

EVIDENCE IN MAN THAT URINARY ELECTROLYTE LOSS INDUCED BY PITRESSIN IS A FUNCTION OF WATER RETENTION¹

By ALEXANDER LEAF,² FREDERIC C. BARTTER,³ ROBERTO F. SANTOS,⁴
AND OLIVER WRONG

(From the Department of Medicine, Massachusetts General Hospital and the Harvard Medical School, Boston, Mass., and the Clinic of General Medicine and Experimental Therapeutics, National Heart Institute, Baltimore, Md.)

(Submitted for publication April 21, 1953; accepted May 15, 1953)

INTRODUCTION

There are many conflicting reports on the acute effects of posterior pituitary extract on the renal excretion of sodium and chloride. This subject has been recently reviewed (1).

The present study was made to determine the effects on electrolyte excretion in man of a long-acting posterior pituitary extract administered over a period of several days. The results suggest that the state of hydration determines the effect of pitressin on electrolyte excretion. The relationship of the findings to the problem of volume regulation of the body fluids will be considered.

METHODS

Metabolic studies were conducted at the Metabolism Ward of the Massachusetts General Hospital (2) and at the U. S. Public Health Service Hospital, Baltimore, Maryland. Subjects studied included four normal adults, two patients with anterior pituitary insufficiency, one with anterior and posterior pituitary insufficiency, one with adrenal cortical insufficiency, and one with ovarian agenesis. Subjects were maintained on a constant diet, and the fluid intake was controlled at various levels. No restrictions were made on the activity or position of the subjects. Dietary sodium intake was fixed in each study and ranged in different studies from 20 to 175 mEq. per day. Following an initial control period of several days, posterior pituitary extract was administered intramuscu-

larly as Pitressin Tannate in Oil,⁵ 2.0 units initially, followed by 1.0 unit every 12 hours for two to four days. This dose of pitressin was estimated to be within the physiological range of endogenous antidiuretic hormone production (3, 4). Twenty separate periods of pitressin administration were studied. In all but one instance an antidiuretic effect was maintained throughout the period of hormone administration.

Urine and serum were analyzed for sodium, chloride, potassium, nitrogen, and total solutes. Two studies included calcium and phosphorus determinations and stool analyses. Analytical methods used have been described in a previous communication (5). Recently, however, the saline standard solutions used in the measurement of total solute concentrations have been corrected for activity coefficients calculated from data obtained in the International Critical Tables (6). Thus, in the studies on R. S., N. W., and O. W. in which the new standards were used, the normal level of total solute concentration of serum was found to be approximately 290 mOsm. per L. rather than the previously reported value of 310 mOsm. per L.

Inulin and para-aminohippurate clearances (7) were used as measures of glomerular filtration rate and renal plasma flow in one subject, M. K. H. In two subjects, R. S. and O. W., 24 hour clearances of endogenous creatinine were used as an estimate of glomerular filtration rate⁶ (8). Acute changes in plasma volume were calculated in subjects R. S. and O. W. from hematocrit and hemoglobin measurements (9).

RESULTS

A. The effects of pitressin on electrolyte excretion

Figure 1 shows a typical response of a normal subject to administration of pitressin over a period

¹ Supported by grants from the Milton Fund of Harvard University, the American Heart Association, and the Worcester Chapter of the Massachusetts Heart Association.

² Howard R. Hughes Fellow in Medicine.

³ Senior Surgeon, United States Public Health Service. Present address: National Heart Institute, National Institutes of Health, United States Public Health Service, Bethesda, Maryland.

⁴ Kellogg Fellow of the American College of Physicians. Present address: Bahia, Brazil.

⁵ Lots L888G or L654C were used. The Pitressin Tannate in Oil was generously supplied by Dr. A. C. Bratton, Jr., Parke, Davis and Co., Detroit 23, Michigan.

⁶ Although not as valid a measure of glomerular filtration rate as is the clearance of inulin, the clearance of endogenous creatinine is a useful estimate of the filtration rate over the daily period of urine collection. This avoids the fallacy of correlating the excretion of water and solutes over twenty-four hours with the clearance of inulin over a few minutes.

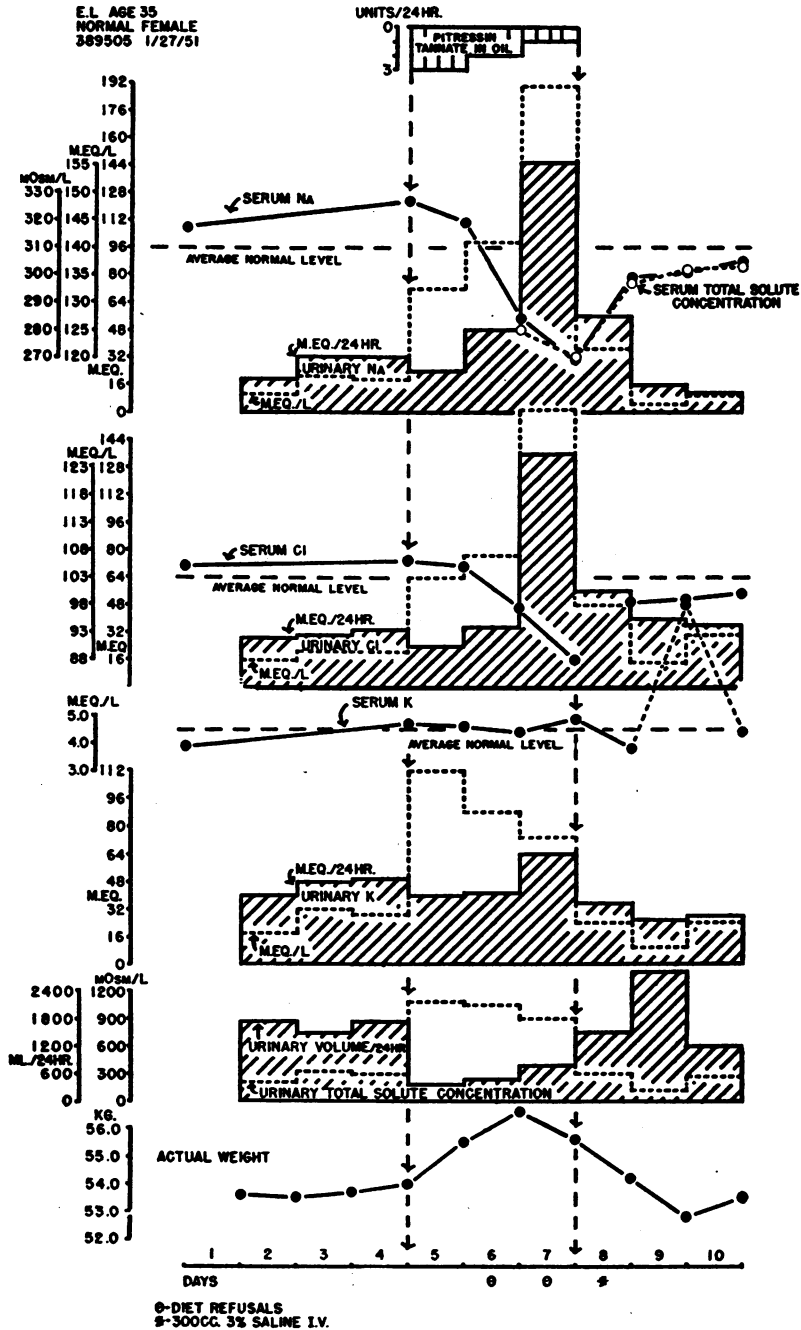


FIG. 1. THE EFFECTS OF PITRESSIN ON SERUM ELECTROLYTE CONCENTRATIONS, URINARY ELECTROLYTE EXCRETION, URINARY VOLUME AND TOTAL SOLUTE CONCENTRATION, AND BODY WEIGHT OF A NORMAL SUBJECT

The hatched columns represent total 24 hour excretions and the broken lines urinary concentrations. Note the prompt antidiuretic effect of pitressin but the delay in the excretion of sodium and chloride. Sodium intake was 60 mEq. per day by analysis. Because of anorexia and nausea, part of the diet and fluid intake was refused on the last two days of pitressin administration. The day after pitressin was stopped 300 ml. of three per cent saline were administered intravenously to correct the extracellular fluid hypotonicity. The abnormally high serum potassium on Day 10 was twice repeated but remains unexplained; no comparable result was seen in any other experiment.

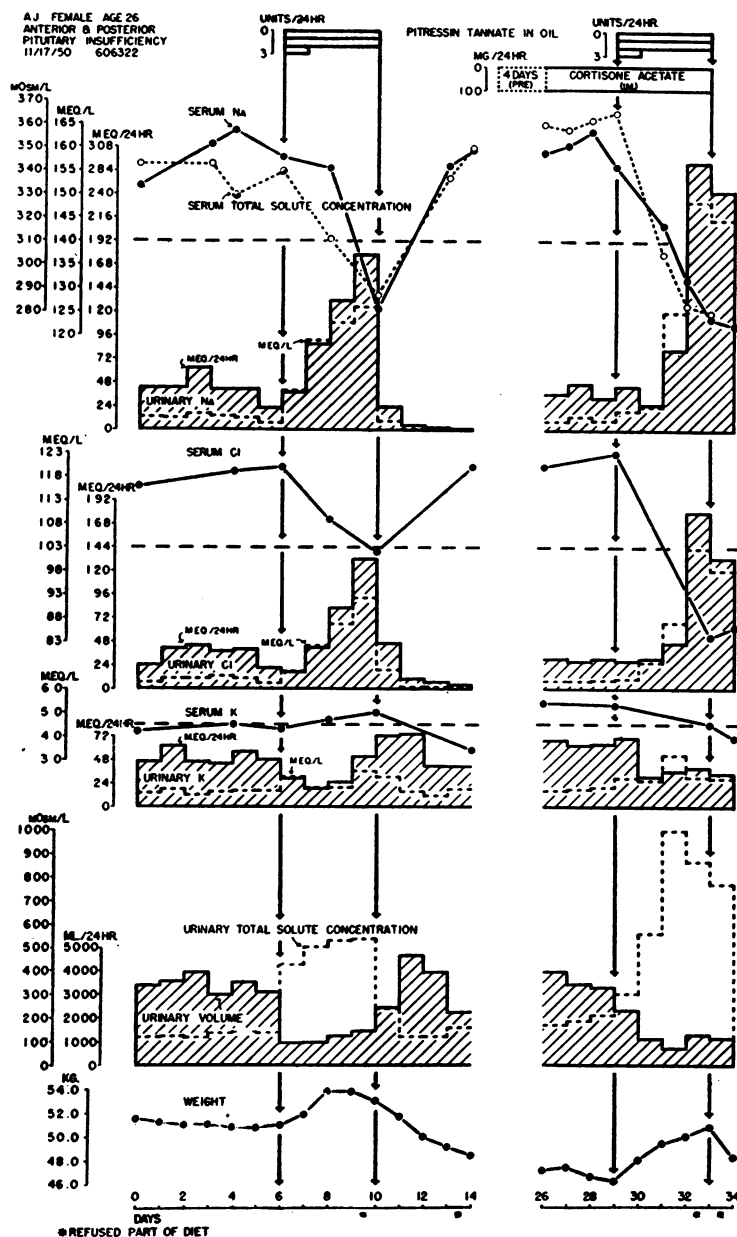


FIG. 2. THE EFFECTS OF PITRESSIN ON SERUM ELECTROLYTE AND TOTAL SOLUTE CONCENTRATIONS, URINARY ELECTROLYTE EXCRETION, URINARY VOLUME AND TOTAL SOLUTE CONCENTRATION, AND BODY WEIGHT IN A SUBJECT WITH ANTERIOR AND POSTERIOR PITUITARY INSUFFICIENCY

The concentrations of serum sodium, chloride and total solute were abnormally high during the control period. In the left-hand section of this figure the effects of pitressin alone are presented. In the right-hand section the effects of pitressin following seven days of continuous cortisone therapy are presented. On cortisone the antidiuretic effect of pitressin was increased, as indicated by the higher urinary total solute concentration. The sodium and chloride loss and symptoms of water intoxication were also increased with cortisone. Sodium intake was 49 mEq. per day by analysis.

of three days. The antidiuretic effect of pitressin is seen in the abrupt decrease in urine volume and increase in urine total solute concentration that occurred on the first day of hormone administration. The water retention produced by this antidiuretic effect resulted in an increase in body weight and dilution of serum sodium, chloride and total solutes. On the third day of pitressin administration a large increase in urinary sodium and chloride excretion occurred. The large water diuresis following the pitressin period represented excretion of water retained during pitressin administration, and returned the weight to the control level. The urinary sodium excretion fell to low levels in the post-pitressin period. These after-effects have been constant sequelae of pitressin administration when the latter has caused fluid retention and sodium loss.

The left-hand section of Figure 2 shows similar data obtained from a patient with anterior and posterior pituitary insufficiency. Again pitressin had a prompt antidiuretic effect. Only after the first 24 hours was there an increased excretion of sodium and chloride, which progressed stepwise throughout the remainder of the pitressin period.

Similar, though less striking, results were obtained in a subject with adrenal cortical insufficiency (Figure 3) treated with 25 mgs. of cortisone acetate orally every eight hours throughout the study.

The delay in the appearance of the increases in electrolyte excretion suggested that they could be dissociated from the prompt antidiuretic action of pitressin. The increased sodium excretion during administration of pitressin was associated with progressive retention of water. This suggested that the effects on electrolyte excretion could result from the overhydration produced by pitressin rather than from a primary action of pitressin.

B. Prevention of the effects of pitressin on electrolyte excretion by dehydration

To test this hypothesis pitressin was given to three normal subjects during periods of low fluid intake which would preclude fluid retention in spite of exogenous antidiuretic activity. The same dose of pitressin was repeated later in the study during periods of high fluid intake. Figures 4

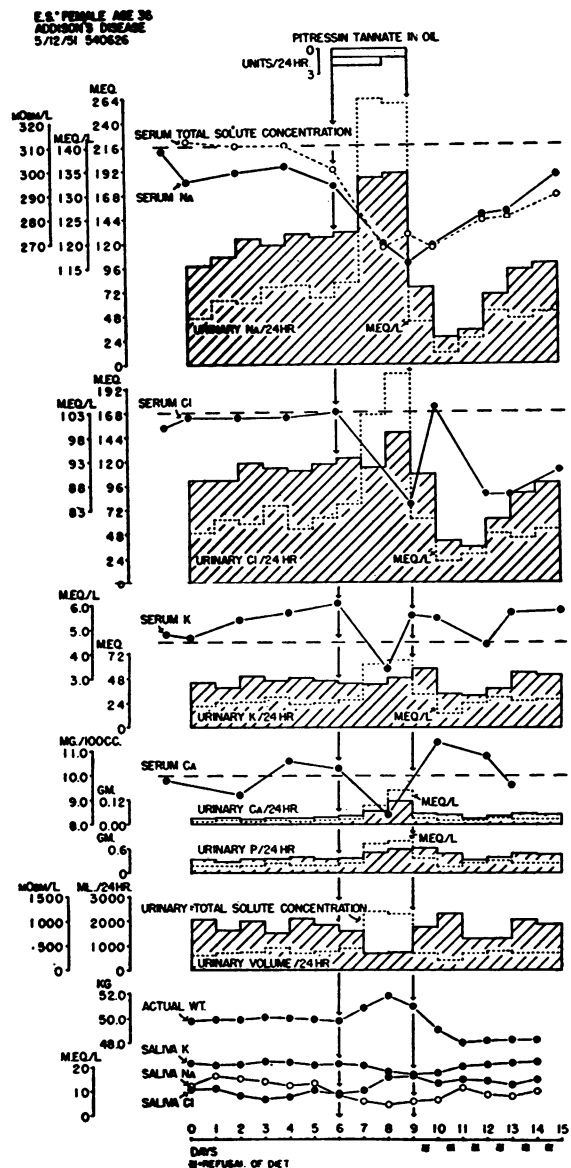


FIG. 3. THE EFFECTS OF PITRESSIN ON SERUM ELECTROLYTE AND TOTAL SOLUTE CONCENTRATIONS, URINARY ELECTROLYTE AND NITROGEN EXCRETION, URINARY VOLUME AND TOTAL SOLUTE CONCENTRATION, AND BODY WEIGHT IN A SUBJECT WITH ADRENAL CORTICAL INSUFFICIENCY

The subject was maintained on 25 mg. of cortisone acetate orally every eight hours throughout the study. Urinary calcium and phosphorus excretion and salivary electrolyte concentrations are also shown. Sodium intake was 124 mEq. per day by analysis.

and 5 show the influence of the state of hydration on renal sodium excretion.

Figure 4 shows the effects of two different levels

of fluid intakes on the excretion of electrolytes during pitressin administration. The urine volume was small, and its total solute concentration high, during the control period of low fluid intake. Administration of pitressin resulted in no further increase in urine total solute concentration and no fluid retention, as indicated by the constancy of

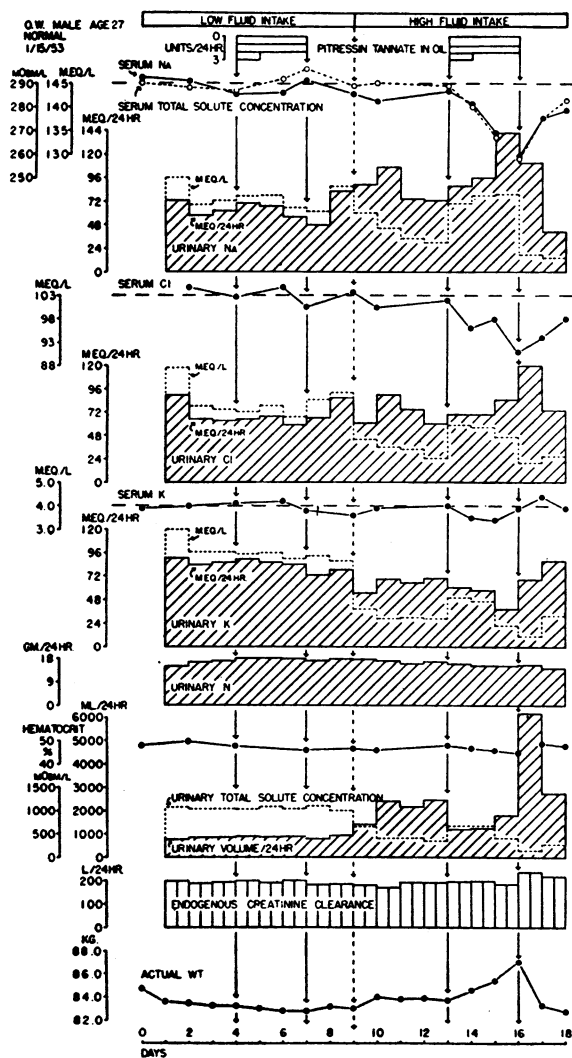


FIG. 4. THE EFFECTS OF PITRESSIN ON SERUM ELECTROLYTE AND TOTAL SOLUTE CONCENTRATIONS, URINARY ELECTROLYTE AND NITROGEN EXCRETION, URINARY VOLUME AND TOTAL SOLUTE CONCENTRATION, HEMATOCRIT, ENDOGENOUS CREATININE CLEARANCE, AND BODY WEIGHT IN A NORMAL MAN ON LOW AND HIGH FLUID INTAKES

Fluid restriction prevented all of the effects of pitressin (weight gain, serum dilution, sodium and chloride loss) observed during the high fluid intake. Sodium intake was 78 mEq. per day by analysis.

body weights and the absence of serum dilution. No change in sodium or chloride excretion resulted from pitressin in the absence of fluid retention.

The fluid intake was then increased. Urine volumes increased and urine concentration dropped. When pitressin was again given, its prompt antidiuretic effect was evident. Water was now retained, as indicated by the weight curve. A definite increase in urinary sodium was present on the third day of pitressin. The increase in chloride occurred the following day. The marked dilution of the serum sodium and total solute concentrations resulted both from water retention and, to a lesser extent, from excretion of sodium.

Figure 5 shows the effect of three different levels of fluid intake on the excretion of electrolytes during pitressin administration. Again with restriction of fluid intake to low levels (920 ml. of water in total fluid intake daily) no significant changes in sodium and chloride excretion were noted. When a further 1,500 ml. of water were given daily, repetition of the pitressin dosage now resulted in definite fluid retention and weight gain, accompanied by a slight increase in sodium and chloride excretion. Finally, the fluid intake was increased to a total of 4,000 ml. daily; pitressin now caused a very large increase in sodium and chloride excretion.⁷ Because of the rapidity of water retention and the marked dilution of serum solute concentration that occurred with the high fluid intake, the last pitressin period was shortened to two days' duration.

These observations suggested that the electrolyte excretion following pitressin is determined by the state of hydration. Figure 6 shows the relationship between per cent increase in body weight and cumulative sodium loss during fifteen periods of pitressin administration. It is apparent that a rough correlation exists between fluid retention and sodium loss.

The possibility existed that the late increase in sodium and chloride excretion might simply be the result of an accumulation of pitressin activity in excess of physiological amounts. The total of

⁷ Although the sodium and chloride excretion had not reached "base-line" control values by the time the third course of pitressin was started, the effects on salt-loss are unequivocal.

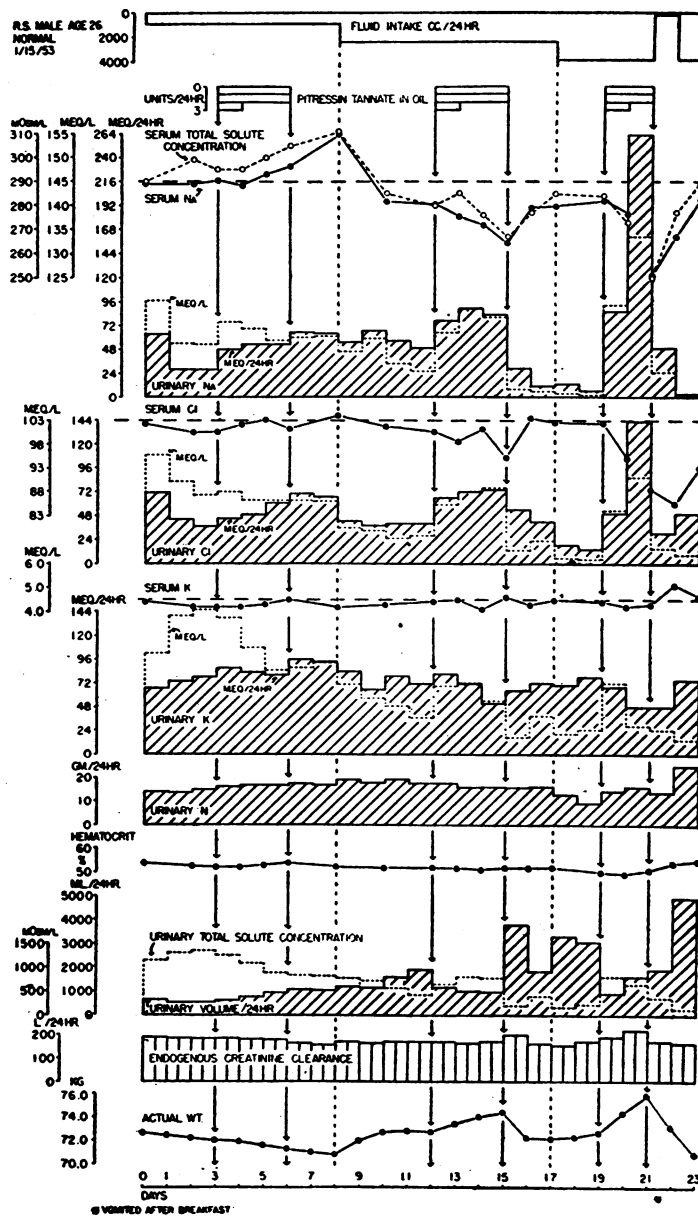


FIG. 5. THE EFFECTS OF PITRESSIN ON SERUM ELECTROLYTE AND TOTAL SOLUTE CONCENTRATIONS, URINARY ELECTROLYTE AND NITROGEN EXCRETION, URINARY VOLUME AND TOTAL SOLUTE CONCENTRATION, HEMATOCRIT, ENDOGENOUS CREATININE CLEARANCE, AND BODY WEIGHT IN A NORMAL MAN DURING THREE DIFFERENT LEVELS OF FLUID INTAKE

During dehydration there was no loss of sodium in excess of intake, which by analysis was 60 mEq. per day. With a moderate and a large increase in water intake, pitressin produced corresponding degrees of fluid retention and sodium and chloride loss. The severity of dehydration on the low fluid intake in this subject was indicated by the high serum sodium and total solute concentrations and the weight loss. The progressive fall in urine total solute concentration and increase in urine volume, in spite of this dehydration and in spite of pitressin is unexplained and was not observed in other studies.

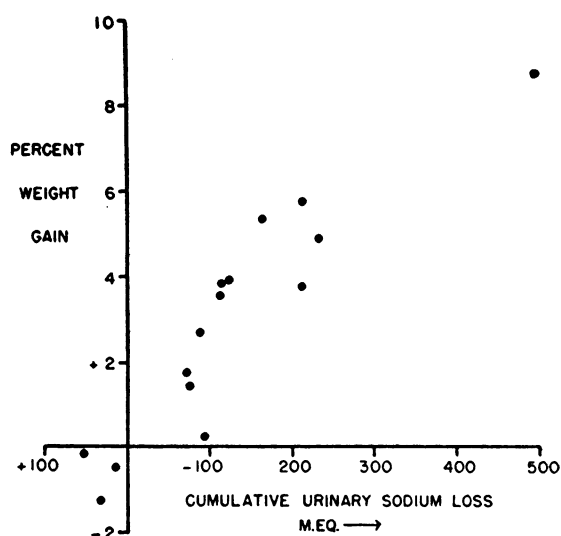


FIG. 6. THE RELATIONSHIP BETWEEN PITRESSIN-INDUCED WATER RETENTION (PER CENT INCREASE IN BODY WEIGHT) AND CUMULATIVE SODIUM LOSS

The data include all the pitressin periods except those during ACTH and very low sodium intake, both of which prevented sodium loss. The cumulative sodium loss was calculated as that in excess of control urinary sodium excretion and per cent weight gain was calculated from the maximum weight increase achieved during pitressin.

7.0 units of pitressin had previously been administered over three days in most of the studies. Therefore, this dose was administered to one subject, O.W., all at one time in five injection sites, in order to produce hormone levels higher than any that could have occurred in the other studies. Antidiuresis was accompanied by a weight gain of only 0.5 per cent (0.4 Kg.), and no increase in sodium or chloride excretion. This indicates that the large sodium and chloride losses from pitressin in these experiments cannot be explained as effects of excessive doses of pitressin.

C. Prevention of the effects of pitressin on electrolyte excretion by ACTH

In two studies adrenocorticotrophic hormone was administered concomitantly with pitressin to see whether the sodium and chloride excretion could be prevented by stimulation of the adrenal cortex. Figure 7 shows the results in one subject; the other subject responded in similar fashion. Pitressin alone produced the expected fluid retention, weight gain, serum dilution and diuresis of

sodium and chloride. A potent corticotropin,⁸ 0.5 mg. administered subcutaneously every six hours for five days, completely abolished the sodium and chloride diuresis during pitressin; in fact, sodium excretion was markedly inhibited until ACTH was stopped.

The maximum increase in body weight was 2.3 kilograms when pitressin alone was administered but 3.1 kilograms when pitressin and ACTH were given. In spite of the larger fluid retention that occurred during the ACTH administration the serum dilution was less because of the concomitant sodium retention.

A very low sodium intake, 20 mEq. per day, during another study on this subject likewise prevented the expected sodium loss during pitressin administration. Such a low sodium intake is a potent stimulus for renal conservation of sodium (10).

D. Hemodynamic and renal changes during pitressin administration

In none of the subjects was there a significant change in blood pressure during administration of pitressin. Expansion of plasma volume subsequent to water retention was shown in subjects O. W. and R. S. The peak increase of plasma volume (as calculated from changes in hematocrit and hemoglobin) was 8 per cent in O. W. and 13 per cent in R. S.

In the three studies in which renal clearances were measured they were found to be increased during the fluid retention. In subject R. S. the clearance of endogenous creatinine rose from a mean control level of 171 ± 11 liters per day to a maximum of 217 liters during the second day of the last pitressin period. In O. W. the clearance of endogenous creatinine rose from a mean control level of 189 ± 9 liters per day to a maximum of 226 liters on the day after the second pitressin period.

In subject M. K. H. the inulin clearance rose from control values of 105, 102, and 85 ml. per minute per 1.73 m^2 to 124 ml. per minute per 1.73 m^2 during the last day of the first pitressin period in Figure 7. The simultaneous clearances of para-aminohippurate showed no significant changes.

⁸ Generously supplied by Dr. E. B. Astwood.

E. The effects of pitressin on excretion of other electrolytes and nitrogen

In the two studies in which calcium and phosphorus balances were measured there occurred an increase in urinary calcium excretion similar to

that shown in Figure 3. Changes in phosphorus excretion were equivocal. No constant pattern of potassium excretion was noted. Urinary nitrogen was not significantly affected by pitressin administration (Figures 4 and 5).

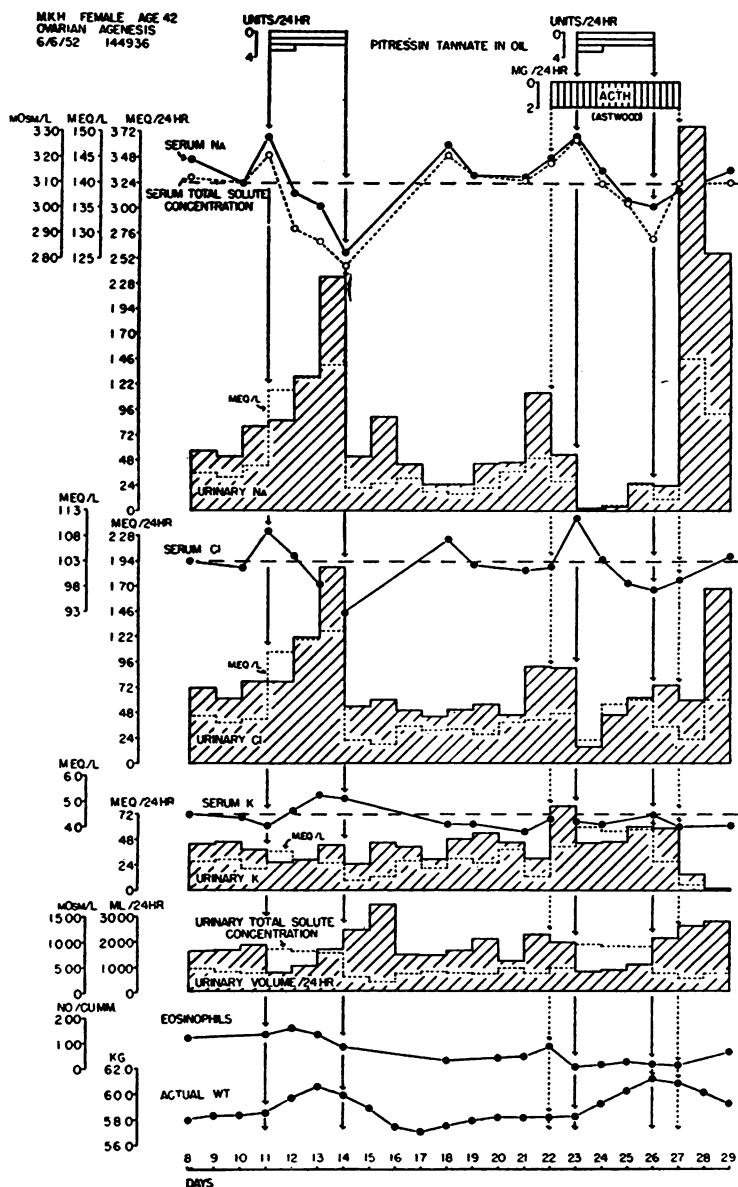


FIG. 7. THE EFFECTS OF PITRESSIN, ALONE AND WITH ACTH, ON SERUM ELECTROLYTE AND TOTAL SOLUTE CONCENTRATIONS, URINARY ELECTROLYTE EXCRETION, URINARY VOLUME AND TOTAL SOLUTE CONCENTRATION, CIRCULATING EOSINOPHILS, AND BODY WEIGHT IN A SUBJECT WITH OVARIAN AGENESIS

The natriuresis and chloruresis induced by pitressin were completely abolished by ACTH. The serum dilution was less with ACTH together with pitressin than with pitressin alone, although the weight gain was greater.

DISCUSSION

The confusion that exists regarding the effects of pitressin on renal excretion of electrolytes is probably the result of a diversity of experimental conditions. Species' differences, variations in dose, and the use of different extracts may have contributed to the conflicting findings. The present study indicates that the state of hydration of the experimental subject may be an important variable. Thus, dehydration prevented the large excretion of sodium and chloride produced by pitressin in hydrated subjects. This finding is in accord with the observation that endogenous antidiuretic hormone, released by dehydration (11-13), does not cause natriuresis or chloruresis (14, 15).

That pitressin does not exert a direct effect on electrolyte excretion in man is suggested by the results of the present study. The increased electrolyte excretion associated with pitressin appeared to be secondary to water retention and was prevented by dehydration.

The possibility, however, of a direct effect of pitressin on renal excretion of sodium and chloride has not been excluded. Mechanisms for sodium retention may come into play during dehydration and obscure a pitressin effect. Increased adrenal cortical activity was capable of inhibiting the natriuresis and chloruresis, and it is possible that dehydration stimulates the adrenal cortex.

Further evidence that pitressin has no direct electrolyte effects was seen in the dissociation between its rapid antidiuretic action and the late increase in electrolyte excretion which occurred in the hydrated subjects. That this late effect was not simply the result of an accumulation of pitressin activity in excess of physiological amounts is indicated by the following: 1) no such effect was seen from the same pitressin dosage during dehydration;⁹ 2) when the total dosage was administered at one time to a hydrated subject there were no electrolyte effects.

The effects of a given quantity of water retention on sodium diuresis could be prevented by

adrenal stimulation or salt restriction.¹⁰ This was demonstrated by studying exactly comparable pitressin periods at the same level of fluid intake, in the same subject, "on" and "off" ACTH, and on low and high salt intakes. Thus it was apparent that this degree of water retention had not served to release endogenous sodium-conserving mechanisms.

In these experiments, small doses of pitressin were given over periods of several days, and it is possible that acute effects were obscured. However, several recent studies in which physiological amounts of posterior pituitary extract were given to human subjects in acute experiments reveal no effect on sodium and chloride excretion (1, 16-20).

Water retention with dilution of extracellular fluid, when marked, was accompanied by symptoms of water intoxication. These were progressive mental confusion, fatigue, headache, and nausea leading to vomiting. Muscle cramps occurred in only one instance. No essential differences were observed in the response of normal subjects and those with endocrine disorders, including adrenal cortical insufficiency. In two subjects, cortisone neither diminished the antidiuretic response to pitressin nor ameliorated the symptoms of water intoxication. Adrenocorticotrophic hormone, on the other hand (Figure 7) did not diminish the antidiuretic response to pitressin, but did ameliorate the symptoms of water intoxication, presumably by causing sodium retention, and hence less serum and extracellular fluid dilution.

Low serum sodium concentrations have been considered to be a cause of decreased renal function. Serum sodium levels as low as 120 mEq. per L. were found in this study in the absence of any evidence of diminished renal function. Inulin, para-aminohippurate, and creatinine clearances, as well as renal concentrating ability, were unimpaired. This would suggest that the cause of reduced renal function in other low sodium states is not the low serum sodium concentration per se, but rather some associated abnormality, such as a

⁹ Theoretically at least, during dehydration, administered pitressin is superimposed upon endogenous antidiuretic hormone, so that the total quantity of circulating hormone is greater than that obtaining when pitressin is administered to the hydrated subject.

¹⁰ Since an increase of glomerular filtration rate was found to accompany water retention, it is likely that water retention of a sufficient magnitude could produce sodium loss in spite of a maximal stimulus to tubular reabsorption of sodium.

reduced plasma volume or cardiac output commonly found in these disorders.

It has been postulated that edema may result from an excess of endogenous antidiuretic hormone activity (21, 22). The assumption is made that primary water retention will cause sodium retention and the resultant isotonic over-expansion of extracellular fluid volume which characterizes the edematous state. The present studies, though of short duration, suggest that this sequence of events does not occur. Pitressin had no effect on the dehydrated subject; when the fluid intake was increased water retention occurred but sodium loss rather than retention ensued. It appears that antidiuretic hormone activity itself, in the absence of simultaneous and independent mechanisms for sodium retention, does not result in edema. No edema was noted in our subjects; indeed it is unlikely that pure water retention could cause significant clinical edema without lethal water intoxication.

Some increase in glomerular filtration apparently occurred during the water retention and natriuresis. However, the demonstration that ACTH prevented such sodium and chloride loss indicates that tubular reabsorptive capacity for sodium and chloride had not been exceeded. The finding of large excretions of sodium and chloride at a time when their concentrations in the serum were falling, clearly indicates that serum concentrations of sodium and chloride were not effective stimuli for their renal conservation. It is thought that this renal loss of sodium and chloride in the presence of obligatory retention of water is a homeostatic response to over-expansion of fluid volume.

It is of interest that Stewart and Rourke (23) produced an increased excretion of sodium by acutely over-loading a normal subject with water to the point of water intoxication. No posterior pituitary extract was administered to their subject. Strauss, Davis, Rosenbaum, and Rossmeisl (24) have increased renal sodium excretion by expansion of extracellular fluid volume with hypotonic saline solutions. These results appear to be manifestations of the same phenomenon: increased sodium excretion in response to over-expansion of body fluid volume.

SUMMARY AND CONCLUSIONS

The effects of a long-acting posterior pituitary extract on electrolyte and water excretion were studied in man. Observations were made during twenty 2 to 4 day periods of pitressin (1 unit per 12 hours) in nine subjects maintained on constant diets and fluid intakes.

It was found that pitressin produced in normally hydrated subjects the following: 1) prompt water retention and resulting weight gain; 2) serum dilution; and 3) a marked increase in urinary sodium and chloride excretion manifest on the second or third day of pitressin administration.

Cessation of pitressin administration was followed by: 1) a large water diuresis with fall in body weight; 2) return of serum concentration to control levels; and 3) a decrease in urinary sodium and chloride excretion to levels below the control.

All the above changes were prevented by restriction of fluid intake during pitressin administration. This strongly suggests that the increased excretion of sodium and chloride was the result of water retention and not a direct effect of pitressin.

Some increase in glomerular filtration rate apparently occurred during the water retention and natriuresis. Evidence that the tubular reabsorptive capacity for sodium and chloride had not been exceeded, however, was indicated by the prevention of such electrolyte loss by ACTH or by a low sodium diet.

The renal loss of sodium and chloride resulting from obligatory retention of water is interpreted as a homeostatic response to over-expansion of fluid volume.

ACKNOWLEDGMENTS

The authors wish to express their gratitude to Dr. Fuller Albright in whose laboratory this study was initiated and to Miss Dorothy B. Richardson for invaluable technical assistance.

REFERENCES

1. White, A. G., Rubin, G., and Leiter, L., Studies in edema. III. The effect of pitressin on the renal excretion of water and electrolytes in patients with and without liver disease. *J. Clin. Invest.*, 1951, 30, 1287.
2. Reifenstein, E. C., Jr., Albright, F., Wells, S. L., The accumulation, interpretation, and presentation

- of data relating to metabolic balances, notably those of calcium, phosphorus, and nitrogen. *J. Clin. Endocrinol.*, 1945, **5**, 367.
3. Shannon, J. A., The control of the renal excretion of water. II. The rate of liberation of the posterior pituitary antidiuretic hormone in the dog. *J. Exper. Med.*, 1942, **76**, 387.
 4. Lauson, H. D., Eder, H. A., Chinard, F. P., Cotzias, G. C., and Greif, R. L., Estimation of the rate of antidiuretic hormone secretion in normal men. *Federation Proc.* 1948, **7**, 69.
 5. Leaf, A., Mamby, A. R., Rasmussen, H., Marasco, J. P., Some hormonal aspects of water excretion in man. *J. Clin. Invest.*, 1952, **31**, 914.
 6. International Critical Tables of Numerical Data, Physics, Chemistry and Technology. Vol. IV, p. 258. McGraw-Hill Book Company, Inc., New York, 1928.
 7. Goldring, W., and Chasis, H., Hypertension and hypertensive disease. The Commonwealth Fund. New York, 1944.
 8. Leaf, A., and Camara, A. A., Renal tubular secretion of potassium in man. *J. Clin. Invest.*, 1949, **28**, 1526.
 9. Elkinton, J. R., Danowski, T. S., Winkler, A., Hemodynamic changes in salt depletion and in dehydration. *J. Clin. Invest.*, 1946, **25**, 120.
 10. Leaf, A., and Couter, W. T., Evidence that renal sodium excretion by normal human subjects is regulated by adrenal cortical activity. *J. Clin. Invest.*, 1949, **28**, 1067.
 11. Gilman, A., and Goodman, L., The secretory response of the posterior pituitary to the need for water conservation. *J. Physiol.*, 1937, **90**, 113.
 12. Ames, R. G., Moore, D. H., and Van Dyke, H. B., The excretion of the posterior pituitary antidiuretic hormone in the urine and its detection in the blood. *Endocrinol.*, 1950, **46**, 215.
 13. Leaf, A., and Mamby, A. R., The normal antidiuretic mechanism in man and dog; its regulation by extracellular fluid tonicity. *J. Clin. Invest.*, 1952, **31**, 54.
 14. Elkinton, J. R., and Taffel, M., Prolonged water deprivation in the dog. *J. Clin. Invest.*, 1942, **21**, 787.
 15. Peters, J. P., Sodium, water and edema. *J. Mt. Sinai Hosp.*, 1950, **17**, 159.
 16. Sinclair-Smith, B. C., Sisson, J., Kattus, A. A., Genecin, A., Monge, C., McKeever, W., and Newman, E. V., The effects of posterior pituitary extract and smoking on water, sodium and chloride excretion in normal subjects and in patients with congestive cardiac failure. *Bull. Johns Hopkins Hosp.*, 1950, **87**, 221.
 17. Chalmers, T. M., Lewis, A. A. G., and Pawan, G. L. S., The effect of posterior pituitary extracts on the renal excretion of sodium and chloride in man. *J. Physiol.*, 1951, **112**, 238-242.
 18. Black, D. A. K., and Thompson, A. E., Day to day changes in sodium and water output with and without posterior pituitary extract. *Clin. Sc.*, 1951, **10**, 511.
 19. Murphy, R. J. F., and Stead, E. A., Jr., Effects of exogenous and endogenous posterior pituitary antidiuretic hormone on water and electrolyte excretion. *J. Clin. Invest.*, 1951, **30**, 1055.
 20. Nelson, W. P., III, and Welt, L. G., The effects of pitressin on the metabolism and excretion of water and electrolytes in normal subjects and patients with cirrhosis and ascites. *J. Clin. Invest.*, 1952, **31**, 392.
 21. Robinson, F. H., Jr., and Farr, L. E., The relation between clinical edema and the excretion of an antidiuretic substance in the urine. *Ann. Int. Med.*, 1940, **14**, 42.
 22. Ralli, E. P., Robson, J. S., Clarke, D., and Hoagland, C. L., Factors influencing ascites in patients with cirrhosis of the liver. *J. Clin. Invest.*, 1945, **24**, 316.
 23. Stewart, J. D., and Rourke, G. M., The effects of large intravenous infusions on body fluid. *J. Clin. Invest.*, 1942, **21**, 197.
 24. Strauss, M. B., Davis, R. K., Rosenbaum, J. D., and Rossmeisl, E. C., Production of increased renal sodium excretion by the hypotonic expansion of extracellular fluid volume in recumbent subjects. *J. Clin. Invest.*, 1952, **31**, 80.