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Central α-Adrenergic Agonists and Need-Induced 3% NaCl and Water Intake

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YADA, M. M., P. M. de PAULA, J. V. MENANI AND L. A. de LUCA, JR. *Central α-adrenergic agonists and need-induced 3% NaCl and water intake.* PHARMACOL BIOCHEM BEHAV **57**(1/2) 137–143, 1997.—In the present study, noradrenaline (NOR, α-non-specific adrenergic agonist), clonidine (CLO, α_2), phenylephrine (PHE, α_1) or isoproterenol (ISO, β-agonist) was injected in the medial septal area (MSA) of water-deprived, sodium-deplete or food-deprived rats. NOR (80, 160 nmol) inhibited the intake of 3% NaCl, water deprivation-induced and meal-associated water intake. Food deprivation-induced food intake and 10% sucrose intake were not altered by NOR. CLO (10, 20, 30, 40 nmol) inhibited (80–100% inhibition compared to control during 60 min) the intake of 3% NaCl, water deprivation-induced and meal-associated water intake. CLO had a weaker inhibition on food and 10% sucrose intake (30–50% less than the control during 60 and 15 min, respectively). PHE (160 nmol) inhibited 3% NaCl intake and 10% sucrose intake (30% less than the control for 15–30 min). ISO (160 nmol) did not alter water or 3% NaCl intake. NOR induced an increase, CLO and ISO induced a decrease, and PHE no alteration in mean arterial pressure. NOR did not alter water or 3% NaCl intake when injected unilaterally into the caudate nucleus. The results suggest that NOR injected in the MSA acts on α_2-adrenergic receptors inducing a specific inhibition of 3% NaCl and water intake. © 1997 Elsevier Science Inc.

Noradrenaline Clonidine Phenylephrine Isoproterenol Adrenergic agonists Medial septal area Water intake Salt intake Food intake Blood pressure

INGESTIVE behavior is controlled by noradrenaline (NOR) acting on the central nervous system. This neurotransmitter is important for the activation of food intake and it has an inhibitory action on water intake (10,13).

How specific is the inhibitory action of NOR on fluid intake is still a question to be solved. The contrast between induction of food intake and inhibition of water intake produced by NOR when injected centrally (10,13) is a suggestion that the inhibition is specific for water intake. However, we do not know if NOR also inhibits the intake of other fluids such as mineral or sweet solutions.

In the present work we investigate two situations of interest to test the central effect of NOR and other adrenergic alpha (clonidine-α_2, phenylephrine-α_1) and beta (isoproterenol) agonists on the intake of water and watery solutions. In one situation, animals ingest a fluid (water or a solution of NaCl) in response to an internal deficiency (need-induced intake) induced by water deprivation or sodium depletion. In another situation, ingestive behavior is expressed for a fluid, sucrose solution, when energetic nutrients are available continuously to the animal (need-free intake). Rats submitted to an acute sodium depletion for 24 h by the combination of furosemide injection and removal of ambient sodium show a quick and consistent sodium intake (12). The rats also ingest excess amounts of sucrose solution even when they have free access to balanced food (19).

Noradrenergic terminals and receptors are found in the medial septal area (MSA) (1,2,16). The injection of NOR or of clonidine in the MSA inhibits water intake induced by carbachol (15,18). An interesting question is whether NOR and its agonists injected in the MSA also inhibits need-induced water and sodium intake.

In the present study we demonstrate that NOR and its alpha adrenergic agonists, chiefly clonidine, injected in the MSA have a preferential inhibitory effect on need-induced water and 3% NaCl intake.

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METHODS

General Procedures

Animals. Male Holtzman rats weighing 260–300 g at the beginning of the experiment were individually housed in a room on a 12:12 light/dark cycle, beginning at 0700. Standard Purina pellets containing more sodium (0.5–1.0%) than the minimum daily requirement of the rat (The Harvard Bioscience Whole Rat Catalog), tap water and 3% NaCl solution were available ad lib unless otherwise noted. All experiments began between 1000 and 1400.

Brain surgery. The animals were maintained under ether or tribromoethanol (Aldrich) (20 mg/100 g b.w., IP) anesthesia throughout the surgery. A stainless steel guide-cannula (12 × 0.7 mm OD) was stereotaxically implanted in the brain with its opening protruding into the top of the MSA (co-ordinates: AP = 1.0 mm anterior to bregma; V = 3.9 mm from skull surface; L = 0.0 mm from sagittal midline; incisor bar: 2.5 mm above the interaural line). The cannula was secured to the top of the skull by dental cement and fastened with two screws. The insertion of a close fitting stylet kept the lumen free of debris and clots. A prophylactic dose of penicillin (30,000 IU) was given IM presurgically. The animals were allowed to recover from the surgery for at least four days before submitted to any experimental test.

Intracerebral injection techniques. Single pulse intracranial injections were made after gently removing the animal from its cage, replacing the stylet with an injector that protruded 2.0 mm beyond the tip of the guide-cannula and that was connected by PE-10 tubing to a 10.0 μl microsyringe, and injecting a total volume of 1.0 μl in a period of 30 s. Stylet and injector were always wiped with cotton soaked with 70% alcohol. After the injection, the injector was removed, replaced by the stylet, and the animal was returned to its cage for observation of its behavior.

Drugs. Noradrenaline hydrochloride (NOR), clonidine hydrochloride (CLO), phenylephrine hydrochloride (PHE) and isoproterenol (ISOP) were used. The drugs were dissolved in 0.9% saline (SAL). Doses ranging from 10 to 160 nmol were chosen from previous works (8,9,15). The natriuretic/diuretic compound furosemide was used for sodium depletion. All drugs were purchased from Sigma.

Histology. At the end of the experiments, the animals were deeply anesthetized with ether and perfused with 10% formalin through the left ventricle of the heart. After fixation in 10% formalin the brains were frozen, cut coronally into 20–30 μm sections and stained with hematoxylin-eosin for examination by light microscopy.

Statistics. Data are expressed as means ± SEM. Paired and non-paired *t*-tests were performed within and between groups, respectively. Two-way (drug and time as factors) analysis of variance (2-way ANOVA) was used for multiple comparisons between groups. All intervals of measurement (15, 30, 60 and 120 min) of ingestive behaviors were taken into account for factor time, except where indicated. Significance level was set at $p < 0.05$ for all tests. Each test described in the experimental protocols below was performed with a new group of animals. Each animal received different doses of one drug in a counterbalanced design. Each animal submitted to a test underwent from one to four experimental sessions according to the number of different doses of the drug that it received, separated by two- to three-day interval.

Experimental Protocols

Water deprivation-induced water intake test. Water and 3% NaCl were removed from the cage and then only food remained available for 30 h before water was returned to the animals. NOR, CLO, PHE, ISOP (10–160 nmol) or SAL was injected in the MSA 20 min before water burettes became available to the animals. Water intake was recorded only at 60 min in the absence of food.

Sodium depletion and the sodium appetite test. Sodium deficient food (powdered corn meal, 0.001% sodium, 0.33% potassium) replaced the regular pellets and the rats were sodium depleted by combining injection of furosemide with removal of ambient sodium. Each rat received one subcutaneous injection of furosemide (10 mg/ml). At the time of the first injection, food pellets were replaced by the sodium deficient food, the 3% NaCl (but not water) was removed, and the rat's cage was thoroughly washed to remove all ambient sodium. Thus, only sodium deficient food and water were available overnight for consumption.

The following day (24 h after the furosemide injection) the food was removed and the 3% NaCl was returned to the animals for ingestion. The cumulative intakes of 3% NaCl and water were measured at 15, 30, 60 and 120 min. This 120 min access to 3% NaCl will be referred to as the appetite test. Standard chow was returned to the rat at the end of the appetite test and was available continuously with water and 3% NaCl until the next depletion. NOR, CLO, PHE, ISOP (10–160 nmol) or SAL was injected in the MSA 20 min before the test.

Sodium appetite training tests. The rats were depleted at least twice before the actual experiments began, one time before the surgery and a second time at least four days after the surgery. The appetite tests corresponding to each one of those first two depletions were the appetite training tests and the animals received no drug but furosemide. Only animals which drank at least 3.0 ml of 3% NaCl in 30 min of the appetite training tests were admitted to the experiments.

Sucrose intake test. After two days of free access to 10% sucrose, the animals had it available with only water for two h daily, during five days. On the 5th day, NOR (160 nmol), CLO (20 nmol), PHE (160 nmol) or SAL was injected in the MSA 20 min before the sucrose solution was offered. The intakes of 10% sucrose and water were measured for 15, 30, 60 and 120 min.

Solid food and meal-associated water intake tests. The animals were deprived of food for 24 h, with only water available. NOR (160 nmol), CLO (20 nmol), PHE (160 nmol) or SAL was injected 20 min before food was returned and then food and water intakes were measured for 15, 30, 60 and 120 min.

Site specificity test. In order to investigate if the effects observed on fluid intake were due to spread of the agonists to remote sites of injection, we have also injected NOR (80 or 240 nmol) or CLO (20 or 30 nmol) in one μl unilaterally into the caudate nucleus of new groups of either water deprived or sodium deplete animals. The co-ordinates for reaching this nucleus were the same anteroposterior and depth used to reach the MSA, but with a lateral of 3.7 mm from the sagittal suture.

Arterial pressure test. Direct mean arterial pressure (MAP) was measured in unanesthetized and unrestrained rats. Three days after the cannula-implant, the animals were anesthetized with ether and a polyethylene (PE) tube (PE-10 connected to PE-50) was inserted into the abdominal aorta through the femoral artery of the right leg on the day before the experiment. The PE-50 end was tunneled subcutaneously to the back of the animal and connected, through a 30 cm length of PE-90, to a Narco (P-1000B) pressure transducer coupled to a multichannel recorder (NarcoTrace 40, Narco Bio-System, USA). After stabilization of the baseline recording, NOR (80

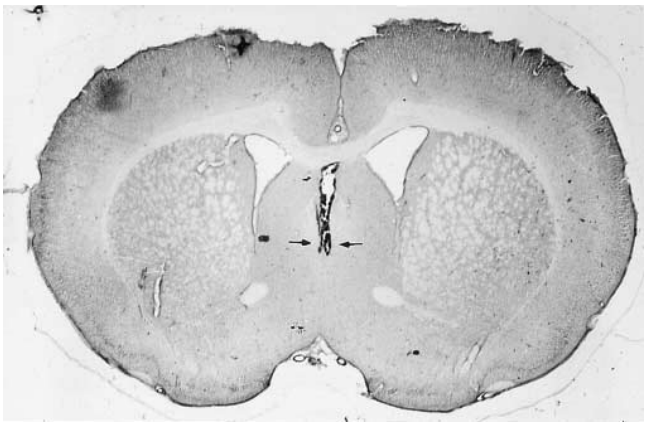

FIG. 1. Site of drug injection in the MSA as indicated by the arrows.

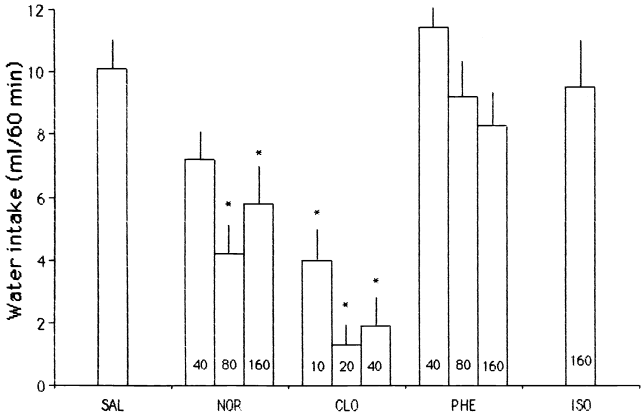

FIG. 2. Water intake of 30 h water deprived rats after 0.9% saline (SAL), noradrenaline (NOR), clonidine (CLO), phenylephrine (PHE) or isoproterenol (ISO) injection in the MSA. The numbers within bars represent the dose (nmol) of the drug. $n = 10$ animals per group. $*p < 0.05$ compared to SAL.

and 160 nmol, waterdeprivation and sodium deplete state, respectively), CLO (20 and 30 nmol, water-deprivation and sodium deplete state, respectively), PHE (160 nmol), ISOP (160 nmol) or SAL was injected in the MSA of either water deprived or sodium deplete animals and blood pressure was recorded for 20 min thereafter.

RESULTS

Histology

The central injection site was determined as the area of gliosis which is formed at the point of injection. The arrow indicates the injection site in the MSA (Fig. 1).

General Behavior

Except for clonidine, none of the agonists produced an observed alteration in behavior during the 20 min after the injection in the MSA. The highest dose (40 nmol) of clonidine produced immobility and midriasis that lasted from 10 to 20 min after the injection. The smaller doses (20 and 30, but not 10 nmol) produced a more transient immobility (about 5 min) in 50% of the animals.

Water Deprivation-Induced Water Intake

Water intake of 30 h water-deprived rats was inhibited by NOR (80, 160 nmol) and CLO (10, 20, 40 nmol) (Fig. 2) injected in the MSA. The maximum effective dose of CLO (20 nmol) induced a more potent inhibition than the smallest (80 nmol) effective dose of NOR (80% vs 40%, respectively). Neither PHE (40, 80, 160 nmol) nor ISO (160 nmol) altered water intake. Neither agonist altered water intake in hydrated animals. All differences are significant for $p < 0.05$ (unpaired *t*-test).

Sodium Appetite Test

NOR [$F(3, 26) = 5$), CLO ($F(3, 20) = 8.7$] and PHE [$F(2, 17) = 5.8$] injected in the MSA inhibited the 3% NaCl intake ($p < 0.05$, 2-way ANOVA, between doses) of 24 h sodium deplete rats (Figs. 3A,B,C). The inhibition induced by CLO was more effective both in duration and in strength (60–80% during at least 60 min for CLO versus 50–60% during 15–30 min for NOR and PHE). Also, the doses of CLO (20, 30 nmol) necessary to induce inhibition was 5 to 12 times smaller than the doses of NOR and PHE. ISO (160 nmol) did not alter the 3% NaCl intake (Fig. 3D). Neither agonist altered sodium intake in replete animals. There were no interaction between drug and time.

Sucrose Intake Test

The 10% sucrose intake was not altered by NOR (160 nmol) injected in the MSA (Fig. 4). CLO (20 nmol) and PHE (160 nmol) inhibited the 10% sucrose intake. CLO produced a 30% decrease in 10% sucrose intake at 15 min, but the intake went back to normal at 30 min and no significant difference between CLO versus SAL was found considering drug and time as factors for the 2-way ANOVA. PHE also induced a 30% decrease in 10% sucrose intake that stayed significant for 30 min and which was also significant ($p < 0.05$) for the 2-way ANOVA [$F(1, 11) = 8.5$, between doses]. There were no interaction between drug and time.

Food Intake and Meal-Associated Water Intake Test

Food intake (Fig. 5A) of 24 h food-deprived rats was not altered by NOR (160 nmol) or by PHE (160 nmol). CLO (20 nmol) induced a 40–50% inhibition of food intake at 15 and 60 min, but not at 30 and 120 min ($p < 0.05$, *t*-test). The inhibition was also significant for 2-way ANOVA [$F(1, 10) = 16$, between doses]. Meal-associated water intake (Fig. 5B) was inhibited ($p < 0.05$, 2-way ANOVA, between doses) by NOR (160 nmol) [$F(1, 9) = 18$] and by CLO (20 nmol) [$F(1, 10) = 21$]. The inhibition of meal-associated water intake induced by NOR and by CLO lasted 120 min and its magnitude was of 90–100% until 60 min and declined to about 50% at 120 min. Meal-associated water intake was not altered by PHE (160 nmol). There was no interaction between drug and time.

Site Specificity Test

The results of this test are summarized on Table 1. Water intake was inhibited by NOR (80 nmol) injected in the MSA, but it was not altered by NOR (80 nmol) injected into the caudate nucleus. CLO (20 nmol) injected in the MSA or into the caudate induced 98% and 43% inhibition of water intake, respectively, compared to control. The 3% NaCl intake was measured for 120 min, but the data show in the Table 1 is from 15 min since the effect of NOR is manifested mainly

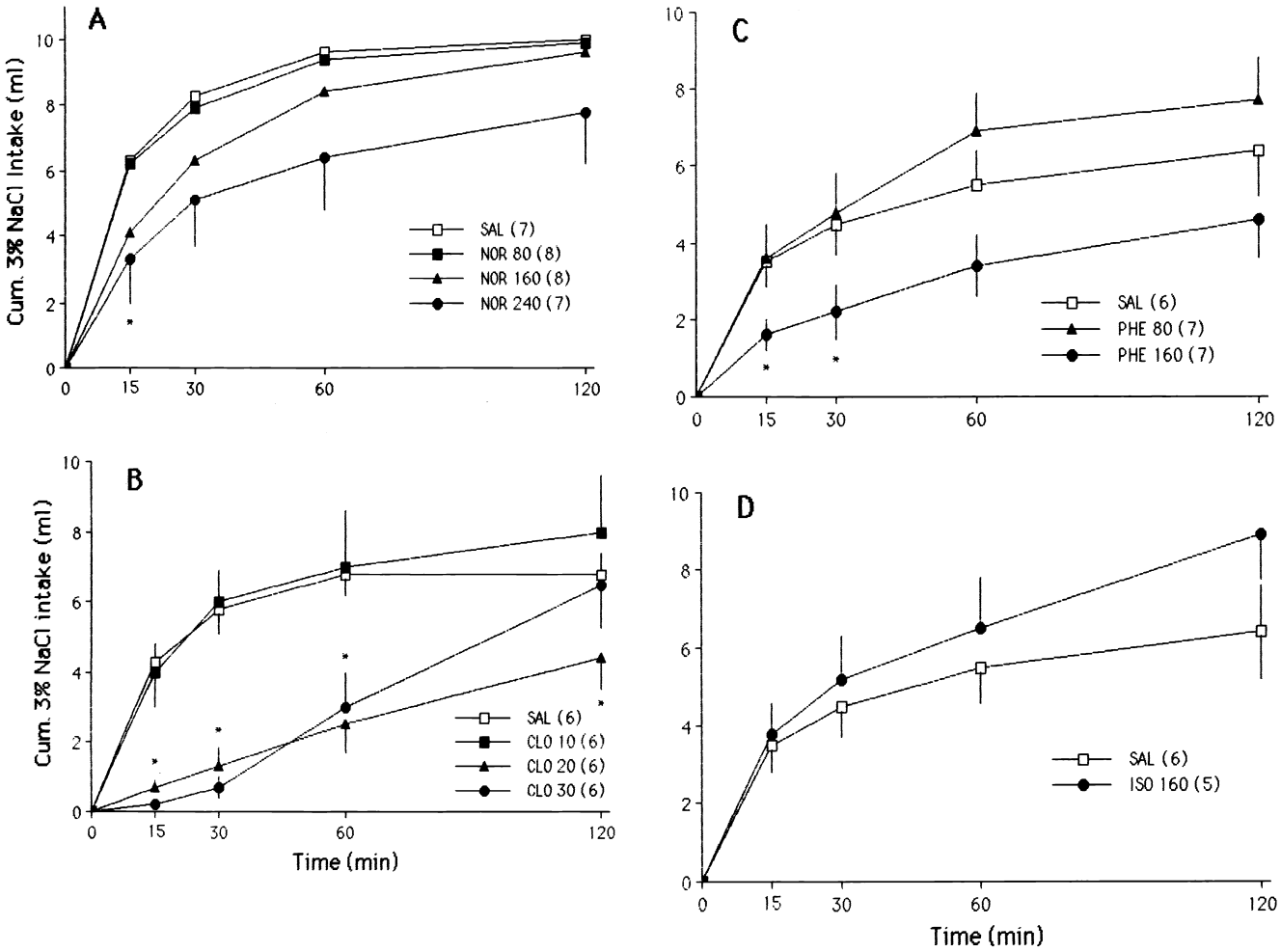

FIG. 3. Cumulative 3% NaCl intake of 24 h sodium deplete rats after injection of adrenergic drugs in the MSA: A) noradrenaline (NOR, 40, 80, 160 nmol), B) clonidine (CLO, 10, 20, 30 nmol), C) phenylephrine (PHE, 80, 160 nmol) and D) isoproterenol (ISO, 160 nmol). Number of animals in parenthesis. *$p < 0.05$ compared to SAL (0.9% saline) injection.

during this time (Fig. 3A). The 3% NaCl intake was inhibited by NOR (240 nmol) and by CLO (30 nmol) injected in the MSA. The injection of NOR (240 nmol) into the caudate did not alter the 3% NaCl intake, although the result was not different from the injection in the MSA. The injection of CLO (30 nmol) into the caudate produced the same amount of inhibition of the 3% NaCl intake as its injection in the MSA. All differences are significant for $p < 0.05$ (unpaired *t*-test).

Arterial Pressure Test

Average basal mean arterial pressure (MAP) did not differ between sodium deplete ($n = 13$) and water deprived ($n = 13$) rats, and it was 116 ± 3 mmHg and 117 ± 2 mmHg, respectively. Peak alterations in MAP obtained with drug injection in the MSA were similar whether the rats were sodium-deplete or water-deprived (Fig. 6). NOR (80, 160 nmol) induced a 20–30% peak increase in basal MAP within the first 5 min after the injection, after which it declined; MAP returned to baseline between 5 to 10 minutes after the injection and thus remained until 20 min after the injection. In sodium deplete rats, CLO (30 nmol) induced a transient 12% peak decrease in basal MAP whithin 10 min after the injection; then, MAP stabilized at a value 5% lower than baseline and thus remained until 20 min after the injection. CLO (20 nmol) did not alter MAP in water deprived rats, although there was a trend of decreasing it. PHE (160 nmol) did not alter MAP in either sodium deplete or water deprived rats. ISO (160 nmol) induced a transient 18% peak decrease in MAP in both sodium deplete and water deprived rats within 1 min after the injection; then, MAP stabilized at a value 5% lower than baseline and thus remained until 20 min after the injection. All differences are significant for $p < 0.05$ (unpaired *t*-test).

DISCUSSION

The results show that noradrenaline injected in the MSA inhibits the 3% NaCl intake of sodium deplete rats. They also show that deprivation-induced and meal-associated water intake are inhibited by noradrenaline injected in the MSA. The inhibitory effect on the 3% NaCl and water intake have some characteristics in common. First, the inhibition is specific for 3% NaCl intake and water, since noradrenaline did not alter food (see also ref. 14) or 10% sucrose intake. Second, the

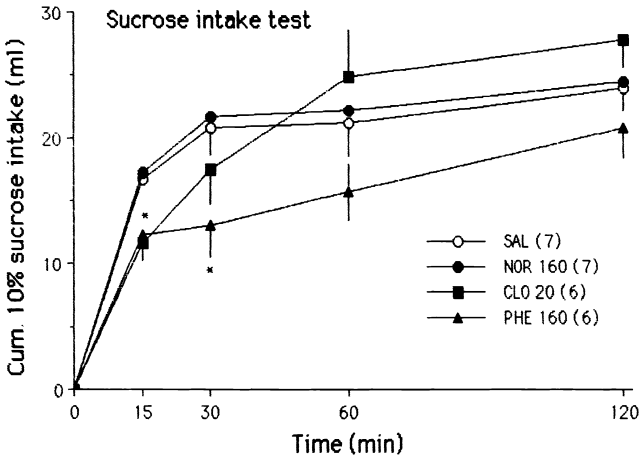

FIG. 4. Cumulative 10% need-free sucrose intake after 0.9% saline (SAL), noradrenaline (NOR, 160 nmol), clonidine (CLO, 20 nmol), or phenylephrine (PHE, 160 nmol) injection in the MSA. Number of animals in parenthesis. *$p < 0.05$ compared to SAL (0.9% saline) injection.

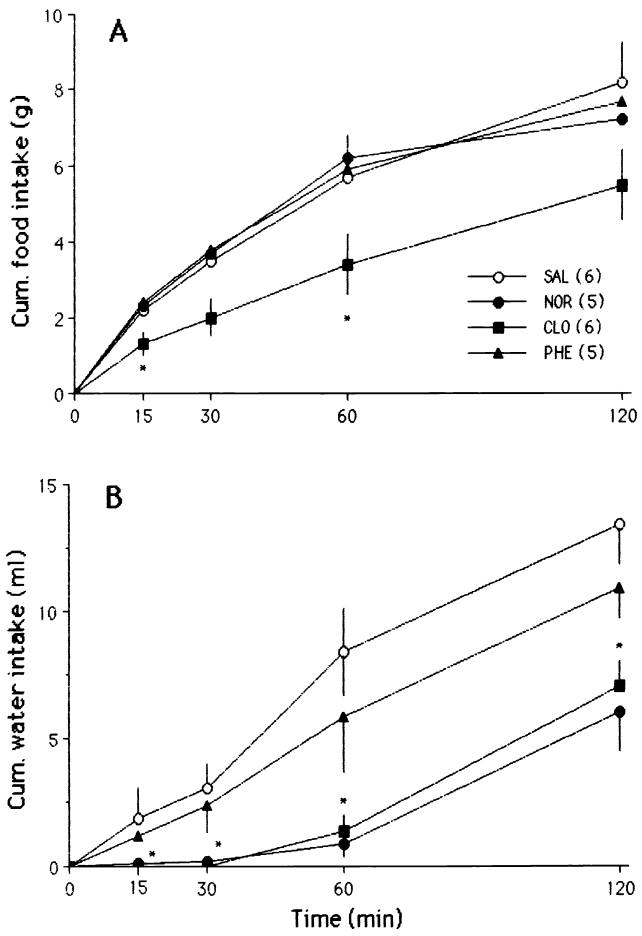

FIG. 5. A) Cumulative food and B) meal-associated water intake after 0.9% saline (SAL), noradrenaline (NOR, 160 nmol), clonidine (CLO, 20 nmol), or phenylephrine (PHE, 160 nmol) injection in the MSA. Number of animals in parenthesis. *$p < 0.05$ compared to SAL (0.9% saline) injection.

TABLE 1

WATER AND 3% NaCl INTAKE OF WATER-DEPRIVED AND OF SODIUM-DEPLETED RATS, RESPECTIVELY, AFTER ADRENERGIC AGONIST OR VEHICLE INJECTION INTO EITHER THE MEDIAL SEPTAL AREA (MSA) OR THE CAUDATE NUCLEUS (CN)

Site of Injection Treatment	Water Intake (ml/h)	3% NaCl Intake (ml/15 min)
MSA injection		
Saline	10.1 ± 1.7 (8)	6.5 ± 0.5 (7)
Noradrenaline	4.2 ± 1.0† (9)	2.7 ± 0.8† (7)
Clonidine	1.3 ± 0.4† (9)	1.5 ± 0.8† (7)
CN injection		
Saline	11.2 ± 1.8 (9)	5.5 ± 0.5 (6)
Noradrenaline	9.7 ± 1.4* (9)	4.6 ± 1.0 (7)
Clonidine	6.3 ± 0.6†* (10)	1.2 ± 0.8† (6)

†$p < 0.05$ compared to saline injected into the same site. *$p < 0.05$ compared to injection in the MSA. Number of animals between parenthesis.

inhibition is likely dependent on alpha-adrenergic receptors because only the alpha agonists, and not the beta agonist isoproterenol, had similar effects to noradrenaline. Third, the inhibition is likely dependent on activation of α_2-adrenergic receptors since clonidine, an α_2 agonist, was much more effective in inducing an inhibition of the intake of both fluids than noradrenaline (mixed agonist) or phenylephrine (α_1 agonist). It is important to recall that α_2-adrenergic receptors are found in the MSA (1,2). Fourth, the inhibitory effect is possibly not dependent on MAP increase induced by noradrenaline,

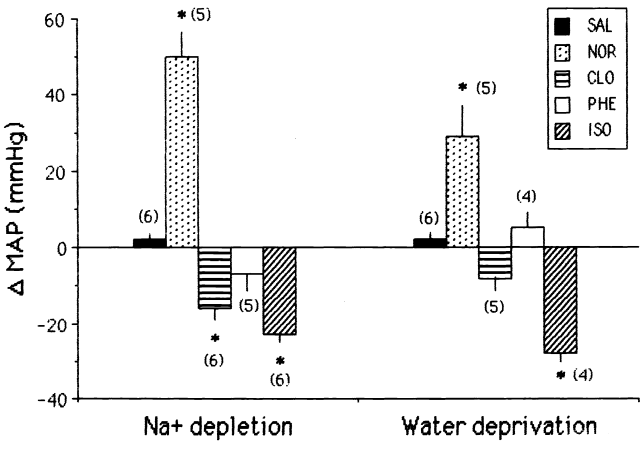

FIG. 6. Peak variations in mean arterial pressure (ΔMAP) from baseline after 0.9% saline (SAL), noradrenaline (NOR, 160 and 80 nmol, sodium deplete and water-deprivation state, respectively), clonidine (CLO, 30 and 20 nmol, sodium deplete and water-deprivation state, respectively), phenylephrine (PHE, 160 nmol) or isoproterenol (ISO, 160 nmol) injection in the MSA. Number of animals in parenthesis. *$p < 0.05$ compared to SAL (0.9% saline) injection.

because 1) MAP is back to baseline several minutes before the time the animals have access to the fluids (20 min after the injection) and 2) clonidine had an opposite effect to noradrenaline (sodium deplete state) or no effect (water deprived state) on MAP. In addition, injection of carbachol in the MSA induces a pressor effect that is very intense in sodium deplete rats, but it does not inhibit the need-induced 3% NaCl intake (unpublished results).

The inhibitory effect of noradrenaline seems to contradict the view that this neurotransmitter is important for the induction of water and NaCl solution intake (3,4). The hypothesis that noradrenaline has a dual role, inhibitory and excitatory, in the control of hydromineral fluid intake (5,9) helps to conciliate these contradictory findings. This could be a reason for the need of much higher doses of NOR than CLO to produce an inhibition, i.e., whereas NOR acts on excitatory and on inhibitory paths, CLO would act only on inhibitory ones. In addition, the differences between the present and previous results (3,4) may lie in the 1) amount of NOR injected, which is higher in the present case and probably must be so in order to overcome its excitatory role, and 2) situation in which the animal is ingesting fluids; in the present work, NOR was injected when an ongoing mechanism of fluid intake was working, whereas previously it was injected in normovolemic animals (3) or in animals of which an excitatory mechanism dependent on NOR had been disrupted by pharmacological treatments (4).

Some possibilities to be tested could explain how alpha-adrenergic agonist injection into the brain inhibits hydromineral fluid intake. First, the agonists could act on pre-synaptic alpha-2 receptors that regulate the synaptic release of NOR, thereby inhibiting further release of endogenous noradrenaline that is important for the production of fluid intake. This would explain the effect of injections of CLO and PHE, the latter acting non-specifically on pre-synaptic alpha-2 receptors. However, in order to this hypothesis be valid for NOR one has to assume that exogenous NOR has access to only pre-synaptic receptors, which is unlikely true. Thus, a second possibility is that there are separate systems in the brain that utilize noradrenaline in two different pathways, one that activates and another utilizing α_2-receptors that inactivates the behavior. The inhibition observed in our experiments would result from each agonist acting predominantly on this second pathway. In this regard, it is interesting to recall that there are cells in the subfornical organ, an area of the brain that is important for water and salt intake, that respond with either excitation or inhibition to alpha-2 adrenergic activation (17). Cells like these ones are candidates to subserve those putative separate pathways.

Clonidine and phenylephrine inhibited the need-induced 3% NaCl intake, but only clonidine inhibited deprivation-induced and meal-associated water intake. Clonidine induced 90–100% inhibition of water and 3% NaCl intake during the first hour of tests at doses five to eight times smaller than the dose of phenylephrine that produced 50% inhibition of 3% NaCl intake during only 30 min of the sodium appetite test. Clonidine also inhibited need-induced food intake, thus challenging the previous view (7,8) that its inhibitory effect is exclusive for water and NaCl intake. However, one has to consider that the inhibition does not follow the same profile for every commodity tested. For instance, meal-associated water intake (Fig. 5B) was completely inhibited at 15 and 30 min whereas food intake (Fig. 5A) was 50% inhibited at 15 min and was not different from the control at 30 min. At 60 min, meal associated water intake was 80% inhibited and food intake 40%. The former was still 50% inhibited whereas the latter was not different from the control at 120 min. Previous (7,8) measurements of need-induced food intake began 60 min after the food became available to the animals. This methodological difference, plus animal variability, may have contributed to the absence of the effect of clonidine on food intake in our previous works. A slight 30% inhibition of 10% sucrose intake also occurred only at 15 min; this inhibition was of shorter duration than the inhibition induced by phenylephrine and suggests that the two alpha agonists may be inhibiting the same pathways. Clonidine is reported to have sedative effects (11) and this may have contributed partially to the inhibitions observed. However, sedation is not enough to explain the strong and more persistent preferential inhibition of 3% NaCl and water intake. If we discount the percent inhibition that occurred on food and 10% sucrose intake, we would still have a more potent inhibition of 3% NaCl and water intake with clonidine than with noradrenaline or phenylephrine. In addition, rats receiving a sedative dose of diazepam (2 to 4 mg/kg) that induces at least one hour-lasting prostration, drink NaCl solution as much as controls do, or even more, in need-free protocols (6). In accordance to this discussion, an activation of a general inhibitory system of ingestive behavior or a motor impairment also do not explain the preferential inhibition of 3% NaCl and water intake compared to food and sucrose solution intakes. Nausea or another sensation of malaise is also an unlikely explanation for the preferential inhibition induced by clonidine if we consider that a typical aversive compound such as lithium chloride inhibits food, but not water intake (20). Finally, alterations in arterial pressure are not important for the inhibitory effects of clonidine, since it either did not alter MAP in water-deprived rats, or it induced a fall in MAP in sodium deplete rats similar to that induced by the beta-agonist isoproterenol, and isoproterenol did not alter fluid intake.

The failure to produce alteration on fluid intake by NOR injection into the caudate suggests that the effects observed by NOR injection in the MSA are not simply a result of a generalized spread of this agonist in the brain. The much stronger inhibitory effect of CLO on water intake when injected in the MSA versus its injection into the caudate (98% vs 43%) favors this conclusion. The same inhibitory effect of CLO injected in the MSA or into the caudate on the need-induced 3% NaCl intake suggests a ubiquitous distribution of the receptors involved with this inhibition. Similar result for water intake comparing the caudate with the lateral hypothalamus was obtained in a previous work (8).

In short, our results show that noradrenaline injected in the MSA has a specific inhibitory effect on water and 3% NaCl intake, possibly dependent on the activation of α_2 receptors. This specificity directed to fluids related to hydromineral balance suggests that central noradrenaline is important for an inhibitory control of hydromineral fluid intake.

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