 | 417.2 | 65.9 | 99.9 | 17.0 | 0.33 | 1.02 | 28.0 | 53.8 | 0.6 | 0.94 | 275.1 | 82.6 | 9.8 | 439.6 | 35.7 | 108.5 | 12.9 | |
| | VIII | | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 293 | 24.1 | 13.8 | 0.36 | 44.5 | 11.8 | 7.8 | 3.7 | 0.05 | 0.19 | 5.8 | 10.8 | 0.1 | 0.26 | 18.4 | 4.7 | 0.73 | 28.8 | 2.1 | 7.0 | | 2nd adrenalectomy at the beginning of this period. Weighed on the 4th day of this period |
| Nov. 4–10 | Total | 2050 | 168.7 | 96.4 | 2.54 | 311.5 | 82.6 | 54.8 | 26.0 | 0.38 | 1.30 | 40.6 | 75.8 | 0.7 | 1.82 | 128.8 | 32.9 | 5.11 | 201.6 | 14.7 | 49.0 | 11.9 | |

* T.B. = total inorganic base.

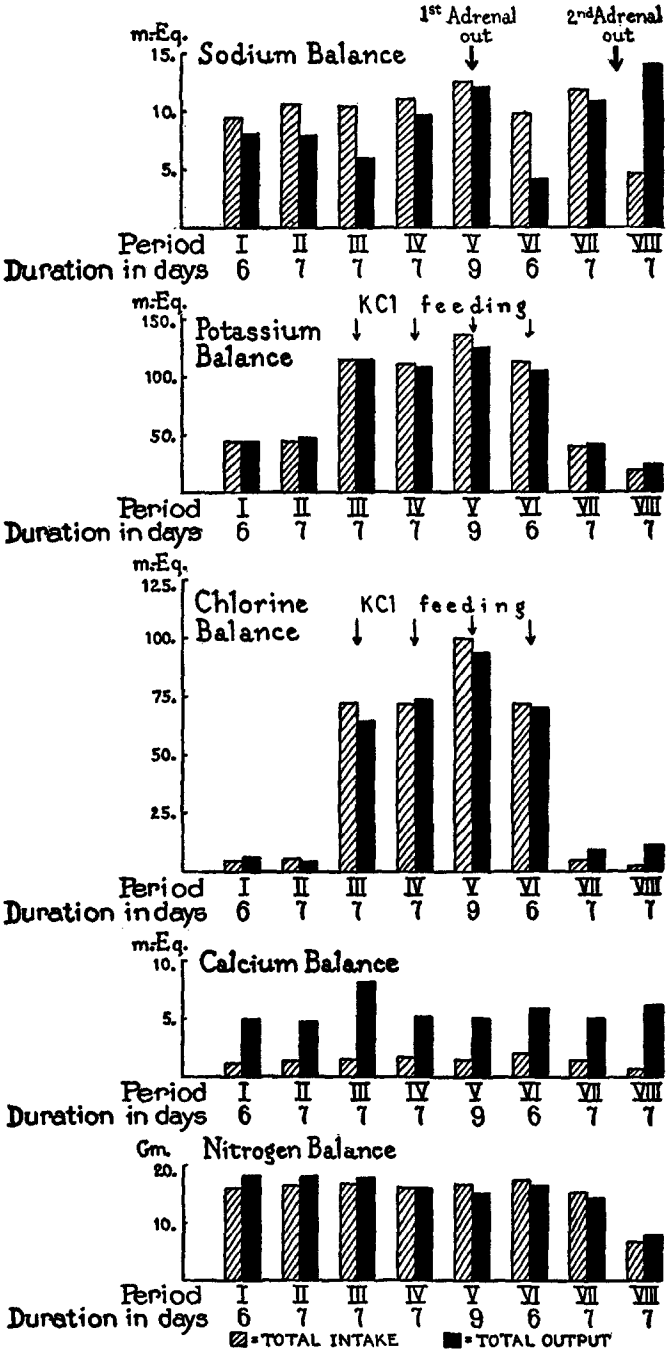


FIG. 1. Balance studies on Dog 1.

the second adrenalectomy, the urine volume was augmented from a daily average of 315 cc. in the fore period to 405 cc. Thereafter, when the animal became severely ill and took neither food nor fluid, the volume fell sharply. This accounts for the fact that the daily volume as presented in Fig. 4 is less following adrenalectomy than before, in contrast with the other animals in which the periods presented in the figure were closed before the animals reached a moribund state. Fig. 4 shows that in addition to the actual increase in sodium excretion following removal of the second adrenal gland there is also an increase in relation to the water output. No such change was noted as a result of the operative procedure at which the first gland was removed. *Blood*: Numerous studies of the blood were made (Table II) and show no significant variations before the second adrenalectomy.

TABLE II

*Observations on the Blood of Dog 1*

| Date | Total base | Total acid | Non-protein nitrogen | Protein | Protein | $HCO_3$ | Cl | K | Na | Remarks |
|---|---|---|---|---|---|---|---|---|---|---|
| *1932* | *m.-eq. per liter* | *m.-eq. per liter* | *mg. per 100 cc.* | *per cent* | *m.-eq. per liter* | *m.-eq. per liter* | *m.-eq. per liter* | *m.-eq. per liter* | *m.-eq. per liter* | |
| Sept. 20 | 162.3 | 154.1 | 28 | 7.86 | 18.6 | 26.1 | 109.4 | 5.2 | 148.0 | |
| Sept. 28 | 158.5 | 152.8 | 30 | 7.34 | 17.3 | 26.5 | 109.0 | 4.5 | 149.0 | |
| Oct. 6 | 156.3 | 149.0 | 37 | 6.58 | 15.5 | 24.7 | 108.8 | 4.5 | 146.6 | |
| Oct. 19 | 158.8 | 150.2 | 31 | 6.74 | 15.9 | 25.0 | 109.3 | 4.8 | 147.9 | 1st adrenalectomy on Oct. 20 |
| Nov. 2 | 156.3 | 149.7 | 26 | 6.78 | 16.0 | 25.2 | 108.5 | 4.9 | 147.3 | |
| Nov. 11 | | | 214 | 7.31 | 17.3 | | 88.8 | 15.4 | 127.5 | 2nd adrenalectomy on Nov. 4. Last blood taken after death |

The last blood sample was unfortunately taken from the right heart shortly after death on the morning of the 8th day after operation. The changes are all similar to those seen in the other animals, except that the K content of the blood was greatly increased. As the blood was slightly hemolyzed and was taken after death, this determination is of little value. The drop in Na and Cl concentrations was 20 m.-eq. per liter, and was accompanied by an increase in non-protein nitrogen to 214 mg. per 100 cc. *Stools*: Analyses of the fecal matter after adrenalectomy failed to reveal changes from those of the fore period and the actual amounts of Na, K and Cl excreted were minimal.

*Dog 2.*—The first adrenal gland was removed from this animal 1 month before the metabolic observations were begun and it was kept on the standard meat diet for 3 weeks before the beginning of the balance study, the results of which are shown in Fig. 2 and Tables III and IV. During the fore period, which lasted 27 days,

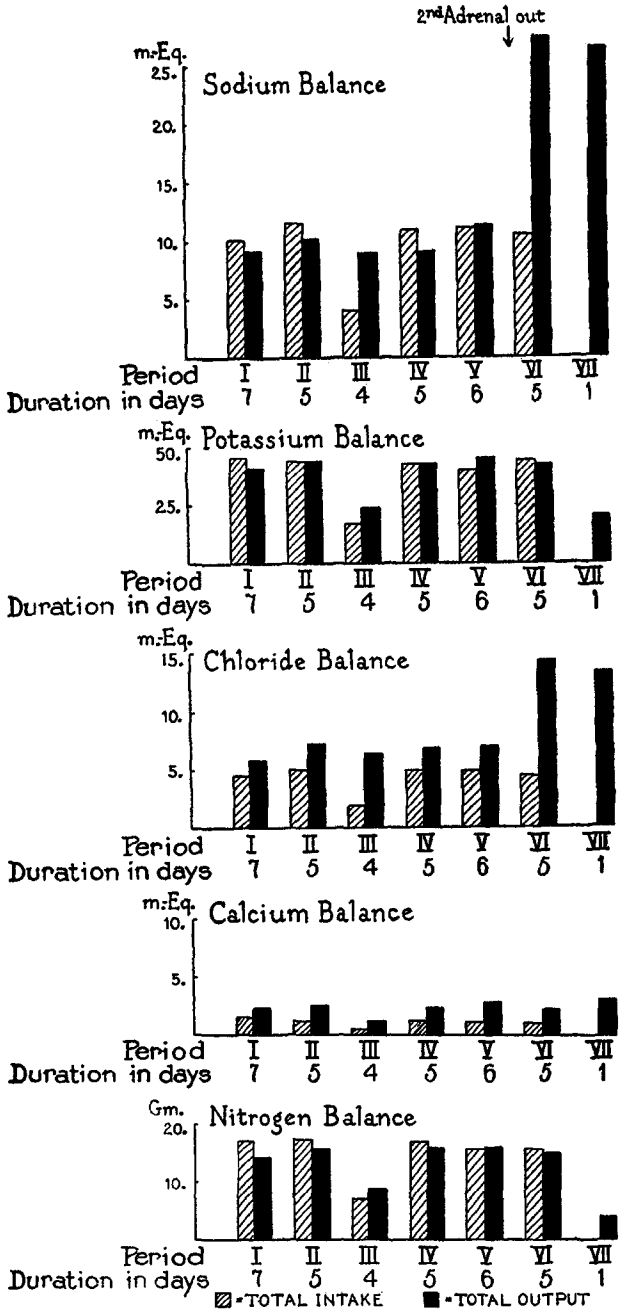


FIG. 2. Balance studies on Dog 2.

TABLE III

*Balance Study on Dog 2*

| Inclusive dates | Period | Urine | | | | | | | Stool | | | | | | | Intake | | | | | | Body wt. | Remarks |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Vol. | K | Na | Ca | T.B.* | Cl | T.N | Dry wt. | K | Na | Ca | T.B.* | Cl | T.N | K | Na | Ca | T.B.* | Cl | T.N | | |
| *1932* | | *cc.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *gm.* | *gm.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *gm.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *gm.* | *kg.* | |
| | I | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 295 | 40.4 | 8.3 | 0.66 | 55.7 | 5.84 | 14.3 | 3.4 | 0.20 | 0.73 | 1.70 | 6.24 | 0.14 | 0.35 | 44.0 | 9.96 | 1.47 | 63.8 | 4.52 | 16.9 | | |
| Nov. 14–20 | Total | 2065 | 282.9 | 58.4 | 4.59 | 390.4 | 40.91 | 100.0 | 25.0 | 1.42 | 5.12 | 11.91 | 43.71 | 0.95 | 1.73 | 308.0 | 69.72 | 10.29 | 446.8 | 31.64 | 118.3 | 14.2 | |
| | II | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 336 | 42.9 | 9.2 | 1.28 | 62.1 | 7.05 | 15.9 | 2.6 | 0.41 | 0.90 | 1.23 | 5.09 | 0.36 | 0.24 | 43.7 | 11.36 | 1.36 | 69.4 | 5.00 | 17.6 | | |
| Nov. 23–27 | Total | 1680 | 214.7 | 45.8 | 6.42 | 310.2 | 35.30 | 79.5 | 13.0 | 2.05 | 4.48 | 6.16 | 25.46 | 1.78 | 1.20 | 218.5 | 56.80 | 6.80 | 347.0 | 25.00 | 88.0 | 13.7 | |
| | III | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 186 | 23.4 | 8.6 | 0.67 | 38.5 | 6.15 | 8.9 | 1.5 | 0.14 | 0.21 | 0.57 | — | 0.05 | 0.13 | 16.7 | 3.97 | 0.52 | 26.9 | 1.86 | 6.9 | | |
| Nov. 28–Dec. 1 | Total | 742 | 93.6 | 34.3 | 2.68 | 153.8 | 24.60 | 35.6 | 6.0 | 0.55 | 0.84 | 2.33 | — | 0.21 | 0.53 | 66.8 | 15.88 | 2.08 | 107.6 | 7.44 | 27.7 | 13.1 | |
| | IV | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 332 | 42.6 | 8.5 | 0.93 | 63.7 | 6.82 | 16.1 | 2.8 | 0.15 | 0.62 | 1.41 | 4.29 | 0.03 | 0.23 | 42.7 | 10.82 | 1.35 | 70.0 | 4.94 | 17.2 | | |
| Dec. 2–6 | Total | 1660 | 212.8 | 42.7 | 4.64 | 318.6 | 34.10 | 80.8 | 14.0 | 0.73 | 3.09 | 7.03 | 21.43 | 0.16 | 1.15 | 211.7 | 54.11 | 6.75 | 349.9 | 24.69 | 85.8 | 13.4 | |
| | V | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 281 | 42.7 | 10.4 | 0.77 | 62.9 | 6.92 | 15.7 | 4.0 | 0.51 | 0.92 | 2.15 | 7.21 | 0.24 | 0.32 | 40.0 | 11.12 | 1.28 | 66.5 | 4.86 | 15.7 | | |
| Dec. 7–12 | Total | 1690 | 256.0 | 62.4 | 4.62 | 377.6 | 41.50 | 94.2 | 24.0 | 3.07 | 5.51 | 12.88 | 43.28 | 1.42 | 1.92 | 240.0 | 66.73 | 7.70 | 398.8 | 29.14 | 94.1 | 13.0 | |
| | VI | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 456 | 40.4 | 26.9 | 0.65 | 77.4 | 13.9 | 14.5 | 4.6 | 0.41 | 0.45 | 1.76 | 6.86 | 0.24 | 0.76 | 42.7 | 10.40 | 1.20 | 62.3 | 4.28 | 15.7 | | |
| Dec. 13–17 | Total | 2280 | 201.9 | 134.5 | 3.27 | 387.0 | 69.5 | 72.5 | 23.0 | 2.04 | 2.26 | 8.81 | 34.31 | 1.19 | 3.80 | 213.8 | 52.02 | 6.00 | 311.3 | 21.40 | 78.7 | 12.4 | |
| | VII | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 345 | 19.0 | 24.7 | 1.82 | 51.1 | 13.2 | 4.84 | 4.0 | 0.66 | 1.79 | 1.36 | — | 0.72 | | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | | |
| Dec. 18 | Total | 345 | 19.0 | 24.7 | 1.82 | 51.1 | 13.2 | 4.84 | 4.0 | 0.66 | 1.79 | 1.36 | — | 0.72 | | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 0.0 | 12.2 | 2nd adrenalectomy at the beginning of this period |

* T.B. = total inorganic base.

the food intake during Period III (4 days) was reduced from 500 gm. of meat to 250 gm. in order to note the effects of reduced food intake upon the balance. Following this, the food intake was raised to 500 gm. daily and after 11 days (Periods IV and V) the second adrenalectomy was performed. In this dog vomiting occurred once on the 5th day after operation, as was mentioned above, and the stools were small and loose. The factor of reduced diet fortunately did not occur in this dog during Period VI following the second adrenalectomy. *Sodium*: The Na balance, except in the period of reduced food intake, was constant and showed an apparent average retention of 1 m.-eq. daily. Following the removal of the second adrenal gland, the urinary Na excretion increased from an average of 10.8 m.-eq. daily to 26.9 m.-eq. and the Na lost from the body in the first 5 days after operation was 85 m.-eq. On

TABLE IV

*Observations on the Blood of Dog 2*

| Date | Total base | Total acid | Base minus acid | Protein | Protein | Non-protein nitrogen | Cl | $HCO_3$ | K | Na | Remarks |
|---|---|---|---|---|---|---|---|---|---|---|---|
| *1932* | *m.-eq. per liter* | *m.-eq. per liter* | *m.-eq. per liter* | *per cent* | *m.-eq. per liter* | *mg. per 100 cc.* | *m.-eq. per liter* | *m.-eq. per liter* | *m.-eq. per liter* | *m.-eq. per liter* | |
| Oct. 11 | 157.0 | 142.2 | 14.8 | 5.24 | 12.4 | 16 | 106.4 | 23.4 | 5.1 | 144.6 | 1st adrenalectomy on Oct. 12 |
| Oct. 25 | 158.0 | 146.0 | 12.0 | 6.17 | 14.6 | 33 | 107.9 | 23.5 | 5.2 | 146.5 | |
| Dec. 18 | 144.3 | 128.9 | 15.4 | 6.28 | 14.8 | 185 | 100.0 | 14.1 | 6.3 | 132.8 | 2nd adrenalectomy Dec. 12—Died Dec. 21 |

the 6th day, Period VII, the Na excretion was about that of the average for the first 5 days, though the stool excretion increased and no food was taken. *Potassium*: In contrast to the increased Na excretion following adrenalectomy, there was no increase in the loss of potassium, the balance being exactly like that of the fore period. This is in agreement with the findings in Dog 1, where the negative potassium balance could be explained by reduction in food intake. *Chloride*: The behavior of the chloride excretion was, as in the case of Dog 1, qualitatively like that of the Na ion; *i.e.*, there was an actual increase in excretion after adrenalectomy, though not equivalent to that of sodium. No explanation presents itself for the fact that this animal and Dog 3 were in slightly negative chloride balance before the second adrenalectomy. *Calcium*: The removal of the second adrenal gland did not appear to affect the negative Ca balance, present throughout the

experiment. *Nitrogen*: As in the case of Dog 1, nitrogen equilibrium was maintained during the first 5 days after operation, while the dog was still able to eat. *Water*: It may be seen from Fig. 4 and Table III that during the first 5 days following adrenalectomy the daily urine volume increased over the daily volume of the fore period. Nevertheless, the concentration of Na in the urine actually increased as it did in Dog 1. *Blood*: The sample of blood taken on the 6th day after the second adrenalectomy showed striking changes from the analyses made before and after the first adrenal gland had been removed (Table IV). The total inorganic base decreased 13 m.-eq. per liter, the Na concentration fell 12 m.-eq. per liter and the K increased 1 m.-eq. per liter. The fall in Na was accompanied by a loss of 7 m.-eq. per liter of chloride and about 9 m.-eq. in the concentration of bicarbonate. The non-protein nitrogen increased from a normal level to 185 mg. per 100 cc. and this occurred during a period of relative diuresis. The serum protein concentration increased but slightly. The fact that the difference between the total inorganic base of the blood serum and the summation of those acid radicals determined by analysis increased only slightly after the second gland was removed, suggests that the loss of Na was probably not the result of acidosis.

*Dog 3.*—The right adrenal gland was removed from this animal almost 3 months before the period of metabolic study was begun. It was given a standard diet of 500 gm. of lean meat 2 weeks before the collection of specimens was started and 6 days before the experiment began, this ration was decreased to 400 gm. daily and was maintained at this level. The dog was almost moribund on the morning of the 7th day after the second adrenalectomy and was chloroformed after the last sample of blood had been taken. This animal did not vomit and there was no diarrhea until the 5th day after the second operation. The data obtained from the study of this dog are presented in Fig. 3 and in Tables V and VI. *Sodium*: As in the other dogs, there was a striking increase in the sodium excreted in the urine during the first 4 days following the second adrenalectomy (Period III). The daily balance changed from an average of +1.1 m.-eq. in the fore period to −22.6 m.-eq. as insufficiency developed. The total loss of Na from the body in these 4 days amounted to 90.4 m.-eq. During the next 2 days when the dog had stopped taking nourishment, and after 80 cc. of blood had been removed on each of the 2 successive days, the Na excretion fell to 5.0 m.-eq. and 7.0 m.-eq. respectively. *Potassium*: As in the other dogs, there was no increase in K excretion following the second adrenalectomy, although the balance was negative, again probably resulting, at least in part, from the decrease in food intake. *Chloride*: Following the removal of the second adrenal gland, there resulted, as in Dogs 1 and 2, an increase in Cl excretion as compared with the fore period, and in this animal, too, it will be seen that this loss is not as great as that of sodium. Chloride excretion decreased during the last 2 days of life, as did the elimination of the sodium ion. *Calcium*: The negative Ca balance present in this animal is even more striking than in the other dogs and this quantitative difference might perhaps be explained by the fact that it had received a pure meat diet for a shorter time before

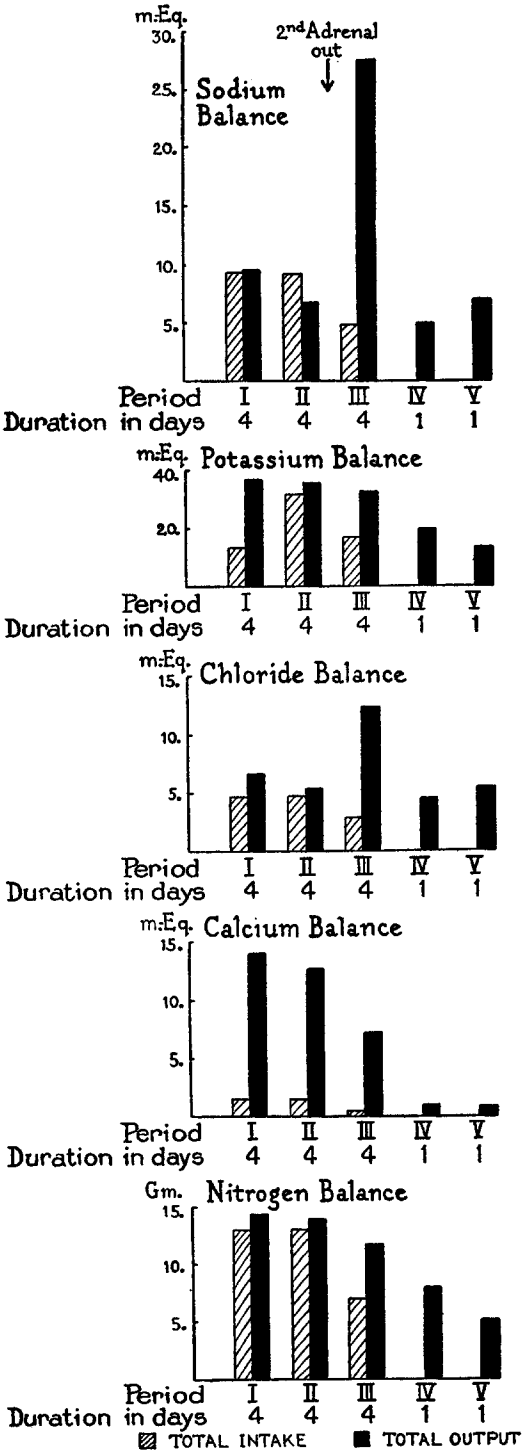


FIG. 3. Balance studies on Dog 3.

TABLE V

*Balance Study on Dog 3*

| Inclusive dates | Period | Urine | | | | | | | Stool | | | | | | | Intake | | | | | | Body wt. | Remarks |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | Vol. | K | Na | Ca | T.B.* | Cl | T.N | Dry wt. | K | Na | Ca | T.B.* | Cl | T.N | K | Na | Ca | T.B.* | Cl | T.N | | |
| *1933* | | *cc.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *gm.* | *gm.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *gm.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *m.-eq.* | *gm.* | *kg.* | |
| | I | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 276 | 36.3 | 9.0 | 1.66 | 54.4 | 6.6 | 13.9 | 7.5 | 0.04 | 0.43 | 12.2 | 22.1 | 0.11 | 0.48 | 33.5 | 9.16 | 1.56 | 50.6 | 4.80 | 13.1 | 15.0 | |
| Jan. 10–13 | Total | 1105 | 145.3 | 36.1 | 6.64 | 217.4 | 26.2 | 55.7 | 30.0 | 0.15 | 1.73 | 48.9 | 88.2 | 0.42 | 1.92 | 134.0 | 36.64 | 6.24 | 202.4 | 19.20 | 52.4 | 14.7 | |
| | II | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 284 | 35.5 | 6.4 | 1.85 | 52.7 | 5.3 | 13.3 | 8.3 | 0.04 | 0.36 | 10.8 | 19.1 | 0.14 | 0.55 | 33.5 | 9.16 | 1.56 | 50.6 | 4.80 | 13.1 | | |
| Jan. 14–17 | Total | 1135 | 142.1 | 25.5 | 7.38 | 210.6 | 21.2 | 53.1 | 33.0 | 0.14 | 1.43 | 43.0 | 76.4 | 0.56 | 2.18 | 134.0 | 36.64 | 6.24 | 202.4 | 19.20 | 52.4 | | |
| | III | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 353 | 32.4 | 27.1 | 0.95 | 65.3 | 14.4 | 11.5 | 4.3 | 0.02 | 0.27 | 6.1 | 8.9 | 0.08 | 0.32 | 17.2 | 4.76 | 0.57 | 29.8 | 2.88 | 6.9 | | 2nd adrenalectomy at the beginning of this period |
| Jan. 18–21 | Total | 1410 | 129.5 | 108.5 | 3.81 | 261.1 | 57.7 | 46.3 | 17.2 | 0.08 | 1.08 | 24.4 | 35.5 | 0.33 | 1.27 | 68.8 | 19.04 | 2.28 | 119.2 | 11.52 | 27.6 | | |
| | IV | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 275 | 19.4 | 5.00 | 1.06 | 29.8 | 4.7 | 7.24 | | | | | | | | | | | | | | | |
| Jan. 22 | Total | 275 | 19.4 | 5.00 | 1.06 | 29.8 | 4.7 | 7.24 | | | | | | | | | | | | | | | |
| | V | | | | | | | | | | | | | | | | | | | | | | |
| | Daily | 210 | 13 2 | 7.00 | 0.79 | 25 5 | 5.6 | 5.27 | | | | | | | | | | | | | | | |
| Jan. 23 | Total | 210 | 13.2 | | | 25.5 | 5.6 | 5.27 | | | | | | | | | | | | | | | |

* T.B. = total inorganic base.

TABLE VI

*Observations on the Blood of Dog 3*

| Date | Total base | Total acid | Base minus acid | Protein | Protein | Non-protein nitrogen | Cl | $HCO_3$ | K | Na | Remarks |
|---|---|---|---|---|---|---|---|---|---|---|---|
| *1932* | *m.-eq. per liter* | *m.-eq. per liter* | *m.-eq. per liter* | *per cent* | *m.-eq. per liter* | *mg. per 100 cc.* | *m.-eq. per liter* | *m.-eq. per liter* | *m.-eq. per liter* | *m.-eq. per liter* | |
| Oct. 9 | 153.5 | 143.6 | 9.9 | 5.76 | 13.6 | 20 | 106.0 | 24.0 | 4.9 | 141.3 | |
| *1933* | | | | | | | | | | | |
| Jan. 8 | 161.3 | 149.6 | 10.6 | 7.15 | 16.9 | 27 | 107.9 | 24.8 | 4.7 | 149.6 | 1st adrenalectomy on Oct. 10 |
| Jan. 14 | 155.5 | | | | | | 105.8 | | 4.6 | 144.7 | |
| Jan. 22 | 133.0 | 121.4 | 12.4 | 6.77 | 16.0 | 64 | 89.8 | 15.6 | 5.6 | 120.3 | 2nd adrenalectomy on Jan. 18 |
| Jan. 23 | 133.0 | 119.8 | 14.2 | 6.80 | 16.0 | 52 | 87.1 | 16.7 | 6.0 | 118.6 | |
| Jan. 24 | 132.9 | 119.8 | 14.1 | 6.90 | 16.3 | 57 | 89.2 | 14.3 | 5.9 | 119.5 | Dog chloroformed on Jan. 24 |

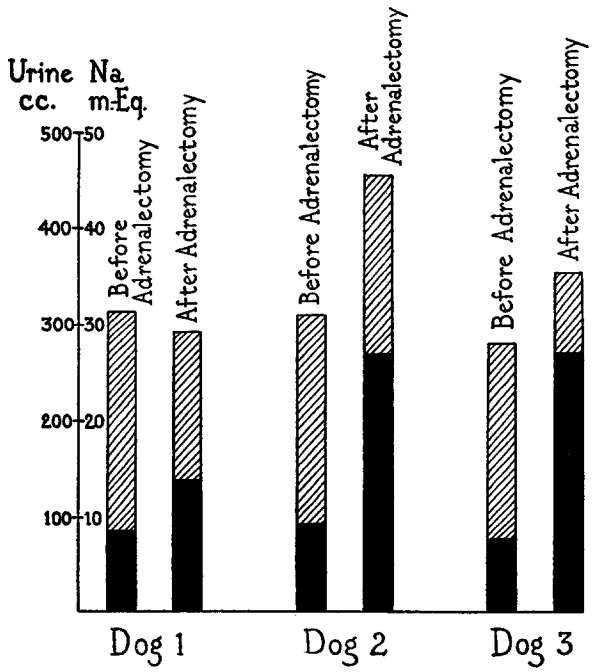


FIG. 4. The effect of adrenalectomy upon the excretion of sodium in relation to urine volume. The total columns represent the daily urine volume in cubic centimeters and the black columns represent the total sodium excretion in milliequivalents.

the metabolic study was begun. *Nitrogen*: In contrast to the other animals, this dog developed a significantly negative balance in the first 4 days after the second operation. *Water*: The urine excretion in this dog was augmented from a daily output of 280 cc. in the fore period to an average of 353 cc. after adrenalectomy. There was, as in both of the other dogs, a marked increase in the Na concentration of the urine in spite of the greater volume. After the 4th day, when symptoms of adrenal insufficiency were marked, the urine volume and Na output decreased. *Blood*: Blood taken on the morning of the 5th day after the second adrenalectomy (Table VI) showed a decrease of about 25 m.-eq. of Na per liter, and was equalled by the drop in chloride and bicarbonate. The K concentration of the blood serum increased slightly and a decided rise was found in the non-protein nitrogen. It is of interest to note that blood samples of 80 cc. each taken on the 6th and 7th days showed no further changes of significance. As was the case in Dog 2, the increase in the undetermined acids of the blood accompanying the loss of sodium was small.

## DISCUSSION

From the data presented, it is apparent that the loss of sodium from the body is an important factor in the development of the clinical picture of adrenal insufficiency in the dog. It is also clear that this loss occurs almost entirely through urinary excretion. That it is not a result of the operative procedure seems certain in view of the fact that it was not demonstrable in Dog 1 after the removal of only one gland. Furthermore, Harrop (9) has found "a distinct rise" in the inorganic base excreted in the urine of an adrenalectomized dog when the administration of cortical extract was stopped. In this animal, the operative wounds had been healed for some time before the extract therapy was withdrawn. Finally, the loss of Na from the body might be anticipated from our studies of the blood of patients suffering from Addison's disease and from the work of Marine and Baumann and that of Zwemer on adrenal insufficiency in cats. Loss of sodium and chloride as a result of vomiting or diarrhea was not a factor in the experiments reported in this paper.

The mechanism of this loss of sodium following total ablation of the adrenal glands is not clear, but at least three possibilities present themselves: (1) that, as in diabetic acidosis, fixed base is called upon to participate in the excretion of large amounts of acid; (2) that the loss of Na is secondary to the loss of water through the kidneys; (3) that the loss of sodium is primary; *i.e.*, that the adrenal glands exert a regulatory effect upon the Na metabolism analogous to that of the

parathyroid glands upon Ca and P metabolism. In other words, one function of the adrenal glands may be to control the concentration of Na in the blood and tissue fluids. The second possibility seems untenable, because it has been shown in this work (Fig. 4) that the relative loss of Na from the body is notably greater than the loss of water after adrenalectomy. Furthermore, with primary dehydration one would expect an increase rather than the established decrease in the concentration of Na in the blood under these circumstances. The commonly known acids produced in the body in amounts sufficient to cause the extreme loss of Na described above are the ketone acids and lactic acid. The ketone acids are not known to be present in adrenal insufficiency and the lactic acid content of the blood of dogs suffering from hypoadrenalism has been shown by Harrop (4) to be unusually low. In the work here presented comparisons of total base and total acids in the blood serum gave no evidence of any significant accumulation of abnormal acid radicals. Sulfate (1) and phosphate ions increase with the retention of non-protein nitrogen, but they can hardly be held responsible for such extreme losses of Na in the urine. Thus it appears that the loss of a primary regulatory mechanism is the most satisfactory hypothesis to explain the behavior of the base sodium in adrenal insufficiency. From the evidence presented it is difficult to ignore the possibility that this assumed influence of the adrenal glands may be exerted upon renal function.

The retention of the K ion in adrenal insufficiency is of interest because this ion in normal subjects and in certain nephritic individuals has been shown (7) to pass through the kidney with greater facility than either the chloride or sodium ions.

It has been pointed out that the retention of non-protein nitrogen occurs in certain disease states in which loss of inorganic base and chloride is marked. Retention of non-protein nitrogen has also been repeatedly observed both clinically and experimentally in adrenal insufficiency, and the work here reported confirms this finding. A current explanation for this apparent kidney insufficiency has been based upon the idea that water available for urea excretion is reduced because of dehydration. Fig. 4, however, indicates clearly that during the period of nitrogen accumulation after adrenalectomy, the rate of water excretion was actually greater than that of the fore period.

Swingle *et al.* (10) have recently stated that the accumulation of non-protein nitrogen is secondary to the fall in blood pressure and the consequent failure of renal filtration. The results of our experiments, as pointed out above, show that this hypothesis is untenable.

## CONCLUSIONS

1. Balance studies have been made on three dogs before and after adrenalectomy, performed in two stages.

2. It has been shown that the sodium concentration of the blood decreases in adrenalectomized dogs, as is true in patients suffering from Addison's disease and in cats experimentally adrenalectomized.

3. There are also decreases in the chloride and bicarbonate concentrations which together are approximately equivalent to the decrease in sodium.

4. An increase in the potassium concentration of the blood occurs after adrenalectomy, as reported in other studies. There is no obvious correlation of this change with changes in potassium balances.

5. The balance studies show a striking loss of sodium from the body during the development of adrenal insufficiency. This loss of Na results from an increased excretion of sodium in the urine and is not complicated by loss of base as a result of vomiting or diarrhea.

6. Following adrenalectomy, both the total amount of sodium and its concentration in the urine are markedly increased. This increase in concentration of sodium occurs in spite of an augmented urine volume.

7. The behavior of the chloride ion following adrenalectomy parallels that of the sodium ion, but the loss is not equivalent.

8. During the period of accumulation of non-protein nitrogen in the blood, the rate of water excretion by the kidney is even greater than before removal of the adrenal glands.

9. The possibility of a regulatory effect of the adrenal glands upon sodium metabolism and renal function has been discussed.

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