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BRIEF COMMUNICATION

Drinking in Water Deprived Rats after Combined Central Angiotensin Receptor and Converting Enzyme Blockade[1]

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SEVERS, W. B. AND P. A. KLASE. *Drinking in water deprived rats after combined central angiotensin receptor and converting enzyme blockade.* PHARMAC. BIOCHEM. BEHAV. 9(2) 259–260, 1978.—Saralasin, an angiotensin II receptor antagonist, is ineffective in reducing drinking after water-deprivation (WD). Angiotensin III has considerable dipsogenic potency. It may be formed without angiotensin II as an intermediate. To clarify whether angiotensin III formation masks a role for angiotensin in WD, 48 hr WD rats were infused via a lateral cerebroventricle with a) CSF, b) Saralasin, c) SQ 20,881, a converting enzyme inhibitor, or d) Saralasin + SQ. No drug effect on drinking was observed. The results demonstrate that angiotensin III is not required for WD drinking.

Angiotensin-CNS Angiotensin III Saralasin Converting enzyme

ANGIOTENSIN II, the dipsogenic product of the renin-angiotensin system [2], has been extensively studied to determine if it has a role in endogenously activated thirst. Blockade of angiotensin receptors with saralasin (or similar drugs) failed to reduce drinking behavior after water deprivation although the paradigms used indicate that exogenous angiotensin II drinking would be blocked (review [6]). The most positive result obtained with saralasin was that of Malvin *et al.* [3], who delayed drinking in water deprived rats for 30 min.

Angiotensin III (des-asp[1] angiotensin II) retains about 50% of the dipsogenic potency of angiotensin II [2]. The possibility has been raised that angiotensin III is formed without angiotensin II as an intermediate [1,5]. In this scheme, angiotensin I is de-aspartylated and converting enzyme generates angiotensin III from the des-asp[1] angiotensin I. It is uncertain whether drugs that block angiotensin II receptors such as saralasin would be efficaceous in blocking angiotensin III drinking behavior. The experiment described below was designed to assess whether the inability of saralasin to block water deprivation thirst was due to angiotensin III accumulation by this alternate synthetic pathway.

MATERIALS AND METHODS

Adult male Sprague-Dawley rats (275–350 g) were implanted with a lateral cerebroventricular cannula [7] under pentobarbital sodium anesthesia (40 mg/kg IP). Three days were allowed for recovery; then water was withheld for 48 hr. Rats were infused via the cerebral ventricle (IVT) at a rate of 2 μl/min with a tubing and tether system connected to the cannula. The infusions were started 15 min before water access and continued for 60 min. Four infusates were used: a) artificial cerebrospinal fluid (CSF, [4]), b) saralasin, 20 μg/hr, c) SQ 20,881 (SQ, a converting enzyme inhibitor) 50 μg/hr and d) combined saralasin and SQ at the same doses. The drugs were dissolved in artificial CSF. Drinking and urine volumes were recorded at regular intervals for 4 hr. Urine produced during the 4 hr test was assayed for sodium and potassium content by flame photometry. Data from the four groups (N=7 each) were compared by one-way analysis of variance. Confirmation of the ventricular cannula placement was obtained by infusing fast green dye for 15 min and observing its distribution in the ventricular system.

RESULTS

Figure 1 shows the average drinking and urine volumes of the pooled data for all groups of rats and the pooled error variance. The mean of each treatment group is indicated as follows: 1—CSF, 2—saralasin, 3—SQ and 4—saralasin + SQ. Analysis of variance showed that at each time point no differences ($F<2$) occurred either in drinking or urine volume. Absolute sodium and potassium excretion during the 4 hr test is shown in Table 1. No significant differences ($F<2$) were observed.

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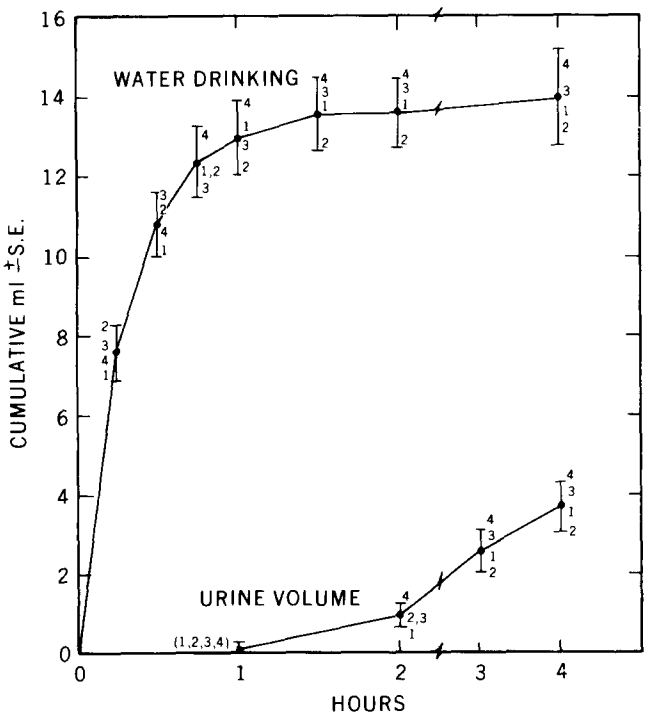

FIG. 1. Cumulative water ingestion and urine volume of 48 hr water deprived rats. Intracerebroventricular infusions were started 15 min before water access (time 0) and continued for 60 min. The pooled mean and SE of four treatment groups (N=28) are shown. The means of each treatment are indicated as follows: 1) CSF infusion, 2) saralasin infusion, 3) SQ 20,881 infusion and 4) saralasin + SQ infusion.

TABLE 1

ABSOLUTE URINARY SODIUM AND POTASSIUM EXCRETION AFTER IVT INFUSIONS OF SARALASIN, SQ 20,881 OR COMBINED TREATMENT IN 48 HR WATER DEPRIVED RATS

IVT Infusion	Na^+, μEq ± SE	K^+, μEq ± SE
CSF	73 ± 33	131 ± 41
Saralasin	47 ± 23	129 ± 30
SQ 20,881	57 ± 15	132 ± 17
Saralasin+SQ	38 ± 9	131 ± 35

DISCUSSION

The importance of angiotensin in water deprivation drinking behavior has been questioned because angiotensin II receptor blockade does not affect [6], or only briefly delays [3], water ingestion. Angiotensin peptide fragments possess various degrees of activity depending upon the specific effect of angiotensin II being examined [5]. Angiotensin III, the 2–8 fragment, retains considerable dipsogenic potency [2]. Formation of this peptide from angiotensin I can occur by two routes, both of which require converting enzyme [1,5]. Therefore, a combination of converting enzyme inhibition and angiotensin II receptor blockade in water deprived rats provides a paradigm where a role for either angiotensin II or III should be detectable. The doses of SQ and saralasin were sufficiently high to block drinking induced by large amounts of IVT angiotensin I or II, respectively [2,8]. Failure of this treatment to alter drinking after water deprivation strengthens the opinion that angiotensin, at sites reached from the cerebral ventricles, is not an absolute requirement for drinking after water deprivation.

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