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# Intracerebroventricularly Infused Angiotensin II or III do not Alter Voluntary Alcohol Intake in Rats

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GRUPP, L. A. AND S. HARDING. *Intracerebroventricularly infused angiotensin II or III do not alter voluntary alcohol intake in rats.* PHARMACOL BIOCHEM BEHAV 51(4) 593-599, 1995.—Subcutaneous injections of angiotensin (ANG) II or III in the periphery reduce alcohol intake and raise water intake. These peptides do not cross the blood-brain barrier and cannot reach the angiotensin receptor-rich sites surrounding the lateral and third ventricles. To examine the effect on alcohol intake of ANG II and III at these ventricular sites, groups of rats were first trained to drink alcohol using a limited access procedure, then surgically prepared with chronic indwelling lateral or third ventricular cannulae, and then reoffered daily 40-min access to alcohol. Neither ANG II (25-200 ng) nor ANG III (25-100 ng) had any effect on alcohol consumption at either of the two ventricular sites. Water consumption was significantly enhanced by both peptides at both sites and could be attenuated by prior treatment with the ANG II antagonist $Sar^1Thr^8$-ANG II. The SC administration of ANG II was able to produce a significant reduction in alcohol drinking. These findings demonstrate that ICV administered ANG II or ANG III do not modulate alcohol drinking and that changes in alcohol intake do not result from the thirst promoted by ANG II. Sites in the periphery may be more involved in the interaction between angiotensin and alcohol consumption.

Alcohol drinking Brain Angiotensin II Thirst Angiotensin III Lateral ventricle Third ventricle Intracerebroventricular Angiotensin antagonist Sarthran

ALCOHOL intake can be altered by a number of transmitters and peptides including dopamine (16), serotonin (22), the opiates (4,14), and angiotensin (6,11). With the exception of angiotensin (ANG) II (19), most of the agents tested gain access to the CNS when given systemically. For example, the dopamine agonist apomorphine (18), the serotonin uptake inhibitor fluoxetine (27), and the opiate antagonist naloxone (14) all produce significant reductions in alcohol intake, possibly through a combination of their peripheral and central actions. ANG II does not normally penetrate the blood-brain barrier. Systemic injections of ANG II reduce alcohol intake as well as stimulate water intake (21) and previous work (9) has shown that part of this action is mediated at the subfornical organ, a circumventricular brain structure rich in ANG II receptors but outside the blood-brain barrier. In that study animals that were first lesioned in the subfornical organ, exposed to alcohol, and then tested with ANG II, displayed significantly less suppression of alcohol intake compared to a control group.

Angiotensin receptors are located in a variety of hypothalamic sites that surround the lateral and third ventricles of the brain. Research (24) has shown that intraventricular (ICV) ANG II antagonists can reduce the uptake of blood-borne ANG II, suggesting that ICV ANG II can act on circumventricular organs. However, some of these brain sites do not appear to be directly accessible by blood-borne (systemic) administration of ANG, because tracer studies have shown that intravenous horseradish peroxidase is found around the subfornical organ but does not spread into the brain parenchyma (19). Similarly, ICV horseradish tracer does not spread to the circumventricular organs (19). It is possible, then, that ICV-administered ANG II might gain access to a set of ANG receptors that is different from those in the circumventricular or-

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gans occupied by systemically administered ANG II. The purpose of the present study was to assess whether ICV stimulation of ANG receptors that are located around the third and lateral ventricles exert an effect on alcohol intake.

A number of studies have shown that ANG-converting enzyme (ACE) inhibitors also reduce alcohol intake (12). It has been suggested that this effect might be mediated through an elevation of central ANG II activity (12). If the periventricular ANG II receptors participate in this effect, then the ICV administration of ANG II would be expected to produce a decrease in alcohol consumption.

## METHOD

### *Subjects*

Naive male Wistar rats (Charles River, Montreal, Canada) weighing 200–250 g at the beginning of the experiment, were used. They were housed singly and maintained on a reverse 12 L : 12 D cycle with lights off at 0700 h. Purina rat chow and tapwater were available in home cages ad lib.

### *Alcohol Intake*

The limited access drinking procedure was used (15,17) to promote alcohol consumption to blood alcohol levels that produce a pharmacodynamic effect (5). This procedure, which produces regular and reliable levels of alcohol intake, allows the administration of drug treatments to coincide with times of maximal alcohol consumption. The daily schedule was to weigh the rats, then transfer them to individual drinking cages (30 × 20 × 15 cm) equipped with two 15-ml graduated tubes (0.1-ml gradations), one containing an alcohol solution in tapwater, the other tapwater. The position of these tubes was alternated daily, and no food was available in the drinking cage. After 40 min elapsed the animals were returned to their home cages and the amounts of alcohol and water consumed were recorded. This took place during the dark cycle, when the animals were awake and most active. The limited access procedure employs a gradual exposure of the animals to the taste and effects of alcohol. Initially the animals were offered a choice between 3% (wt./vol.) alcohol and water for at least 10 days and then a choice between 6% (wt./vol.) alcohol and water for a further 10 days. Only those animals whose average 6% alcohol intake over the final 5 days exceeded the metabolic rate of 0.3 g/kg (5 ml/kg) went on to surgery. Seventy-five of 90 animals met this criterion.

### *Surgery*

The rats were divided into two groups matched for 6% alcohol drinking and designated to receive either lateral or third ventricle cannulae. Rats were anesthetized with 60 mg/kg pentobarbital, and 22-ga guide cannulae (Plastic Products, Roanoke, VA) were chronically implanted into either the left lateral ventricle (coordinates: A-P −0.8 mm from bregma; lateral 1.5 mm from sagital suture; depth 4.5 mm from skull surface; tooth bar −2.5 mm) or the third ventricle (coordinates: A-P −2.5 mm from bregma; lateral 0 mm from sagital suture; depth −9.5 mm from skull surface; tooth bar −3.3 mm). Obturators were cut to the same length as the guide cannulae and inserted into the cannulae when they were not in use. The injection cannula protruded 0.2 mm beyond the tip of the guide cannula. At least 1 week of recovery was allowed before the experiment proceeded.

### *Cannula Placement*

After surgery, the functionality and accuracy of the cannula placement were tested. Because ICV ANG is known to elicit water intake in sated rats (2), all rats were given one infusion through the cannulae with 100 ng ANG II. They were immediately transferred to drinking cages where free access to water was available and then returned to their home cages after 40 min. Only those rats whose consumption exceeded 10 ml/kg continued in the experiment. Thirteen rats in the third ventricle group and seven in the lateral group did not meet the criterion. Animals in each of the lateral and third ventricle groups were then subdivided into three groups equated for 6% alcohol intake.

### *ICV Treatment*

Ten days before ICV treatment a baseline of 6% alcohol and water intake was re-established for the three lateral and the three third-ventricle groups. The daily limited access procedure continued as before except that before being placed into the drinking cages the animals received their respective ICV infusions. The solutions were infused slowly over a 10-s period and the injection cannula remained in place for an additional 4 s so as to allow for diffusion of the solution away from the tip of the cannula.

The three lateral ventricle groups received daily ICV infusions of either ANG II, Sar$^1$Thr$^8$-ANG II, or the saline vehicle. ANG II (25, 50, 100, 200 ng; i.e, 25, 48, 96, 192 pmol) and Sar$^1$Thr$^8$-ANG II (1, 5, 10 μg; i.e., 1, 5, 10.5 nmol) were given in ascending order of dose with each dose repeated on 5 successive days. Two days intervened between doses. The doses of ANG II (Hypertensin Ciba) or Sar$^1$Thr$^8$ ANG II (Bachem) were infused in a volume of 4 μl. The saline vehicle infusion volume was also 4 μl over a 10-s period.

The three third-ventricle groups received daily ICV infusions of either ANG II, ANG III (Bachem), or the saline vehicle. ANG II (25, 50, or 100 ng) and ANG III (25, 50, or 100 ng; i.e., 27, 54, or 107 pmol) were given in ascending order of dose with each dose repeated on 5 successive days. Two days intervened between doses. In the next phase (5 days), all groups continued to receive their respective doses of either 100 ng ANG II, ANG III, or saline, but in addition they were pretreated with 1 μg of the ANG II antagonist Sar$^1$Thr$^8$-ANG II. Infusions of Sar$^1$Thr$^8$-ANG II occurred immediately before the ANG infusions and were given in a similar manner to the ANG infusions. ANG II, ANG III, and Sar$^1$Thr$^8$-ANG II were dissolved in saline and stored in plastic containers. ICV infusion or injection used only plastic syringes and accessories. All solutions were prepared fresh daily.

To assess the ability of peripherally administered ANG II to alter alcohol intake, animals from the ANG II and ANG III groups were combined and designated to receive saline by the ICV route followed immediately by SC injections of ANG II, 400 μg/kg. The saline control group received saline ICV and saline SC. This phase lasted 5 days.

### *Data Analysis*

Alcohol and water intake for each rat were averaged over the 5 days of each phase. One-way analyses of variance (ANOVA) were performed followed by post-hoc Duncan's tests, which examined relevant pairwise group comparisons. The $\alpha$-coefficient was set at 0.05.

## RESULTS

### *Lateral Ventricle*

*Effect of saline vehicle on alcohol and water intake.* Figure 1A shows that alcohol intake was generally not altered by the ICV administration of the saline vehicle [$F(6, 54) = 2.03$, NS]. Compared to the baseline phase, alcohol intake in the first saline phase was significantly reduced, but this reduction did not persist through the following four saline phases and may have been related to the initiation of the central infusions. Water intake was low and did not change significantly over the course of the experiment [$F(6, 54) = 1.61$, NS]. Both alcohol and water intake were not changed in the final phase when the ICV infusions were stopped. Taken together, these findings indicate that the infusion per se of the saline vehicle had no systematic effect on either alcohol or water consumption.

*Effect of ANG II antagonist Sar$^1$Thr$^8$-ANG II on alcohol and water intake.* Figure 1B shows that the three doses of this ANG II antagonist had no effect on either alcohol [$F(4, 36) = 0.17$, NS] or water intake [$F(4, 36) = 1.05$, NS] across all phases of the experiment. The substitution of the saline vehicle for the Sar$^1$Thr$^8$-ANG II in the final phase did not result in a change in either alcohol or water consumption, confirming the lack of effect of the antagonist on intake. There was a tendency for the 5-$\mu$g dose to increase water intake when compared to baseline intake. This agonist-like effect of an ANG II antagonist has been reported previously (23), but in the present experiment the effect was not large and was observed only at the intermediate 5-$\mu$g dose. These findings suggest that the ICV administration of this ANG II antagonist does not influence the intake of alcohol and water.

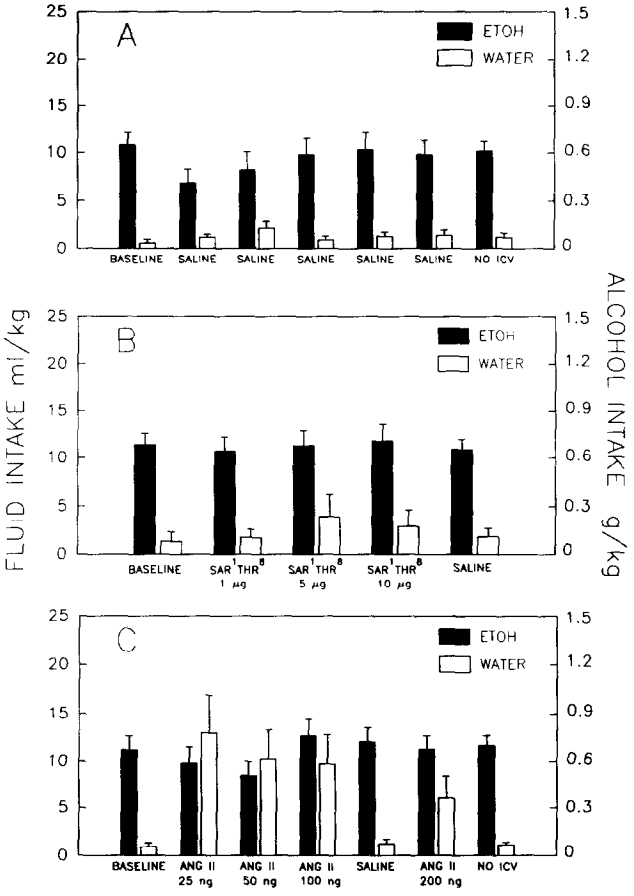


FIG. 1. The effect of lateral ventricle infusions of saline (A), Sar$^1$Thr$^8$-ANG II (B), and ANG II (C) on 6% (wt./vol.) alcohol and water intake during 40-min limited access drinking sessions before (baseline) and during treatment (subsequent phases). 1.05 ng ANG II = 1 pmol ANG II; 0.96 ng Sar$^1$Thr$^8$-ANG II = 1 pmol Sar$^1$Thr$^8$-ANG II. All treatment drugs were dissolved in 0.9% saline and were administered over a 14-s period in a volume of 4 $\mu$l immediately before the drinking session. T bars represent the SEM.

*Effect of ANG II on alcohol and water intake.* Figure 1C illustrates the effect of four doses of ANG II on alcohol intake. A one-way ANOVA revealed a nonsignificant effect of phase [$F(6, 54) = 1.78$, NS], indicating that none of the doses produced a significant reduction in alcohol intake. There was a tendency for the 50-ng dose to decrease alcohol intake, but post-hoc tests did not show this change to be significantly different from baseline intake.

Figure 1C also illustrates the effect of ANG II on water intake. In contrast to its effect on alcohol drinking, ANG II produced a robust increase in water intake [$F(6, 54) = 6.61$, $p < 0.0001$]. Post-hoc Duncan's tests revealed that compared to baseline levels, water intake was significantly elevated at least 10-fold by the 25, 50, and 100-ng doses of ANG II. Although a dose-response effect is usually seen with ascending doses of the peptide, the maximal effect in the present experiment was achieved by the lowest (25-ng) dose. Although the 200-ng dose increased water intake, the change was not significant compared to either baseline, saline, or no ICV levels of water consumption, and was significantly smaller compared to the effect of the 25-ng dose. It is possible that the repetitive dosing with ANG II may have led to the development of tolerance and a less than optimal effect of the 200-ng dose on water consumption. These findings illustrate that doses of ANG II administered by the ICV route that produce profound increases in water consumption do not have any significant effect on alcohol intake.

### *Third Ventricle*

*Effect of saline vehicle on alcohol and water intake.* Figure 2A illustrates mean alcohol and water intake across all phases of the experiment. A one-way ANOVA showed that alcohol intake was not significantly altered by the ICV administration of the saline vehicle [$F(4, 20) = 1.49$, NS]. Water intake was low but changed significantly over the course of the experiment [$F(4, 20) = 4.19$, $p < 0.01$]. Post-hoc Duncan's test indicated that administration of the ANG II antagonist Sar$^1$Thr$^8$-ANG II in the final phase produced an increase in water intake compared to the immediately preceding phase in which only saline was administered. These findings indicate that ICV saline does not change alcohol or water consumption. As was the case with lateral ventricle infusions, the ANG II antagonist appeared to have some agonist properties in that it produced a small but significant rise in water drinking. The stronger agonist effect of Sar$^1$Thr$^8$-ANG II on water intake at the third ventricle may have been related to the easier accessibility of the peptide at this infusion site to the periventricular hypothalamic sites that stimulate water intake.

*Effect of ANG III on alcohol and water intake.* Figure 2B shows that the three doses of ANG III had no effect on alcohol intake [$F(4, 20) = 0.90$, NS). Pretreatment with the ANG II antagonist in the final phase did not alter the pattern of alcohol consumption. On the other hand, ANG III produced a small but significant elevation in water consumption [$F(4,$

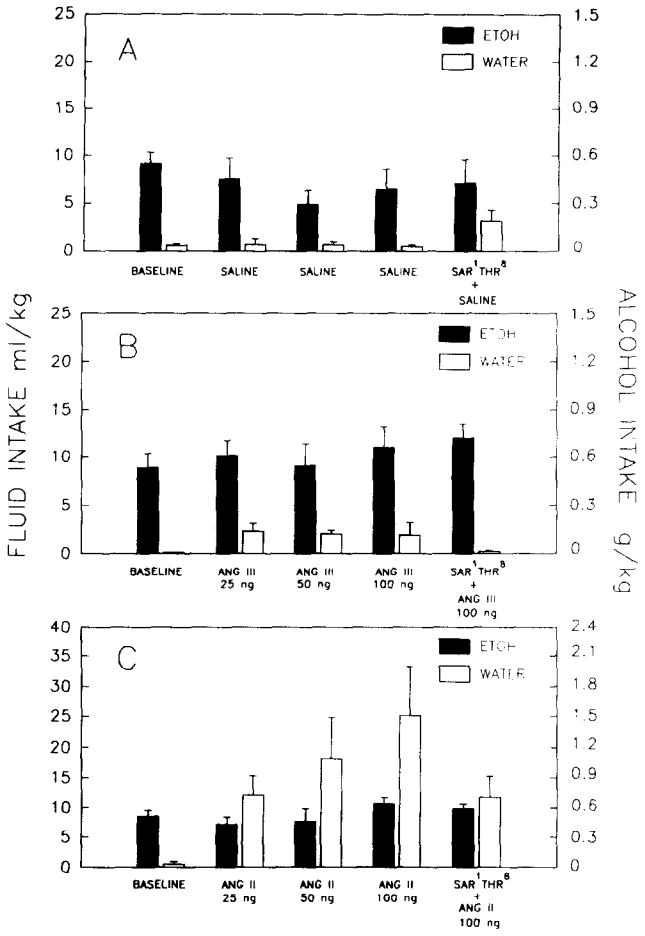


FIG. 2. The effect of third-ventricle infusions of saline (A), ANG III (B), and ANG II (C) on 6% (wt./vol.) alcohol and water intake during 40-min limited access drinking sessions before (baseline) and during treatment (subsequent phases). In the final phase animals received Sar¹Thr⁸-ANG II ICV immediately before treatment. 0.93 ng ANG III = 1 pmol ANG III. All treatment drugs were dissolved in 0.9% saline and were administered in volumes of 4 μl over a 14-s period immediately before the drinking session. T bars represent SEM.

20) = 2.7, $p < 0.05$], with both the 25- and 50-ng doses producing significant increases. Pretreatment with the ANG II antagonist before ANG III treatment in the final phase attenuated the ANG III-induced water intake. These findings show that the effects of ANG III on alcohol and water consumption can be dissociated. Whereas ANG III can induce a small but significant increase in water intake, it has no effect on alcohol intake.

*Effect of ANG II on alcohol and water intake.* Figure 2C illustrates the effect of the three doses of ANG II on alcohol and water intake. A one-way ANOVA of the alcohol data revealed a nonsignificant effect of phase [$F(4, 16) = 1.43$, NS), indicating that none of the doses produced a significant change in alcohol intake. Intake in the final phase in which the animals were pretreated with Sar¹Thr⁸-ANG II before ANG II did not appear to be different from previous phases.

Figure 2C also illustrates the effect of ANG II on water intake. As was the case with the lateral ventricle, and in contrast to its effect on alcohol drinking, ANG II produced a robust dose-dependent increase in water intake [$F(4, 16) = 6.33$, $p < 0.003$]. Post-hoc Duncan's tests revealed that compared to baseline levels, water intake was significantly elevated at all ANG II doses, and that the 100-ng dose produced significantly greater water intake than the 25-ng dose. Pretreatment with the ANG II antagonist in the final phase produced a significant reduction in water intake, illustrating competitive antagonism. These findings again illustrate that the effects of ANG II on alcohol and water intake are not necessarily interrelated and can occur independently of each other.

Figure 3 illustrates the effect of peripherally administered ANG II (400 μg/kg by the SC route) on alcohol and water intake in a group of rats that had previously received ANG II or ANG III ICV. Two-way ANOVA revealed a significant effect of phase for alcohol consumption [$F(2, 24) = 5.55$, $p < 0.01$] and significant effects of group [$F(1, 24) = 9.05$, $p < 0.01$], phase [$F(2, 24) = 16.92$, $p < 0.0001$], and the interaction of group and phase [$F(2, 24) = 8.22$, $p < 0.001$] for water intake. Whereas the SC administration of the saline vehicle had no effect on either alcohol intake ($t_5 = 1.9$, NS) or water intake ($t_5 = 1.3$, NS) compared to baseline (panel A), a small but nonsignificant reduction was evident. ANG II produced a significant increase in water intake ($t_7 = 4.1$, $p < 0.005$) and a significant reduction in alcohol drinking ($t_7 = 2.4$, $p < 0.05$) (panel B) when compared to its own baseline level of intake. Saline injections per se rarely interfere with

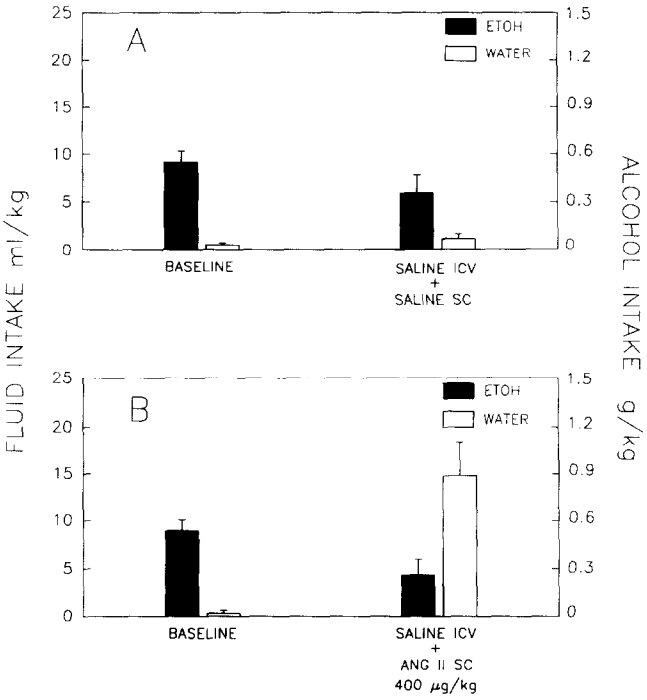


FIG. 3. The effect of peripheral injections of saline (A) and ANG II (B) on 6% (wt./vol.) alcohol and water intake during 40-min limited access drinking sessions before (baseline) and during treatment (subsequent phase). In the treatment phase animals received third-ventricle infusions of saline immediately before SC saline (A) or ANG [400 μg/kg (B)]. All treatment drugs were dissolved in 0.9% saline and were administered in volumes of 4 μl ICV and 1 ml/kg SC over a 14-s period immediately before the drinking session. T bars represent SEM.

normal alcohol drinking behavior, but in this case a nonsignificant but small reduction was observed. The absence of a between-group difference in alcohol intake was attributable to this change in alcohol intake in the saline-injected group. Taken together, these findings are congruent with a large body of evidence showing that peripherally administered ANG II produces a reduction in alcohol consumption (6, for a review), and demonstrate that this reduction can occur in animals whose alcohol drinking did not respond when either ANG II or ANG III was administered into the third ventricle.

## DISCUSSION

Although the peripheral administration of the peptides ANG II, ANG III, or drugs that are known to increase ANG II activity produce a dose-dependent reduction in alcohol intake [see (6) for a review], little is known about the central effect of these peptides on alcohol intake. It is unlikely that the peripheral SC administration of ANG II and ANG III would stimulate the same sites as ICV-administered ANG II because these peptides do not cross the blood-brain barrier. The purpose of this study was to examine the effect on alcohol intake of ANG II and ANG III administered by the central ICV route.

Direct infusions of ANG II into the lateral ventricle of water-sated rats produced the classical increase in thirst, as evidenced by a highly significant elevation in water intake. Alcohol intake, however, was not altered by any of the four doses tested ranging from 25–200 ng. These findings indicate that the functional stimulation of ANG II receptors in the vicinity of the lateral ventricle does not exert an influence on alcohol consumption. Furthermore, the finding that alcohol consumption does not decrease in the face of a robust increase in water consumption points to a clear dissociation between the effects of ANG II on alcohol and water intake. This adds weight to the argument (11) that the decrease in alcohol consumption produced by SC-injected ANG II is not an artifact of the increase in water intake or the necessary consequence of a behavioral conflict between the consumption of alcohol and the thirst-inducing properties of ANG II. Although ANG II did not alter alcohol intake, neither did the blockade of the receptors with three different doses of the ANG II antagonist $Sar^1Thr^8$-ANG II. These findings, which replicate the results with peripherally administered antagonist (8), add further support to the conclusion that the ANG receptors in the vicinity of the lateral ventricle do not appear to play a role in modulating alcohol drinking.

The ICV injection of ANG II or ANG III into the third ventricle again produced the well-documented increase in water intake. ANG II injections induced a dose-dependent and more potent increase in water consumption compared to ANG III, a finding that replicates earlier work showing that doses of ANG II in excess of 25 ng elicit greater water intake than equivalent ANG III doses (25). The greater efficacy of ANG II may be related to a longer effective half-life (25). The dose-dependent stimulation of water intake with third-ventricle infusions contrasts with a uniform increase in water intake seen with lateral ventricle infusions. This might be related to the differences in cannula placement relative to the ANG II-sensitive sites in the brain and possible corresponding differences in accessibility to the receptors. Pretreatment with the ANG antagonist $Sar^1Thr^8$-ANG II significantly reduced the dipsogenic effects of both peptides. Notwithstanding these robust effects on water drinking, no doses of either ANG II or ANG III had an effect on alcohol intake, nor did the ANG antagonist exert an effect of its own. These findings show that the functional stimulation of ANG II receptors in the sites around the third ventricle does not appear to exert an influence on alcohol consumption. These results replicate the finding with lateral ventricular placements in that the ANG peptides did not affect alcohol intake but did enhance water intake. On the other hand, when ANG II was administered by the peripheral SC route to the same animals whose alcohol consumption was refractory to ICV-administered ANG II, alcohol consumption dropped significantly from a mean of 9 ml/kg to a mean of 4 ml/kg. These findings demonstrate that ICV drug administration in no way permanently altered the ability of the animals to respond to the alcohol-inhibiting effects of ANG II. Furthermore, they show that the sites that respond to ANG II stimulation with a decrease in alcohol drinking do not appear to be accessed by ICV-administered ANG II. Previous reports have implicated the subfornical organ (9) and ruled out the area postrema (10) in the inhibitory effect of ANG II on alcohol intake. ANG II, however, can stimulate receptors in the liver, kidney, adrenals, and other peripheral sites, and these sites could play a role by conveying information to the CNS either hormonally or through changes in activity of the vagus nerve.

Taken together, the results indicate that stimulation of the brain areas accessed by the lateral and third ventricles that are rich in ANG receptors does not produce a reduction in alcohol intake. At the same time, ICV application of ANG to these areas does increase water consumption and Wright et al. (26) have reported increases in blood pressure to similar doses of ICV-administered ANG II and ANG III. These considerations illustrate the dissociation between the behavioral and cardiovascular-endocrine properties of ANG II and do not support the view that when the reduction in alcohol intake by SC ANG II is observed, these other actions of ANG II act as nonspecific sources of stimulation that merely interfere with the animals' ability to consume alcohol (3). Instead, these findings support the view that the peripherally initiated effect of ANG II on alcohol intake is an example of a behavioral action of this peptide.

A recent study (3) reported that continuous infusion of ANG II into the lateral ventricle over a 7-day period increased the intake of alcohol. This finding contrasts with the present results, which find no effect of either ANG II or ANG III. Methodologic differences exist between the two experiments. For example, Fitts (3) tested the Long-Evans strain of rats under continuous access conditions in which alcohol was freely available 24 h/day. ANG II was administered continuously using an osmotic minipump. In the present experiment, alcohol was available for a 40-min period and consumed in a bout. The entire dose of ANG II was administered as a pulsatile infusion over a brief period of time, and the Wistar strain of rats was used. Although these differences might, to some extent, account for the different findings, alternative explanations can be considered. Close examination of the data in the Fitts report (3) reveals that ANG II did not produce a consistent increase in alcohol intake. Even though both the 5- and 20-pmol/h doses were able to increase water intake and were therefore both functionally active, only the 20-pmol dose increased alcohol intake. Furthermore, the increase observed with this dose was not reliable. It was observed only slightly more than 50% of the time. Over the 7-day period when ANG II was administered, water intake was consistently elevated, but the increase in alcohol drinking was seen only during the

middle 4 days, with the first and last 2 days showing no significant change. The present data point to the conclusion that ICV ANG II does not have a reliable effect on alcohol intake.

In the present study, care was taken to test only those animals that consumed enough alcohol to achieve a pharmacologic effect. In a 40-min period our animals consumed approximately 600 mg/kg, which is a level clearly above the rats' metabolic rate of 300–350 mg/kg per h. In the Fitts study (3), the alcohol intake was extraordinarily low (i.e., approximately 4 ml of a 4.7% (wt./vol.) alcohol solution or 376 mg/kg per day. This low intake level makes it unlikely that those animals were experiencing a pharmacologic effect from the alcohol they consumed. The different outcomes of the two experiments with respect to the effect of ANG II might be related to the very different levels of the alcohol effect experienced. In fact, the increase in intake of the alcohol solution observed by Fitts (3) parallels the increase in water intake; coupled with the possibility that the alcohol consumed by those animals did not have pharmacologic consequences, this suggests that the animals in that experiment might not have been treating the alcohol solution as a drug solution per se, but rather as an alternative fluid source. Consequently, like water, intake increased when ANG II was administered.

Although peripheral injections of ANG II reduce alcohol intake (7,13), a number of laboratories have also demonstrated that the IP administration of ANG-converting enzyme inhibitors, agents that prevent the synthesis of ANG II, also lead to a reduction in alcohol drinking (1,12,13). One explanation put forward to account for this paradoxical finding proposes that the ACE inhibitors cause a rise in central ANG II because elevated ANG I resulting from peripheral blockade of ANG II synthesis passes into the CNS, where it is converted to ANG II. This increase in central ANG II is hypothesized to cause the reduction in alcohol intake. The results of the present experiment, in agreement with other work (20), do not support this hypothesis, because ICV-administered ANG II did not alter alcohol intake.

In summary, rats receiving ANG II or ANG III in either the lateral or third ventricle increased their water intake but showed no change in their alcohol consumption. These data do not support the idea that changes in alcohol drinking following ANG II administration are related to the increase in thirst and water intake produced by this peptide. The data indicate that the sites mediating the reduction in alcohol intake when ANG II is injected peripherally are anatomically separate from the sites reached when ANG II is infused into the lateral and third-ventricular sites tested here.

ACKNOWLEDGEMENTS

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