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FOREWORD

This report was prepared in the Department of Physiology, Albany Medical College, Albany, New York by—

G. S. KANTER, Ph.D.

The author expresses appreciation to R. H. Lubinski and D. Shimandle for technical assistance.

ABSTRACT

Hydrogen ion secretion by the kidneys of anesthetized hypothermic dogs was measured to ascertain if the hypothermic kidney helps to compensate for the acidosis which develops during exposure to cold. Artificial respiration was not used. Total hydrogen ion excretion was measured as the sum of total titratable acid and ammonia excreted. Though anaerobic arterial pH, corrected to body temperature, fell significantly from 7.41 during control to 7.27 at 27° C., urinary pH remained at control levels of approximately 6.8. Neither titratable acid nor ammonia excretion increased in response to the hypothermic acidosis as would have occurred during normothermia with a similar low pH. Total titratable acid excretion fell from a control value of 0.011 mEq./min. to 0.005 mEq./min. at 27° C. Ammonia excretion, after a mild increase from a control value of 0.008 mEq./min., fell to 0.006 mEq./min. at 27° C.

This technical documentary report has been reviewed and is approved.

Robert B Payne

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Chief, Operations Division

URINE TITRABLE ACIDITY AND AMMONIA EXCRETION DURING HYPOTHERMIA

1. INTRODUCTION

It was observed in a previous study that the pH of urine collected anaerobically from anesthetized dogs, made progressively more hypothermic, did not decrease concomitantly with the fall in arterial pH (1). Artificial respiration was not used. The absolute urine pH was shown to be partially dependent on the rate of urine flow in the intact animal. With low flows, urine pH remained at approximately control level, while with higher flows, it increased to near blood levels. The failure of the acidotic hypothermic dogs to discharge a more acid urine suggested that the hypothermic kidney is impaired in its ability to acidify the urine. The possibility existed that the failure of the urine pH to decrease noticeably during hypothermia was due to an increased excretion of ammonia and titratable acid. Therefore, this investigation was designed (a) to quantitate the excretion of acid during hypothermia by measuring both titratable acidity and ammonia excretion and (b) to determine if, by excreting additional hydrogen ions, the hypothermic kidney does help compensate for the acidosis which develops. Such a renal compensation has been observed during acidosis in normothermia (2-5).

2. METHODS

The five dogs used in this study were anesthetized with 30 mg./kg. of sodium pentobarbital. To insure patency of the respiratory passages, an endotracheal tube was inserted with the aid of a laryngoscope. Artificial respiration was not used. Rectal temperature was measured with a Yellow Springs Tele-thermometer and a rectal ther-

mistor probe inserted to a depth of about 10 cm. To obtain timed, quantitative urine collections and to avoid possible electrolyte exchanges in the bladder (6, 7), both ureters were cannulated through an abdominal incision close to the bladder, and polyethylene catheters (PE 160) were tied in place. Urine was collected under paraffin oil. Heparinized samples of arterial blood were obtained from an implanted stainless steel femoral cannula. Arterial pH was measured anaerobically at the end of every period with a Beckman model G pH meter and Beckman model 290-31 anaerobic blood electrode assembly immersed in a constant temperature bath set at 38° C. Urine pH was similarly measured anaerobically at the end of each collection period. Corrections were made by using Rosenthal's factor of 0.0147 pH/° C. (8) to allow expression of blood pH at the particular hypothermic body temperature at which the sample was taken. Forty-five minutes before the start of the control collection period a constant infusion of 0.9% saline at 2 ml./min. was begun. The infusion was maintained throughout the remainder of the run to maintain a urine flow adequate for the analyses. After a control period of 30 minutes, the animals were covered with crushed ice to induce hypothermia; thereafter, measurements were made at consecutive 30-minute periods for 240 minutes. The methods of measurement and analysis of data were the same as those reported previously (1, 9).

The total titratable acidity of the urine was measured by potentiometric back-titration to the corrected blood pH existing at time of sampling with a Beckman model G pH meter and Beckman shielded external electrodes. The titrant, 0.1 normal NaOH, was delivered with a micrometric syringe buret into the urine

TABLE I
Renal hydrogen ion excretion during hypothermia

	Control	Hypothermia				
	Rectal temperature					
	33°	35°	33°	31°	29°	27°
Anaerobic arterial pH (correlated to body temperature)	7.41 ±.02	7.42 ±.02	7.39 ±.03	7.37 ±.02	7.34 ±.03	7.27 ±.03
Anaerobic urinary pH (at 38° C.)	6.86 ±.29	6.73 ±.19	6.72 ±.19	6.76 ±.20	6.80 ±.18	6.85 ±.21
Urine flow (ml./min.)	.21 ±.07	.22 ±.05	.26 ±.09	.36 ±.11	.40 ±.12	.47 ±.15
Total titratable acid (mEq./min.)	.011 ±.003	.012 ±.002	.011 ±.002	.008 ±.002	.006 ±.002	.005 ±.002
Plasma phosphate (mM./liter)	1.55 ±.10	1.54 ±.14	1.52 ±.15	1.46 ±.15	1.38 ±.14	1.28 ±.11
Urine phosphate (mM./liter)	109.4 ±47.8	90.4 ±33.3	81.8 ±25.1	71.6 ±21.7	62.0 ±19.0	57.8 ±21.7
Excreted phosphate (mM./min.)	.019 ±.006	.016 ±.003	.017 ±.003	.019 ±.004	.018 ±.002	.015 ±.002
Titratable acid carried by phosphate (mEq./min.)	.008 ±.003	.007 ±.002	.006 ±.002	.005 ±.002	.005 ±.002	.004 ±.002
Excreted ammonia (mEq./min.)	.008 ±.002	.011 ±.001	.011 ±.002	.009 ±.002	.007 ±.001	.006 ±.001
Heart rate/min.	135 ±15	130 ±11	116 ±9	100 ±10	81 ±9	67 ±5
Hematocrit (% RBC)	44.3 ±2.4	44.8 ±2.8	45.4 ±2.9	47.4 ±3.4	49.2 ±3.2	50.8 ±3.2

Mean and standard errors of the means for five anesthetized male mongrel dogs; average weight = 17.0 ± 1.0 kg.

sample. Theoretically, titration of urine at ambient room temperature to the blood pH value which existed at various levels of hypothermia introduced a small error. The magnitude of this error was too small to alter the results significantly. Ammonia excretion was measured by a modification of the standard Conway microdiffusion technic (10). Disposable polyethylene Conway dishes were used. In addition, plasma and urine levels of phosphate were determined by the Gomori method (11). The latter measurements, in conjunction with the measurement of urine and blood pH, allowed calculation of the titratable acid carried by the urinary phosphate.

3. RESULTS

Anaerobic arterial pH, corrected to body temperature, fell significantly, as had been observed previously, from 7.41 during the control period to 7.27 at 27° C. (table I; fig. 1). Urinary pH, however, remained at approximately the control level of 6.8. It is interesting to note that the urinary pH did not decrease markedly as it would have with a similar decrease in blood pH during normothermia. Similarly, titratable acidity did not increase in response to the hypothermic acidosis. Excretion of total titratable acid remained nearly constant, at approximately 0.011 mEq./min.,

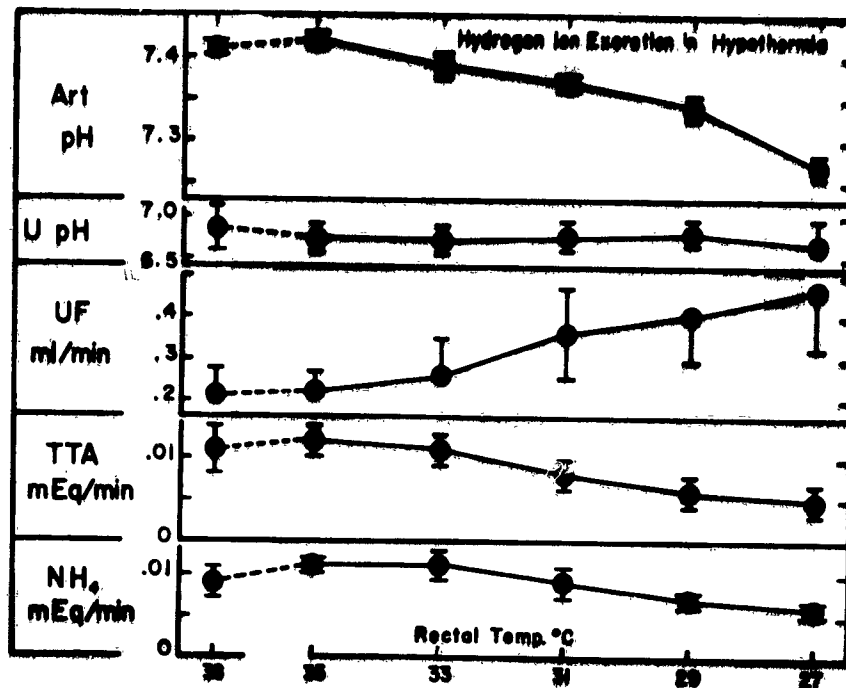


FIGURE 1

Arterial pH, urine pH, urine flow in milliliters per minute total titratable acid excreted in milliequivalents per minute and ammonia excreted in milliequivalents per minute for five dogs cooled to 27° C. Means and standard errors of the means are included.

until the rectal temperature reached 33° C. and thereafter declined. At 29° C. it was half that of control, and at 27° C. it was 0.005 mEq./min.

Although plasma phosphate fell from a control value of 1.55 mM./liter to 1.28 mM./liter at 27° C., phosphate excretion was maintained at the control level of approximately 0.019 mM./min. down to 29° C. Thereafter, the excretion rate decreased to 0.015 mM./min. at 27° C. This relative maintenance of phosphate excretion occurred despite a 17% fall in plasma phosphate at 27° C. and a fall of over 50% in glomerular filtration rate, as shown previously (1, 9). Hypothermic depression of tubular reabsorption, which offset the decrease in the filtered load of phosphate delivered to the tubules, was presumably responsible. Calculation showed that the urinary excretion of phosphate carried from 55 to 85% of the total

titratable acid excreted. The control value was 73% and at 27° C., 80%.

Urine flow increased from 0.21 ml./min. during the control period to 0.47 ml./min. at 27° C. but did not approach the rate of saline infusion of 2.0 ml./min. The decrease in urine phosphate concentration from 109.4 mM./liter in the control period to 57.8 mM./liter at 27° C. was largely a reflection of the increase in urine flow. At lower core temperatures, depression of reabsorption allowed maintenance of a urinary concentration above that which would have been obtained otherwise. The U/P ratio for phosphate decreased from the control level of 70.6 to 45.2 at 27° C.

Similarly, ammonia excretion did not contribute additional hydrogen ion, over control, to the urine. After a mild increase from the

control level of 0.008 mEq./min. to 0.011 mEq./min. at 35° C., ammonia excretion remained constant down to 33° C. and then declined linearly to 0.006 mEq./min. at 27° C. This latter value was 25% below the control and 45% below the peak excretion obtained at 33° C.

The heart rate showed the typical hypothermic depression. Owing to the magnitude of the saline infusion, the increase in hematocrit was not as great as is usually found in deep hypothermia.

4. DISCUSSION

It has been shown that induction of hypothermia without artificial respiration results in a fall in the pH of arterial blood (12, 13). The basis of this hypothermic acidosis is not yet clearly defined. In a recent study, it was shown that bicarbonate excretion does not contribute to this fall in arterial pH in hypothermia (1). Indeed, in spite of renal tubular depression, bicarbonate, in contrast to other substances, is remarkably well reabsorbed (14). It was also found that anaerobic urine pH did not fall as the arterial pH declined (1). The hypothermic kidney was apparently unable to secrete hydrogen ions.

In an earlier study on renal function in hypothermic dogs Segar et al. (15) found an increase in urinary pH from 6.76 to 7.28 and a fall in titratable acid and ammonia excretion. They employed sufficient artificial respiration, however, to maintain blood pH at control levels. As an increase in blood pH from 7.43 to 7.52 occurred, the decrease in acid excretion and the increase in urinary pH might be ascribed to overventilation. What would occur during hypothermic acidosis could not be derived from their data. Hernandez and Coulson (16) in their study on the effect of hypothermia on renal function in alligators found a marked decrease in NH_3 excretion after 24 hours of exposure to 6° C., while urine pH remained at the control level of approximately 7.8. Whether or not an acidosis occurred during cooling is uncertain, as at 6° C. the blood pH was reported

as 7.46. The differences in acid-base regulation, renal conservation of cation, and normal urinary excretion pattern between alligators and most mammals make it difficult to transpose their results to the latter group (17).

The purpose of this investigation was to study the excretion of ammonia and titratable acid during hypothermia in dogs in relation to the alteration in blood and urine pH when artificial respiration was not employed. It was found that the excretion of hydrogen ions by the hypothermic kidney decreased despite the marked decrease in blood pH (table I; fig. 1). The total titratable acid fell 50% while the ammonia excretion declined 25%. Under normothermic conditions and comparable low-blood pH, both total titratable acid and ammonia excretion increase significantly (2-5). That they do not do so at a low-core temperature indicates that the renal acidification mechanism is quite temperature-dependent.

Calculation of the role of the urinary phosphate system during hypothermia shows that, similar to the finding during normothermia, phosphate is the major buffer salt of the urine. While both the total titratable acid excretion and titratable acid carried by urinary phosphate fell approximately to half during hypothermia, the bulk of the titratable acid excreted was carried by the phosphate system.

One determinant of the ability of the kidney to acidify the urine is the ability of the distal tubular cells to take up sodium ion in exchange for hydrogen ion. An important component of this mechanism involves an exchange between hydrogen and potassium ions (18). It is therefore noteworthy that previous studies have demonstrated that the control rate of excretion of potassium is maintained in the hypothermic kidney even though the potassium load presented to the tubules falls to levels 60% below control, because of the fall in glomerular filtration rate and plasma concentration (9). The suggestion has been made that, instead of an exchange of hydrogen ions for sodium ions, the hypothermic kidney may actually excrete potassium ions in place of

hydrogen ions (1). The plausibility of this explanation is strengthened if the excretion data in relation to the changes in plasma potassium in the hypothermia experiments (9) are recalculated to assess total potassium balance. It may be demonstrated that the renal loss of potassium cannot be accounted for by the fall in extracellular concentration of potassium. There must be a loss of potassium from some or all of the body cells.

The change in plasma potassium concentration during hypothermic acidosis contrasts significantly with the influence of acidosis in

normothermia, where plasma potassium concentration increases as plasma pH falls (19, 20). The present study shows that the hypothermic kidney fails to increase the excretion of hydrogen ions in acidosis, and a previous study showed that a relative increase in potassium excretion occurred at the expense of some intracellular potassium (9). These findings give added support to the hypothesis that a failure of normal cation exchange mechanisms in the hypothermic kidney is responsible for both the depressed plasma potassium concentration and the failure of the kidney to compensate for the acidosis of hypothermia.

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