

## Ethanol as a Reinforcer for Rats: Effects of Concurrent Access to Water and Alternate Positions of Water and Ethanol

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*Abstract.* Water and ethanol solutions were concurrently made available on a continuous reinforcement schedule to 4 food-deprived male albino rats during daily 1-hr sessions in an operant conditioning chamber equipped with 2 levers and 2 liquid dippers. The number of ethanol reinforcements substantially exceeded the number of water reinforcements for each rat at each concentration studied (8, 16, and

32% w/v). Water reinforcements were low in number and did not vary with ethanol concentration. As the ethanol concentration was increased, the number of ethanol reinforcements obtained decreased, while the quantity consumed (mg/100 g of body weight/hr) increased. The highest rate of responding occurred at the beginning of the session.

*Key words:* Ethanol – Ethanol Drinking – Water-Ethanol Choice – Concurrent Schedules – Ethanol Concentration – Ethanol Reinforcement – Rats.

Intake of an ethanol solution may be due to its liquid character and not due to the presence of ethanol. To control for this possibility most animal studies of ethanol drinking have employed two drinking bottles—one containing water and the other containing an ethanol solution (see Myers and Veale, 1972; Wallgren and Barry, 1970, for recent reviews). Concurrent access to water, when combined with systematic changes in the relative positions of the different liquids, permits an assessment of the portion of ethanol consumption that may be attributed to nonspecific liquid intake.

Myers has made food, water, and ethanol concurrently available to rats, with the presentation of each being contingent upon a lever press (Myers and Carey, 1961; Myers, 1961a, b). The rats were both food and liquid deprived. With this procedure he has studied the effects of a number of factors, including ethanol concentration, on responding for each of the 3 substances. When the concentrations were presented in an ascending order, ethanol responses exceeded those for water until the ethanol concentration reached 9% (v/v); whereas when the concentrations were presented in a descending order, ethanol responses did not exceed those for water until the concentration was decreased to 4% (v/v) (Myers and Carey, 1961). The procedure used in the present study is similar to Myers', in that water and ethanol were

concurrently available to food-deprived rats contingent upon lever pressing.

The purpose of this study was to investigate the effects of concurrent access to water on ethanol intake. Additionally, the quantity (mg) of ethanol consumed and the time course of intake were measured. The influence of side preference was evaluated by varying the location of water and ethanol. To simplify the experimental analysis, the rats were not deprived of liquid, and food pellets were not available in the operant chamber.

### *Method*

*Subjects.* Four naive male albino Sprague-Dawley rats, about 300 days old at the beginning of the experiment, were individually housed in a constantly illuminated room with the temperature maintained at 24°C. At 80% of their free-feeding weights, at which they were maintained, 3 rats weighed 477 g (Z-7), 450 g (Z-6), and 381 g (Z-5). One obese rat, Z-9, was maintained at 403 g which was 65% of its free-feeding weight. Water was always available in the rats' home cages, except for 5 days during training (see *Procedure* below).

*Apparatus.* A sound-attenuated commercial operant conditioning chamber (LVE, # 1414) was equipped with 2 levers and 2 solenoid-driven liquid dippers (LVE, # 1351). The levers and liquid dippers were symmetrically centered on the front panel with the dippers placed lateral to the levers. Three colored jewel lights located above each lever provided general illumination. A 4.76-W white light was located 3.2 cm above the hole in the panel where the dipper cup was located when

Table 1. Sequence of training and experimental phases

Phases	Home cage		Operant chamber				
	food	water	left side	right side	food	session duration (hrs)	number of sessions
Establishment of lever pressing for water	— <sup>a</sup>	— <sup>b</sup>	— <sup>c</sup>	VI 0%	+ <sup>d</sup>	6	1
	—	—	—	shaping 0%	+	6	1—2
	—	—	—	0%	+	6	3
Establishment of ethanol as a reinforcer	—	+ <sup>e</sup>	—	0%	+	6	5
	—	+	—	2%	+	6	2
	—	+	—	4%	+	6	3
	—	+	—	8%	+	6	4
	+ <sup>d</sup>	+	—	8%	—	6	≥ 5
Establishment of concurrent performance	+	+	8%	—	— <sup>a</sup>	6	≥ 5
	+	+	alternation of 8% and E <sup>f</sup>	—	—	6	≥ 10
	+	+	alternation of 8% and E	—	—	1	≥ 10
Experimental phase	+	+	alternation of 8% and 0%	—	—	1	≥ 10
	+	+	alternation of 16% and 0%	—	—	1	≥ 10
	+	+	alternation of 32% and 0%	—	—	1	≥ 10
	+	+	alternation of 8% and 0%	—	—	1	≥ 10

<sup>a</sup> Absence of food.

<sup>b</sup> Absence of water.

<sup>c</sup> No consequences followed responses.

<sup>d</sup> Sufficient food was provided to maintain rats at 80% of their free-feeding weights.

<sup>e</sup> Free access to water.

<sup>f</sup> Presentations of an empty dipper were produced by lever presses on the side not containing ethanol.

in the up-available position. Each lever press produced 4-sec access to a dipper cup containing 0.12 ml of liquid. Simultaneous with the 4-sec dipper cup presentation was the sounding of a Sonalert and the illumination of the light above the dipper-panel opening. Liquid was contained in partially covered reservoirs to minimize evaporation. White masking noise was constantly present, and an exhaust fan provided ventilation.

Programming and data recording were automatically controlled by standard electromechanical equipment in an adjacent room. The temporal pattern of the responses and reinforcements was continuously recorded by a cumulative recorder and a counter which printed out every 2 min.

The ethanol concentrations, expressed in grams percent (w/v), were prepared using 95% (v/v) ethanol in tap water. For example, the 8% solution was made by adding 10.6 ml of ethanol to a volumetric flask with sufficient tap water to make a total volume of 100 ml. The solutions were prepared at least 20 hrs before use and were kept in stoppered flasks at room temperature. The volume consumed was measured at the end of each session by subtracting the volume remaining from the volume added to the reservoir corrected for evaporation. Evaporation corrections were determined for each concentration by measuring the volume lost from a second reservoir which, during experimental sessions, was placed adjacent to the reservoir used with the liquid dipper.

*Procedure.* Table 1 summarizes the phases of the procedure for establishing concurrent performance maintained by water and ethanol-reinforced lever pressing.

*Establishment of Lever Pressing for Water.* Water bottles were removed from the rats' home cages, and to increase further the probability of drinking, the daily feedings of Purina

Laboratory Chow were placed in a wire food hopper in the operant chamber. During the first daily 6-hr session, the right-hand dipper was presented at a mean rate of once a min, regardless of the rat's behavior, with a variable interval between presentations (1-min variable-interval schedule). When the rats reliably drank from the dipper, the variable-interval water presentations were discontinued, and the presentation of water was used to shape depression of the right-hand lever. Each lever press was reinforced on a continuous reinforcement schedule. After the rats learned to lever press, 3 more sessions were conducted before the water bottles were restored to the home cages.

*Establishment of Ethanol as a Reinforcer.* In-session feedings of Purina Laboratory Chow continued for a series of 14 daily 6-hr sessions. During the first 5 sessions water was the available liquid, then 2% ethanol for 2 sessions, 4% ethanol for 3 sessions, and 8% ethanol for 4 sessions. The in-session feedings were then discontinued, and the food was given to the rats only in their home cages following each session. Sessions with 8% ethanol available at the right-hand dipper continued until 5 consecutive stable days of ethanol-reinforced lever pressing occurred. During this phase responses on the left-hand lever had no consequence.

*Establishment of Concurrent Performance.* After 8% ethanol was established as a reinforcer at the right dipper, availability of 8% ethanol was shifted to the left dipper until the rats emitted an approximately equal number of responses for ethanol in a given session at the left lever as they had in previous sessions at the right lever. Thereafter, 8% ethanol availability was shifted daily between the 2 dippers. Responding on the side opposite ethanol produced presentations of an empty dipper. After 10 consecutive sessions of stable ethanol-

reinforced lever pressing (5 sessions on the left and 5 on the right), the session duration was reduced from 6 hrs to 1 hr.

**Experimental Phase.** After 10 consecutive 1-hr sessions of lever pressing for ethanol, concurrent water availability was introduced at the dipper opposite to the one containing ethanol. Intake of ethanol concentrations of 8, 16, 32, and 8% (retest), in that order, was studied under conditions of concurrent access to water. Changes from one concentration to the next were made after 10 sessions, given that there was no consistent upward or downward trend in the number of reinforcements. The positions of water and ethanol were systematically reversed from session to session except for the 8% retest series, where 8% was held on the right for 2 consecutive sessions and then on the left for 2 consecutive sessions.

### Results

Fig. 1 shows that the number of ethanol reinforcements substantially exceeded the number of water reinforcements for each rat at each concentration regardless of the side of ethanol availability. For each rat at each concentration a *t*-test was made for the difference between the mean number of ethanol reinforcements obtained on each side. In no case was there a significant difference ( $P < 0.05$ ,  $df = 8$ ). Also, shown in Fig. 1 are the mean numbers of reinforcements obtained when the rats were returned to 8% after completing the series of sessions at 32%. On retest at 8%, rat Z-9's and Z-6's reinforcements were decreased 24% and 3%, respectively, relative to the number initially obtained at 8%, whereas reinforcements for rats Z-5 and Z-7 showed increases of 48% and 3%, respectively. On retest with the 8% ethanol solution present for 2 consecutive days on one side and then for 2 consecutive days on the other side, the rats responded on the side where ethanol was available. Thus, the rats did not learn merely to alternate sides between sessions; the rats did discriminate the location of ethanol perhaps on the basis of odor (Meisch and Thompson, 1973).

Dipper presentations on the side opposite ethanol were not increased by the addition of water, and water reinforcements did not vary with ethanol concentration (Fig. 1). Moreover, the concurrent availability of water did not decrease the number of 8% ethanol reinforcements obtained.

As the ethanol concentration was increased, the number of reinforcements obtained decreased (Fig. 1). The decreases were not to a point below one-half that obtained at the adjacent lower concentration, and since the volume drunk was proportional to the number of reinforcements obtained, the quantity (mg) consumed increased with increases in the concentration (Fig. 2).

Fig. 3 shows that for each rat at each concentration most reinforcements were obtained at the beginning of each session. The time course of water intake was

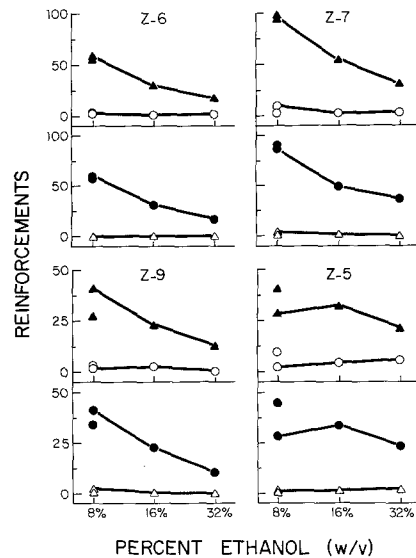


Fig. 1. Ethanol and water reinforcements per 1-hr sessions as a function of ethanol concentration and position, left or right side. Filled triangles: ethanol reinforcements, left side. Unfilled circles: water reinforcements, right side. Filled circles: ethanol reinforcements, right side. Unfilled triangles: water reinforcements, left side. At 8%, where two points are plotted, the unconnected point is the retest value; occasionally, the retest point coincided with the original point, e.g., water reinforcements on left side for rat Z-6. Each symbol represents the mean number of reinforcements obtained during 5 1-hr sessions. Note the different scales of the ordinate

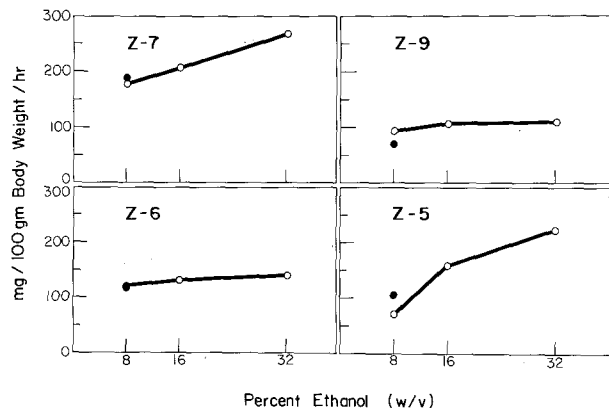


Fig. 2. Quantity mg/100 g body weight/hr of ethanol consumed per 1-hr session as a function of concentration. Each point is the mean value from 10 1-hr sessions. The filled circle at 8% represents the mean value of the retest series of sessions, i.e., the series of sessions following those at 32%

an exception: Reinforcements were low in number and were more evenly distributed throughout the session. When responding occurred it was often in bursts (cf. Fig. 5, Meisch and Thompson, 1974a).

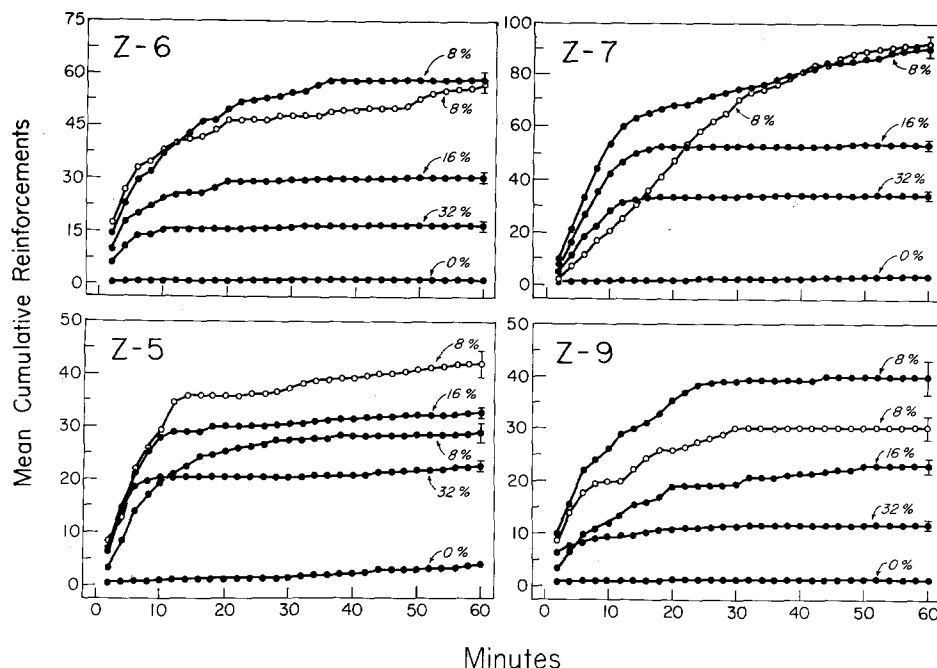


Fig. 3. Mean cumulative reinforcements over 1-hr sessions for each concentration. Each point is a mean based on observations from 10 sessions, except for those points at 0%, which are means from 40 sessions. Unfilled circles represent values from sessions following the series of sessions at 32%. Brackets indicate the standard error of the mean. Absence of brackets indicates that they fell within the area occupied by the plotted point. Note the different scales of the ordinates

### Discussion

Rats concurrently offered water and an ethanol solution chose the ethanol solution regardless of the side of ethanol availability. These results indicate that intake of the ethanol solutions was due to the presence of ethanol and not to their liquid character. It might be argued that the low rate of responding on the side opposite ethanol was due to the fact that early in the experiment such responses resulted in presentations of an empty dipper cup, and thus, such responding may have been extinguished. Two considerations suggest that this interpretation is not correct. First, in experiments where water replaced ethanol solutions, responding was not maintained (Meisch and Thompson, 1972, 1973, 1974a, b). Second, the rats had more than 40 sessions with water on the side opposite ethanol, and this should have provided sufficient opportunity to make contact with its presence.

One interpretation of the present data is that ethanol was consumed by the food-deprived rats because of its caloric value. However, when food-deprived rats were satiated by having unlimited access to food between sessions, ethanol intake persisted over a range of fixed-ratio values (Meisch and Thompson, 1973) and of ethanol concentrations (Meisch and Thompson, 1974a). These results are not consistent with the theory that ethanol's calories are the exclusive

determinant of ethanol intake (*cf.* Meisch and Thompson, 1974b).

The present procedure of providing concurrent access to water and ethanol has several advantages. It eliminates the necessity for separate water control sessions. Certain variables, such as drugs, may be efficiently studied, for the effects of the variables on water and ethanol intake may be examined within the same session. The concurrent procedure also controls within sessions for factors affecting liquid intake that may fluctuate across days. The procedure also provides the additional dependent variable of choice of liquid location. In future experiments the procedure may be used to examine choice between ethanol solutions and other liquids. Finally, this procedure should facilitate comparison of results with those from other laboratories, since most investigators have employed a concurrent water-ethanol option.

The results of this experiment differ from those of Myers and Carey's study (1961). They found that water reinforcements increased as ethanol concentration increased. This discrepancy in results between the two studies is most likely a reflection of the various procedural differences.

The present results extend the generality of conditions under which ethanol serves as a reinforcer to conditions where water is concurrently available. These results also replicate certain earlier findings:

(1) above 8%, reinforcements decreased with increases in the ethanol concentration; (2) quantity (mg) consumed increased with increases in the concentration; and (3) the highest rate of intake occurred at the beginning of the session (Meisch and Thompson, 1972, 1974a, b). This time course of intake is that which should result in ethanol's producing the greatest behavioral effects (Meisch and Thompson, 1974a).

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