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Dilutional Hyponatremia Due to Diazoxide-Produced Polydipsia[1]

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TANG, M. AND J. L. FALK. *Dilutional hyponatremia due to diazoxide-produced polydipsia.* PHARMAC. BIOCHEM. BEHAV. 3(1) 115–119, 1975. – Subcutaneous injection of diazoxide every 3 hr for a total of 5 doses in 15 hr produced a state of elevated drinking and antidiuresis in rats resulting in a massive, positive, self-imposed water load. Dilutional hyponatremia was present, but not serum hyposmolality, owing to the increased serum glucose and BUN. The mechanism by which diazoxide produces a polydipsia even in the presence of an accumulating water load may illuminate the genesis of other pathophysiological dilutional states.

Diazoxide Dilutional hyponatremia Polydipsia Water intake

IN a previous study [10] it was determined that diazoxide (3-methyl-7-chloro-1,2,4-benzothiadiazine-1, 1-dioxide), a direct-acting hypotensive agent, produced a substantial, dose-related increase in water intake in water-satiated rats. Subcutaneous administration of 80 mg/kg diazoxide yielded about a 5.5 ml/100 g water intake in the 3–4 hr post-injection period. Five injections of 40 mg/kg, each separated by 3 hr, gave rise to a sustained polydipsia over the 15-hr period, accompanied by antidiuresis and frank edema.

The present experiments were designed to explore the phenomenon of diazoxide-produced polydipsia under additional conditions, but especially to evaluate the self-imposed alteration in body water status.

METHOD

Animals

Twenty-eight male, Holtzman rats with a mean weight of 393.6 g (range: 328–442 g) were used. They were divided at random into 5 groups as described below. Animals were housed individually in metabolism cages (Acme Research Products) under temperature-controlled, light–dark cycle conditions (12 hr on, 12 hr off).

Drug

Diazoxide, a generous supply of which was obtained from Schering Corp., Summit, N. J., was administered subcutaneously (SC). For injections of 40 mg/kg, the vehicle was 1:7 of 1 N NaOH: isotonic saline; for 80 mg/kg the vehicle was adjusted to a 1:3 proportion.

Procedure

Animals were adapted for 4 days to their metabolism cage environment. Purina Laboratory Chow (pelleted) was available continuously, except during the injection-series procedure, as noted below. Water was continuously available from calibrated animal drinking reservoirs (Richter tube). On the fifth day, animals were injected once every 3 hr for a total of 5 injections and their water intakes and urine outputs were measured hourly for 15 hr, starting 1 hr after the first injection.

During the cage adaptation days, animals were weighed and their food and water serviced at 1:00 p.m. On the fifth day (injection-series day), animals were weighed as usual, injected SC, and food was removed from the cages. There were five groups of animals constituted as to the dose of diazoxide per injection, 80 mg/kg (N = 5), 40 mg/kg (N = 8), and the respective vehicles for the two diazoxide dose levels, designated VH 80 (N = 5) and VH 40 (N = 5), and a group (N = 5) receiving 80 mg/kg/injection of diazoxide but not allowed water to drink during the 15 hr observation period (DZ 80, no H_2O).

At the end of the fifteenth hr, animals were decapitated and blood was collected. Samples were allowed to stand for 20 min and then centrifuged at 3,300 rpm for 30 min. The serums were then frozen for later analysis. Serum sodium values were determined with an Instrumentation Laboratories 143 internal standard flame photometer, chlorides by a Marius-Fiske chloridimeter, and osmolalities by a Fiske G-33 osmometer using small-sample (0.2 ml) tubes. Glucose values were determined by the O-toluidine reaction method

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[14] modified for microsamples. Blood urea nitrogen (BUN) values were determined by the diacetyl monoxime method [5]. A Spectronic 70 spectrophotometer (Bausch and Lomb) was used in the glucose and BUN determinations. Hematocrits were determined by heparinized microhematocrit tubes and a Clay-Adams microhematocrit centrifuge. Whenever possible double determinations were obtained. Instrument calibration was checked with Versatol and Versatol A (General Diagnostics, Warner-Lambert Pharmaceutical Co., Morris Plains, N. J.). The osmometer was calibrated and checked with vials of Fiske certified standards.

RESULTS

Figure 1 shows that repeated diazoxide administration at both 40 and 80 mg/kg/injection resulted in severe and sustained polydipsia relative to the vehicle control groups. The urine output volumes of the diazoxide-injected groups allowed to drink water were only slightly greater than volumes of the diazoxide group not given water and the vehicle control groups. An analysis of variance among all these groups for final, cumulative urine volume failed to yield a significant F-value. Considering the large, self-administered, positive water load, the small urine volume demonstrates the powerful antidiuretic action of diazoxide. The cumulative mean amount of water drunk at 15 hr by the 80 mg/kg/injection group (79 ml ± 7.94 S.E.) is significantly greater than that drunk by the 40 mg/kg/injection group (t = 2.847, df = 10, $p<0.05$) and by its respective vehicle control group t = 10.452, df = 7, $p<0.001$. Estimating the final cumulative, positive water loads by adding the percent increase in body weights of the diazoxide injected groups to the percent weight reductions of their respective vehicle control groups reveals that the DZ 80 group had a 13 percent body weight load while the DZ 40 group had an 8 percent load. Animals injected with 80 mg/kg diazoxide but not given water sustained a 2.6 percent decrease in body weight during the 15 hr experimental period which is not significantly different from that lost by the VH 80 group.

Figure 2 shows the serum sodium values of the diazoxide groups which were allowed to drink water. Owing to the antidiuresis coupled with severe polydipsia a self-imposed dilutational hyponatremia developed. The hyponatremia was not due to any direct, pharmacological effect of diazoxide on water–electrolyte distribution among intracellular and extracellular phases since the serum sodiums of the 80 mg/kg/injection diazoxide group not allowed water (DZ 80, no H_2O) were not statistically different from the two vehicle control groups (VH 40 and VH 80). The serum chloride values reflect the same body water states inferred

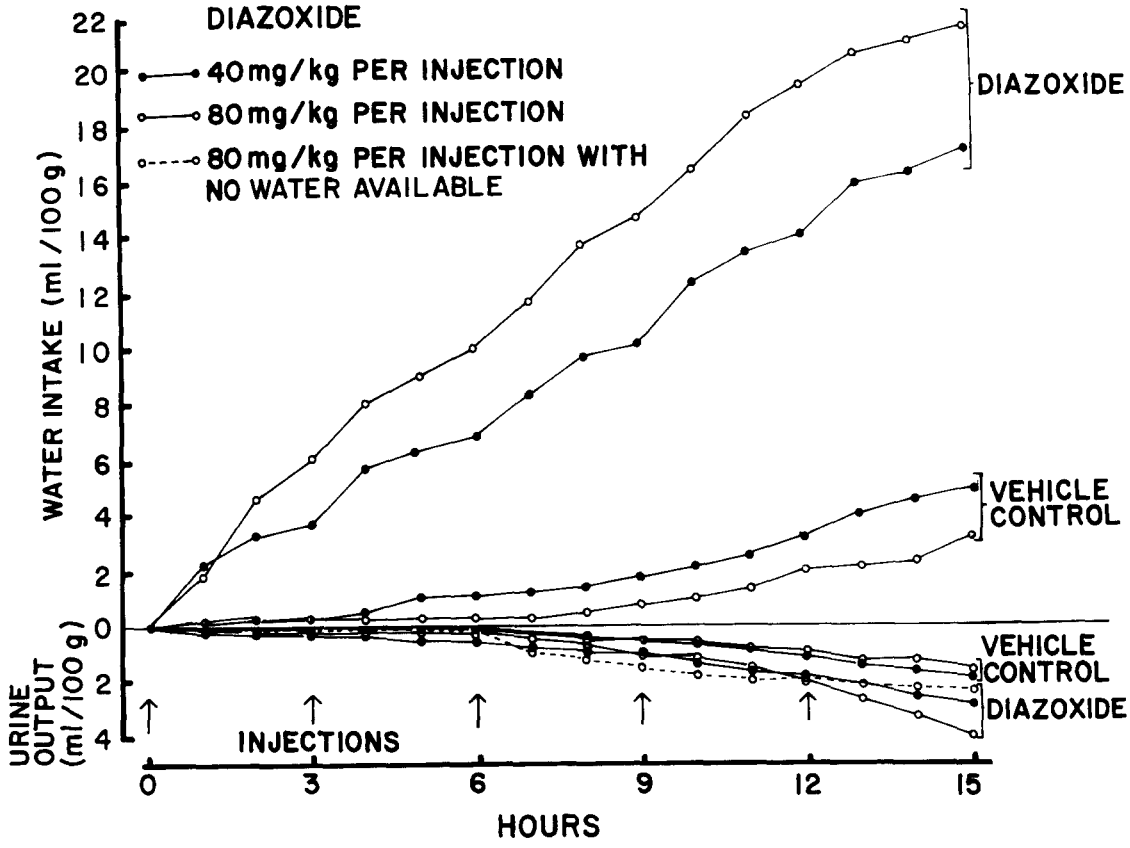

FIG. 1. 15-hr cumulative mean water intakes (percent body weight) and/or urine outputs of water-satiated rats given repeated SC injections of diazoxide, vehicle, or diazoxide with no water available.

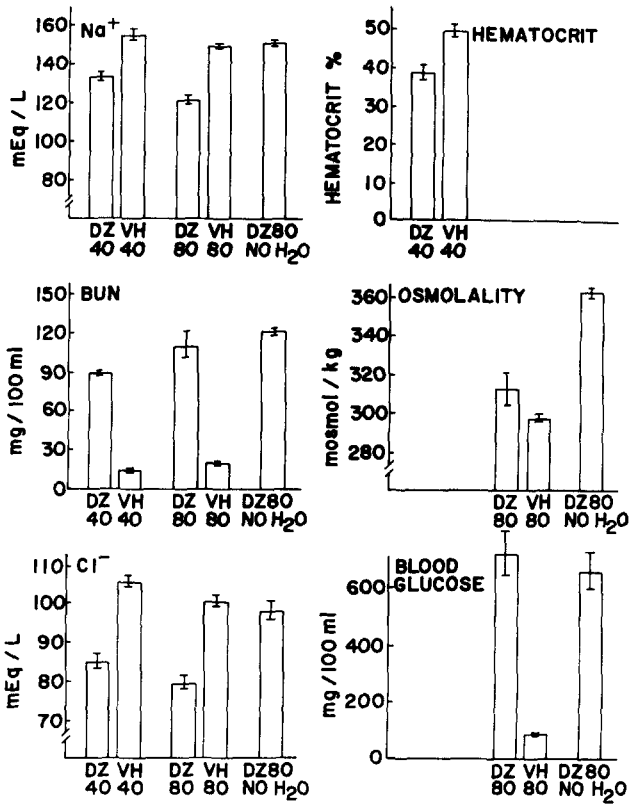

FIG. 2. Mean serum and blood values (± S.E.) for groups shown in Fig. 1 at the 15 hr. DZ 40 and DZ 80 = repeated doses of 40 or 80 mg/kg diazoxide, SC; VH 40 and VH 80 = equivalent doses of vehicle; DZ 80, no H_2O = repeated doses of 80 mg/kg diazoxide with no water available during the 15 hr period.

TABLE 1

SERUM OSMOLALITIES OBSERVED 15 HR AFTER FIVE SPACED DOSES OF DIAZOXIDE (80 mg/kg, SC; DZ 80), AFTER SC VEHICLE INJECTIONS (VH 80) AND AFTER DIAZOXIDE DOSES WHEN NO DRINKING WATER WAS ALLOWED DURING THE 15 HR PERIOD (DZ 80, no H_2O). SECOND COLUMN SHOWS SERUM OSMOLALITIES AS CALCULATED FROM MEASURED SERUM SODIUM, GLUCOSE AND BUN VALUES.

Group	Serum mosml/kg Observed	Serum mosml/kg Calculated
VH 80	298	294
DZ 80	314	310
DZ 80, no H_2O	364	364

from the serum sodium levels. Again, the hematocrit difference between the DZ 40 and VH 40 groups reveal the dilutional state of the polydipsia animals.

The dilutional hyponatremia was not accompanied by serum hyposmolality. The osmolality difference between DZ 80 and VH 80 is not statistically significant ($t = 2.244$, $df = 7$, $p > 0.05$). This apparent discrepancy between serum hyponatremia and absence of hyposmolality can be accounted for completely by the concomitant changes in serum glucose and BUN. Table 1 shows the observed osmolalities and those calculated by summing the osmotic contributions based on the measured concentrations of sodium, glucose and BUN [22]:

$$\text{Calc mosml/kg} = 1.86\ (\text{observed } Na^+) + \frac{\text{glucose}}{18} + \frac{\text{BUN}}{2.8}$$

The hyperglycemia and azotemia resulting from diazoxide injection counteracts the hyponatremia, leaving the DZ 80 group osmolality not different from that of the VH 80 group. The elevated osmolality of the DZ 80, no H_2O group is completely accounted for by the above equation (cf. Table 1 and Fig. 2).

The DZ 40 and DZ 80 groups developed an obvious edema by the end of 15 hr. One animal in the DZ 80 group died following the second injection from what appeared as a beta-adrenergic-stimulated cardiac fibrilation.

DISCUSSION

The remarkable efficacy of diazoxide in maintaining continued water intake in spite of an accumluating, positive water load suggests that such an override mechanism could operate in the maintenance of other pathophysiological dilutional states. The mechanism of action which stimulates fluid intake to an extent that it conspires with antidiuresis to generate and maintain dilutional hyponatremia appears to involve the release of some renal factor. This was inferred from a previous study which showed that nephrectomized animals failed to drink after diazoxide injection [12], although nephrectomized animals are responsive to other dipsogenic stimuli [12,18]. The renal dipsogenic factor is released by beta-adrenergic stimulation [12, 18, 20, 23, 26] and some [15,24] but not all [9,31], evidence points to renal renin as the factor.

Since diazoxide is an antidiuretic agent and increases BUN, as well as producing hyperglycemia [29,30], it might be inferred that the resulting increase in serum osmolality (see Fig. 2, DZ 80, no H_2O) could account for the increase in water intake observed in the present experiment. Several lines of evidence militate against this explanation. The hyperglycemia produced by diazoxide is ameliorated by adrenalectomy in mice [30] and rats [27], but adrenalectomized rats respond with a normal dipsogenic response to diazoxide [12]. Propranolol does not reduce diazoxide-induced hyperglycemia [19], but it did antagonize the drinking response [12]. In nephrectomized mice, there is no attenuation in the hyperglycemic response in diazoxide [13,29] and the BUN level would be at least as elevated as in normal animals. Therefore, the serum osmolality elevation to diazoxide would be similar or greater in nephrectomized animals, but the dipsogenic response was absent [12]. The acute response of serum and blood parameters to an 80 mg/kg diazoxide injection revealed no changes in sodium, potassium, hematocrit, osmolality, etc.

after 30 or 90 min postinjection when no drinking water was allowed [12]. Yet, within this time period animals respond to diazoxide with a marked drinking response ([10,12], see also Fig. 1). Considering the above lines of evidence, it is unlikely that the glycemic, azotemic, or resulting osmolal response to diazoxide can explain the marked dipsogenic property of this agent. Sodium retention resulting in plasma volume expansion and peripheral edema sometimes observed with long-term therapeutic use of diazoxide [2,32] would not be a factor operative in these acute experiments.

It is curious that while diazoxide-produced polydipsia, hyperglycemia and BUN elevation result in dilutional hyponatremia, the BUN and glucose values were not markedly different between animals drinking water and those not allowed water during the 15 hr of diazoxide treatment (cf. Fig. 2). We can only speculate that a condition analogous to the carbohydrate disturbance of uremics called azotemic pseudodiabetes may operate to maintain the glucose elevation [6, 28, 33] in addition to the elevation produced by diazoxide, and that an exaggerated production of urea occurred specifically as a function of the dilutional hyponatremic state ([16], p. 434, [21]). The complete accountability for the observed serum osmolalities in terms of the measured sodium, glucose and BUN levels stands in contrast to the situation in which the hyponatremia is produced by sodium depletion, rather than by dilutional processes. In absolute sodium depletion an unidentified serum component increases and contributes to the development of a severe hyperosmolality [11].

There are a number of physiological and behaviorally-induced situations in which water intake is not inhibited by the development of hyponatremia. "Inappropriate" water intake maintains and aggravates the hyponatremia produced by sodium depletion [3, 7, 25] and of dilutional hyponatremia originating from a variety of etiologies as well [17,34]. Nor is the psychogenic polydipsia of humans [1] or animals [8] terminated by the resulting hyponatremia. Both nephrectomized and ureterally ligated rats allowed access to water drink volumes which produce hyponatremia [4]. Thus, the regulation of serum sodium concentration may be overriden by various pathophysiological and pathobehavioral states. In the present experiments, diazoxide-produced polydipsia and antidiuresis with the resulting hyponatremia was not attributable to serum osmolality or volume changes. Neither the stimulus to drink induced by diazoxide, nor the factors permitting maintenance of drinking after the development of severe hyponatremia are known.

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