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Effects of Sodium Nitrite on DRL Performance in the Rat[1]

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VIVEIROS, D. M. AND L. M. TONDAT. *Effects of sodium nitrite on DRL performance in the rat.* PHARMAC. BIOCHEM. BEHAV. 8(2) 125–127, 1978. – Fifteen Sprague-Dawley rats were administered sodium nitrite 0.10% or 0.15% solutions in their drinking water from the age of 45 days to the end of the study. Six control rats received only tap water. At 80 days of age, rats were trained to bar press for food pellets on a CRF schedule. After reaching criterion performance the rats were switched to a DRL-20 for a period of 6 days to test for response inhibition, which was measured as a ratio of responses to reinforcements. Results indicated no significant differences between groups for response inhibition. All groups showed significant increases in learning as reflected by a decrease in ratios and an increase in total reinforcements over days. However, sodium nitrite rats compared to controls obtained significantly fewer reinforcements over sessions and a greater number of no responding periods (time-outs). One interpretation discussed was the possibility that sodium nitrite produced an increase in responding to distractible (non-task related) cues.

Sodium nitrite Learning DRL performance

SODIUM nitrite is added to processed foods (e.g., ham, hot dogs) as a preservative to prevent the growth of *Clostridium Botulinum* [4], and as a flavor and color enhancer [8]. Nitrites are also naturally reduced from nitrates found in vegetables and water [12]. Recently, a number of studies have reported detrimental physiological and behavioral effects [1, 3, 10, 13].

Physiologically, sodium nitrite is an agent in the formation of nitrosamines which have been found to cause cancerous tumors in rats and numerous other species [11]. Sodium nitrite has also been shown to react with iron in hemoglobin to form methemoglobin, thereby preventing oxygen transport to body tissues. Levels of 70–80% methemoglobin are fatal [11], and this state has been suspected to be the cause of infantile cyanosis [7]. Further, Shuval and Gruener [14] state that nitrites passing through the placenta are responsible for the appearance of methemoglobin in fetal rats. Behroozi and his associates [3] found that the increase in methemoglobin levels in rats given sodium nitrite for two months was accompanied by permanent changes in EEG patterning and a sluggish appearance as compared to controls. In a study by Shimizu [13], the effect of sodium nitrite on rat brain tissue was reported to both activate and inhibit monoamine oxidase activity, depending on the substrate used.

Behaviorally, sodium nitrite has produced reversible increases in aggression [10] and decreases in motor activity in mice [2]. To date no other behavioral changes attributed to sodium nitrite have been reported. This research was designed to test learning performance in rats treated with sodium nitrite. The DRL schedule was selected to test not only learning ability, but also to assess the possible effect of sodium nitrite on the inhibition of active goal seeking responses (response inhibition) [5, 6, 9].

METHOD

Animals

Twenty-one 40 day old Sprague-Dawley rats of CD strain were obtained from the Charles River Laboratory. All animals were given free access to Purina Laboratory Chow and tap water, modifications for treatment groups are indicated below. Rats were housed separately in cages in a room with constant temperature and lighting conditions.

Apparatus

Animals were trained in a Ralph Gerbrands operant conditioning apparatus (20.5 × 21.8 × 23.0 cm, single lever) enclosed in an environmental chamber. The apparatus was modified with an interval timer and counters to implement a DRL schedule and measurement of performance.

Procedure

Following 5 days of monitoring body weights and water intakes, animals were matched according to body weight, and then assigned to 3 groups. Groups 1 and 2 (experi-

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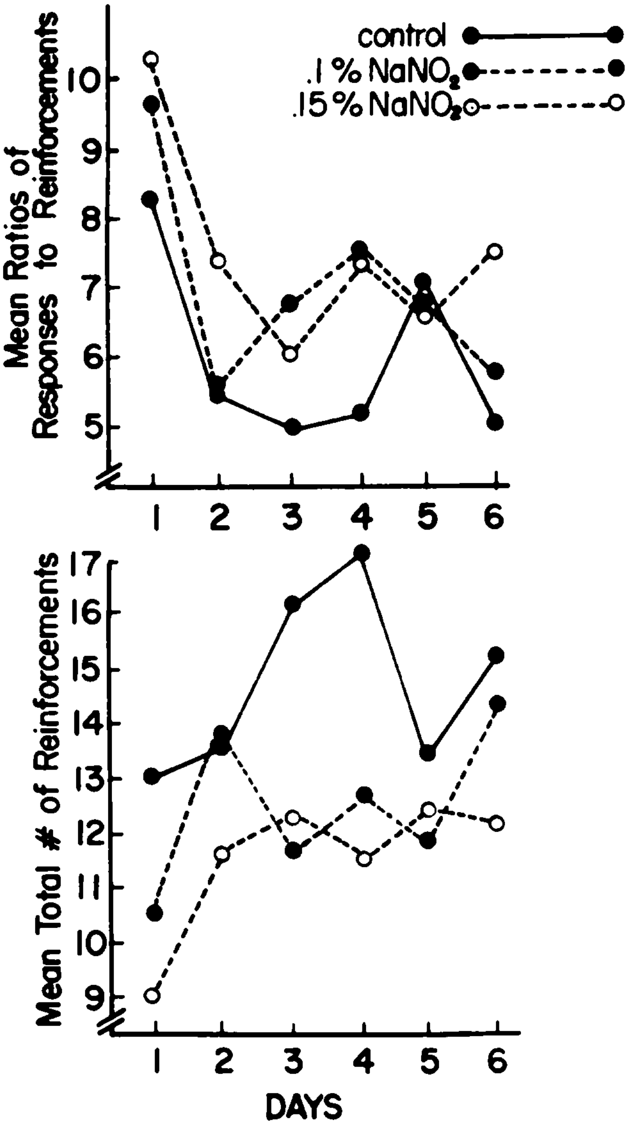

FIG. 1. (Top) Mean ratios of responses to reinforcements for all groups across days. (Bottom) Mean total number of reinforcements for all groups across days.

mental groups, n = 8,7) were maintained on 0.1% and 0.15% sodium nitrite solutions, respectively, in their drinking water. Group 3 (control, n = 6) was maintained on regular tap water. At 70 days of age all rats were adapted to a 23 hr food deprivation schedule for approximately 10 days. Animals were then trained to bar press for 45 mg food pellets in 15 min daily sessions. A continuous reinforcement schedule was maintained until subjects had reached the criterion of 120 presses per session for 2 consecutive days. Rats were then switched to a DRL-20 schedule 15 min daily for 6 days. The ratio of responses to reinforcements as well as total reinforcements per session were recorded for each rat. Body weights and water intakes continued to be monitored daily throughout the study. Behavioral observations were also noted throughout test sessions.

RESULTS

Separate one-way analyses of variance (3 levels, unequal n) [15] conducted on final CRF performance, and water intakes and body weights (prior to deprivation and at end of study) revealed no significant differences between treatment groups ($ps > 0.05$).

Figure 1 (top) depicts mean ratios of responses to reinforcements for the treatment groups. A 3 x 6 (Groups x Days, unequal n) analysis of variance on these data indicated no differences between groups for the ratio of responses to reinforcements ($ps > 0.25$). Ratios were found to decrease significantly over days ($p < 0.01$), with a post hoc analysis [15] revealing the differences to exist between Day 1 and Day 2 ($p < 0.05$) and to have stabilized by Day 5 and Day 6 ($p > 0.25$). The Day by Group interaction was not significant ($p > 0.25$).

Figure 1 (bottom) shows mean total reinforcements for treatment groups over days. Here differences following an analysis of variance (same analysis as above) were found between groups for total number of reinforcements ($p < 0.05$). A post hoc analysis indicated that 0.1% sodium nitrite rats ($\bar{X}$ = 12.3) obtained significantly fewer reinforcements than control rats ($\bar{X}$ = 14.7; $p < 0.05$) and no differences existed between the 0.1% and 0.15% ($\bar{X}$ = 11.3) sodium nitrite groups ($p > 0.25$). A significant Day effect was found ($p < 0.05$) demonstrating an increase in number of reinforcements obtained over days for all groups. No Group by Day interaction was found ($p > 0.25$).

Recorded observation of behavior during DRL testing sessions suggested that sodium nitrite rats made more of the nonreinforced responses immediately after receiving a reinforcement. In general, nitrite rats persisted in pressing five or six times following reinforcement, left the bar and did not return until long after the appropriate IRT had passed. During this time away from the bar general exploratory behavior was noted (i.e., general activity was maintained). By contrast, it was observed that the controls made more of the unreinforced responses toward the end of the appropriate IRT, i.e., pressing after 18 or 19 seconds had passed. These general differences in observed behavioral patterns were reflected by a statistical difference in the total number of time-outs (i.e., minute durations of no responding) over the 6 day session (control total = 8; 0.10% nitrite = 18; 0.15% nitrite = 38; χ^2 = 21.9, df = 2, $p < 0.005$).

DISCUSSION

The results of this study indicated no differences in response inhibition between rats receiving sodium nitrite in their drinking water, as measured by the ratio of responses to reinforcements. The results also revealed that increases in learning were not affected by sodium nitrite on this schedule. All rats became more proficient with the schedule as shown by decreases in ratios of responses to reinforcements and increases in total reinforcements obtained over days. However, there was a significant difference between groups for total number of reinforcements, indicating that sodium nitrite rats received fewer reinforcements compared to controls.

This latter finding suggests that nitrite rats were less persistant at the task than control rats. Other studies have reported decreases in motor activity following sodium nitrite administrations [2,3]. However, it is not believed that a decrease in motor activity for nitrite rats would explain the differences in the number of reinforcements obtained during the DRL task, because of their CRF performance. During CRF training no differences in performance were found, i.e., both nitrite rats and controls reached criterion with similar performance rates. Perhaps a better explanation for these data is that sodium nitrite rats had a lower threshold for responding to other environmental or non task-related cues. This is supported by the significantly greater number of time-out periods for nitrite rats during which these rats continually reexplored the operant chamber. Further research specifically designed to test sodium nitrite's effect on distractibility is necessary to substantiate this view.

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