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Effects of Clonidine and Guanfacine on Drinking and Ambulation in Spontaneously Hypertensive Rats

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TOGASHI, H., M. MINAMI, Y. BANDO, Y. KOIKE, K. SHIMAMURA AND H. SAITO. *Effects of clonidine and guanfacine on drinking and ambulation in spontaneously hypertensive rats.* PHARMAC. BIOCHEM. BEHAV. 17(3) 519–522, 1982.—Present experiment was undertaken to compare the effects of clonidine and guanfacine on water drinking behavior and ambulatory activity in spontaneously hypertensive rats (SHR). Equipotent hypotensive doses of clonidine and guanfacine, 150 μg/kg and 1500 μg/kg, respectively, given twice a day at 8:00 and 20:00, produced a triphasic pattern of behavioral changes; initial increase in water drinking and ambulation during the light period, decrease in water intake and ambulation at the beginning of the dark period, and a second increase in water drinking and ambulation at the end of the dark period. Guanfacine treated SHR showed less change in water drinking behavior and ambulation than the clonidine treated SHR.

Clonidine Guanfacine Water drinking Spontaneously hypertensive rats Ambulatory activity

GUANFACINE, a representative member of centrally acting antihypertensive agents, resembles clonidine in most of its basic pharmacological properties [6]. Although some differences have been reported [3,7], no reports are available in the literature concerning the changes in water drinking behavior after clonidine and guanfacine administration in man and animals. Recently, Kuribara *et al.* [4] and Tadokoro *et al.* [8] developed a sensitive method for simultaneous determination of ambulatory activity and water drinking in rats. Using this equipment, the present experiment was undertaken to compare the effects of clonidine and guanfacine on the rhythms of water drinking behavior and ambulatory activity in spontaneously hypertensive rats (SHR).

METHOD

Animals

Twenty week old male SHR of Wistar Okamoto strain were used. Experiment was done under a 12 hr light and dark period (light-dark cycle). The light period began at 8:00 and ended at 20:00. Illumination was provided by fluorescent light. Room temperature was maintained at 20±2°C throughout the experiment. Rats were fed a regular diet.

Drugs

After the control period in which water was administered PO twice daily, SHR were divided into two groups: the clonidine (Catapres, C. H. Boehringer Sohn) administered group and the guanfacine (Estulic, Sandoz) administered group. It has been reported that guanfacine is approximately one-tenth as effective as clonidine in lowering blood pressure. For this reason, we used oral doses, 150 μg/kg of clonidine and 1500 μg/kg of guanfacine, in this experiment. Drugs were given by stomach tube at 8:00 and at 20:00. Administration of water and drugs was done at a volume of 5 ml/kg.

Measurement of Blood Pressure

The left carotid artery was cannulated for recording blood pressure. Tubing (PE 20), filled with 0.9% saline that contained 0.1% heparin, was run under the skin and out through the back of the neck. The end of the catheter was occluded with a needle. Patency of the catheter was maintained by flushing with 0.2 ml of heparinized saline twice daily. Carotid blood pressure was recorded with a pressure transducer (Nihon Kohden MUP-0.5A) and a pen recorder. Two days after cannulation, a single oral dose of clonidine 150 μg/kg or guanfacine 1500 μg/kg was administered to the respective groups. Before drug administration and every 3 hours thereafter, systolic and diastolic blood pressure was recorded.

Water Drinking Behavior and Ambulatory Activity

The automatic Ambulo-Drinkometer (Ohara & Co. Ltd., Tokyo), developed by Tadokoro *et al.* [8], was used to determine ambulatory activity and water drinking. This apparatus is composed of ten steel cages (38 (D) × 25 (W) × 19 (H)

cm), which are equipped with microswitches that are activated by tilting of the cage floor as the rats move around. A drinking spout is connected to a water tank via an infusion machine, made of acrylfiber and stainless steel tubes, which derivers 0.05 ml drops. The water falls drop by drop, as the rat drinks. There is a weak electrical current in the upper and lower parts of the infusion machine. Short circuits generated by individual drops are amplified to activate an electromagnetic counter. Using this apparatus, we could determine exactly the amount and rate of water drinking. Two weeks before the experiment five rats of each group were put into this apparatus. During the control period, water was administered twice each day for 5 days. The drugs were administered for the following ten days. Ambulatory activity and water drinking were calculated as the mean of the total counts of the 5 rats during 3 hours. Ambulatory activity and water drinking in the drug administration period were compared with those in the control period.

Urine Volume

Before drug administration and 2 days thereafter, urine volume was measured every 3 hours during the dark period and once during the light period. In order to collect urine samples every 3 hours during the dark period, light-dark alternation cycle was reversed (dark-light cycle). At this time, the light period began at 8:00 and ended at 20:00.

RESULTS

Arterial Blood Pressure

Single oral administration of 150 μg/kg clonidine produced the same degree of hypotension as 1500 μg/kg guanfacine in SHR. Namely, at these doses, clonidine and guanfacine produced a significant decrease in systolic and diastolic blood pressure by approximately 30 mmHg during 3 to 12 hours after administration (Fig. 1).

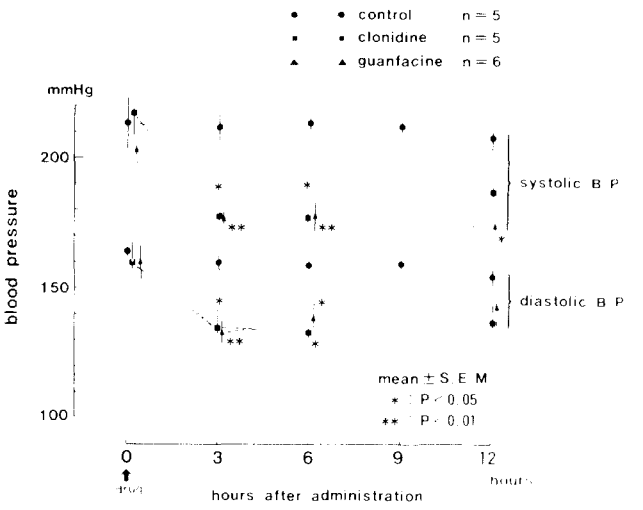

FIG. 1. Effects of clonidine and guanfacine on blood pressure in SHR. The three groups are indicated by the following symbols: filled circles, age-matched control group (n=5); filled squares, clonidine administered group (n=5); filled triangles, guanfacine administered group (n=6). Each value shows mean±S.E.M. Blood pressure was determined before a single oral dose of 150 μg/kg clonidine or 1500 μg/kg guanfacine, and then 3 hours, 6 hours and 12 hours thereafter. Statistical analysis was done by Student's *t*-test. Ordinate: systolic and diastolic blood pressures. Abscissa: time in hours after the drug administration.

Water Drinking and Ambulatory Activity

It was demonstrated that the 24 hour patterns of drinking behavior and ambulatory activity in SHR were well synchronized with the light-dark cycle. The pattern of these two parameters remained nearly constant from day to day. Namely, the activity count of drinking behavior and ambulation in the dark period was higher than those in the light period. Further, the drinking and ambulatory activity patterns showed two high peaks in the dark period.

During the 3 hour period after clonidine administration in the dark period, a marked decrease in water drinking as compared with that in the control period was demonstrated. On the contrary, the water intake at the end of the dark period and the light period was remarkably elevated as compared with the control period. Not only did the water drinking counts decrease, but the counts of ambulatory activity were also markedly depressed during the 3 hour period after clonidine administration in the dark period. Then, the ambulatory activity counts surpassed those of the control. During the light period, an increase of ambulatory activity counts was demonstrated during drug treated period (Fig. 2). On the other hand, after administration, guanfacine produced a slight decrease in water drinking and ambulatory activity at the beginning of the dark period. Although the behavioral effects of guanfacine at the end of the dark period and in the light period resembled those of clonidine, guanfacine treated SHR showed significantly less increase in water drinking behavior and ambulatory activity as compared with the clonidine treated group (Fig. 3).

Urine Volume

Clonidine produced a marked increase in urine volume during the 3 hour period, after the administration in the dark period. During the following 3 hour period, there was a significant decrease in urine volume. Guanfacine, however, did not produce any changes in urine volume (Fig. 4).

DISCUSSION

The most interesting observation in the present experiment using equipotent hypotensive doses of clonidine and guanfacine is that administration of clonidine twice daily produced a triphasic pattern of behavioral changes. An initial increase in water drinking and ambulation during the light period followed by a decrease in water intake and ambulation at the beginning of the dark period and then a second increase in water drinking and ambulation at the end of the dark period were observed. Although the behavioral effects of guanfacine resembled those of clonidine, guanfacine had a less potent effect on the water drinking behavior and ambulatory activity than those observed in the clonidine treated group. With regard to the mechanism of increase in water drinking behavior induced by clonidine two factors have been considered: the inhibition of salivation [9] and the inhibition of ADH secretion [5]. In this experiment, we did not study the direct action of clonidine or guanfacine on salivary flow. However, decrease in salivary flow has been observed in patients one hour after clonidine administration [1]. Accordingly, it may be possible that increase in water drinking in SHR was resulted from the clonidine induced decrease in

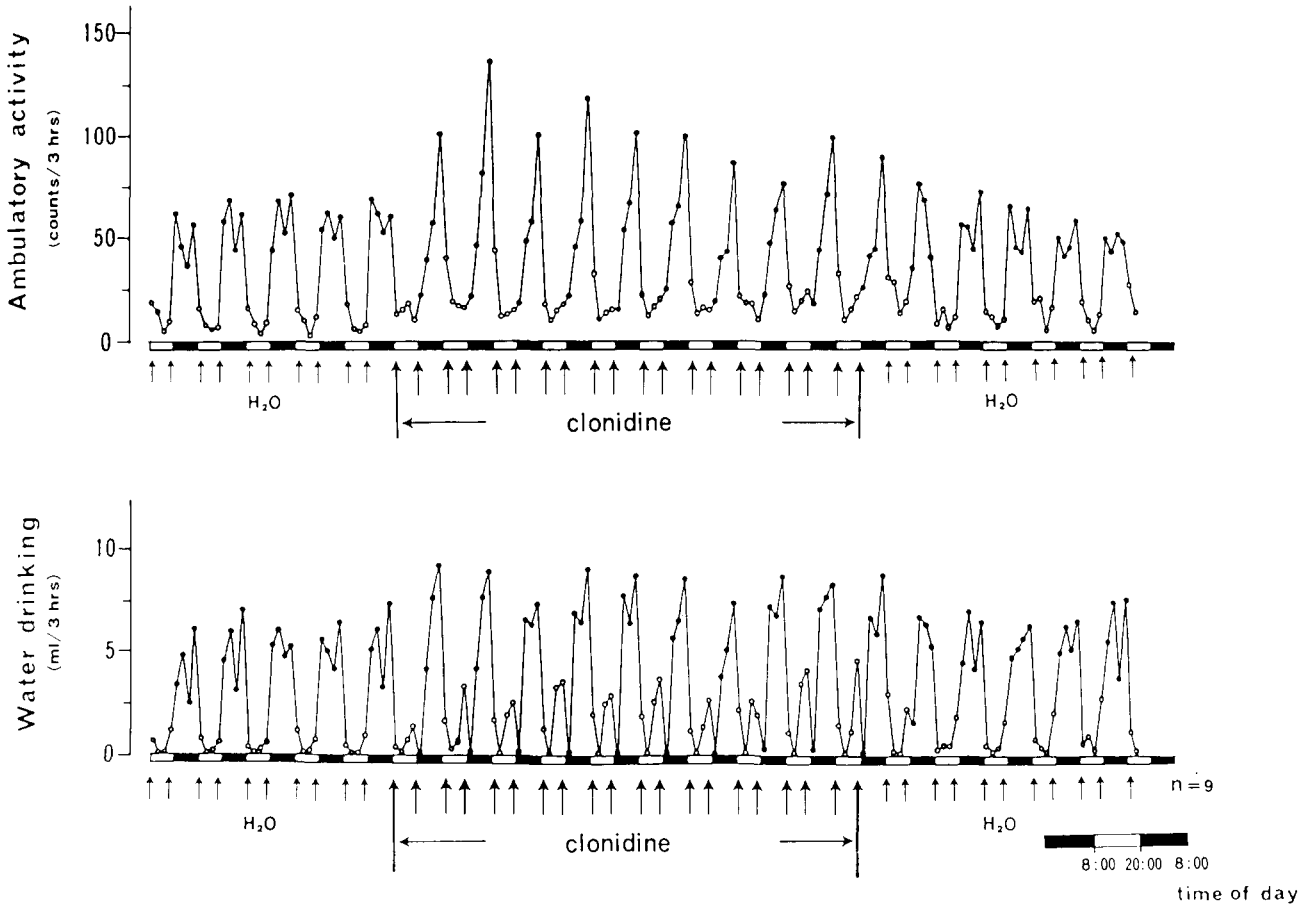

FIG. 2. Effects of clonidine on ambulatory activity and drinking behavior in SHR. In the control period, water was administered for 5 days. During the drug administration period, clonidine at a dose of 150 μg/kg was given twice daily for 10 days. Ambulation and drinking values were calculated as mean of total counts of 9 rats during 3 hour periods. Ambulation and water drinking activity during the light period were expressed by open circles, and during the dark period by filled circles. Small arrows indicate water administration and large arrows indicate clonidine administration.

salivary flow. It has been observed that guanfacine produces dry mouth in patients with essential hypertension [2]. The guanfacine induced increase in water intake may possibly be explained by the dryness of mouth associated with the decrease in salivary flow. Hence, it is assumed that the decrease in salivary flow induced by clonidine and guanfacine is at least in part associated with the increase in water drinking in SHR. In our experiments, it was observed that during the 3 hour period after clonidine administration in the dark period, urine volume increased about five fold. Thereafter it decreased significantly as compared with that in the control period. On the other hand, increase in drinking was observed 9 to 12 hours after clonidine administration. Decrease in humoral volume induced by diuresis also appears to be responsible for the increase in water intake after clonidine administration. In this experiment, guanfacine did not alter the urine volume. Therefore, it is impossible to speculate that the decrease in humoral volume induced by guanfacine is associated with increase in water intake after guanfacine administration. With regard to the decrease in water intake at the beginning of the dark period, drinking behavior which may be increased by clonidine or guanfacine may be masked by the sedative effect just after the drug administration.

Different degrees of behavioral changes in water drinking and ambulatory activity between clonidine and guanfacine were demonstrated in this experiment. The significance of these differences requires further laboratory confirmation and explanation.

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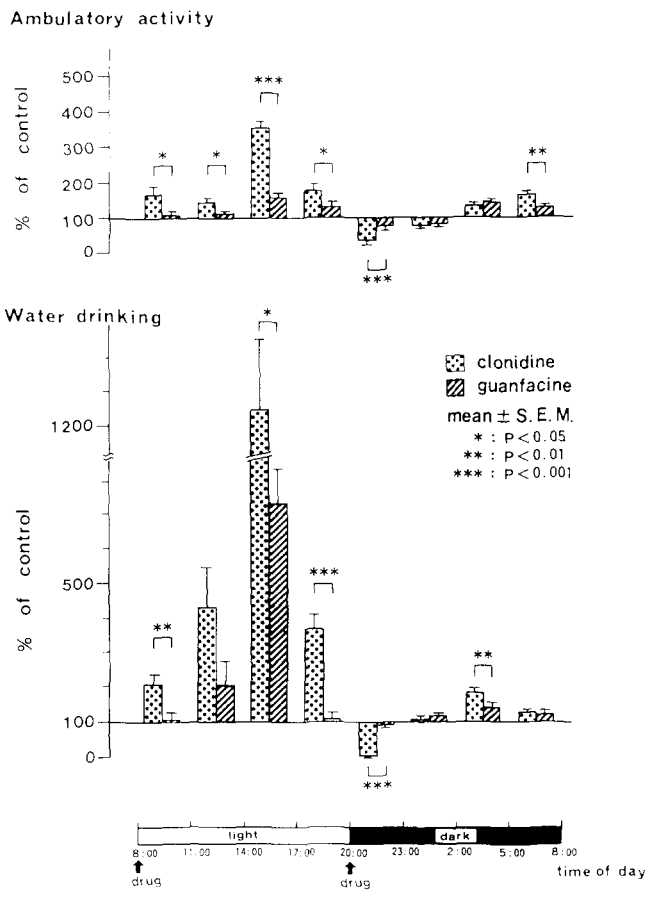

FIG. 3 Time course % changes in ambulation and drinking during clonidine and guanfacine administration period. Control value was expressed as mean of 5 day-summation of 3 hour periods during pretreatment. Counts of ambulation and drinking during clonidine and guanfacine administration period were shown as % changes of each control value during 3 hours. The % value represents a 10 day period. The 10 day-summation of % values between the group treated with 150 μg/kg of clonidine twice daily and the group treated with 1500 μg/kg of guanfacine twice daily were compared. Hatched columns show clonidine treated group. Dotted columns indicate guanfacine treated group. Statistical analysis was done by Student's *t*-test. Ordinate: 10 day-summation of % changes of control value in ambulatory activity and water drinking. Abscissa: time in hours.

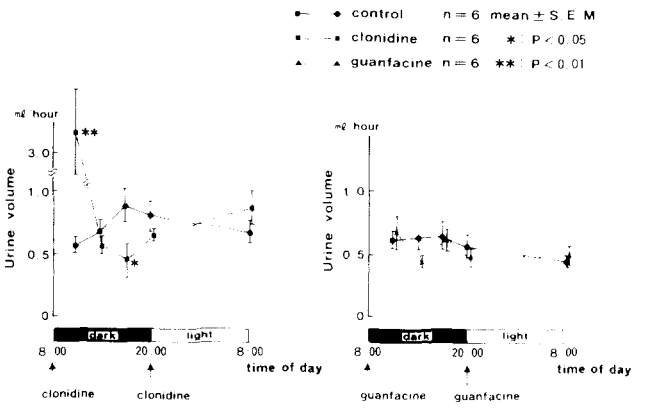

FIG. 4. Effects of clonidine and guanfacine on urine volume in SHR. Before administration of 150 μg/kg clonidine and 1500 μg/kg guanfacine twice daily and two days thereafter, urine samples of 6 rats in each group were collected every 3 hours during the dark period and once during the light period. Urine volume of each sample was measured. The pretreatment period values were expressed as control. Filled circles, filled squares and filled triangles indicate urine volume before administration and after clonidine or guanfacine administration, respectively. Each value shows mean ± S.E.M. Statistical analysis was done by Student's *t*-test. Ordinate: urine volume (ml/hr). Abscissa: time in hours.

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