Ventricular modification in carbachol-induced drinking: Reply to Routtenberg¹

KHALIL A. KHAVARI, University of Wisconsin-Milwaukee, Milwaukee, Wis. 53201

Two recent studies (Myers & Cicero, 1968; Khavari, Heebink, & Traupmann, 1968) presented data which failed to support the view proposed by Routtenberg that carbachol-induced drinking may be due to ventricular modification (Routtenberg, 1967). Routtenberg (1968a, b) has raised some questions regarding the above two studies. In this note I am answering the three questions posed by Routtenberg (1968b) regarding our report (Khavari et al, 1968).

(1) "The purpose of this note is to point out that the negative results of Khavari et al (1968) may be in part attributed to the same problem of application of excessive amounts of carbachol [Routtenberg, 1968b, p. 99]." Routtenberg's previous comment (1968a) on Myers and Cicero's report is in point: "Thus, most prior investigations have administered a small quantity of carbachol (0.5-5 μ g) in a single application, and then tested for the effects of such administration immediately after the single application for about a 1-h period [p. 106]." In our study (Khavari et al, 1968) we tested five dosages which are considered *small* by Routtenberg himself. In addition, the water intake of the rats was measured for a 1-h period immediately after application of a single dose, in the best tradition of "most prior investigations." Clearly, the doses of carbachol used in our study were not excessive.

Routtenberg (1968b) has performed a Spearman rank correlation coefficient test with selected portions of our data (Khavari et al, 1968; Table 1; comparison of mean water intakes with the five doses of carbachol) and reported that there is a significant relation between dose level and water intake. However, the above statistical reasoning not only fails to support his point but ignores the remainder of our data. Specifically, when changes in water intake of each group under a given dose of

carbachol are compared to predrug baseline, we find no dose-drinking relations of any kind (Table 1). Clearly, the most appropriate comparison for detection of carbachol effects on drinking of each group of rats is in utilization of baseline-drug data for that group. Routtenberg (1968b) has used the data in the third column of Table 1 for his statistical treatment. However, when the differences in drinking of each group under carbachol and saline is considered (Table 1, Column 4) we find no significant dose-drinking correlation ($r_s = -.61$).

Finally, to further support the contention that excessive ventricular doses of carbachol have been responsible for failure to obtain drinking, Routtenberg (1968a, p. 106) states that "It should be pointed out, too, that other investigators have, in fact, obtained drinking by application of carbachol to the ventricle. Fisher & Levitt (1967) found that carbachol applied at low doses produced drinking behavior, but that higher doses did not." I cannot find any statement by Fisher and Levitt to the effect that they have obtained drinking with low intraventricular doses of carbachol. However, in the same paper that Routtenberg has cited, Fisher and Levitt state that "Direct ventricular injection of the amount of crystalline carbachol we usually insert into brain (1 to 3 μ g) produced catatonia, tremors, or bizarre motor behavior. Significant amounts of drinking did not occur [Fisher & Levitt, 1967, p. 840]."

(2) Routtenberg's second point deals with the fact that we have used water-deprived Ss. He states that "... the authors report the effect of carbachol on 23 h water deprived animals. Such a procedure would raise baseline or control levels, and could mask any effects of carbachol-induced drinking that were present [Routtenberg, 1968b, p. 99]." It is difficult to appreciate the above point for two reasons. First, in our study (Khavari et al, 1968) measures of drinking for determination of baseline control and carbachol conditions were taken

when the Ss were exposed to the same identical water deprivation schedule, i.e., both predrug and drug tests were performed when the Ss were water-deprived for exactly 23 h. Second, the fact that carbachol application in the central thirst circuit of water-deprived rats significantly increases their drinking over the nonstimulated controls is well documented (e.g., see Miller, 1965; Grossman, 1967).

(3) Routtenberg's third comment is in regard to the vehicle employed in our study (10 μ l of distilled water). He suggested that "Such a hypotonic solution when injected into the ventricle might act to suppress drinking behavior [Routtenberg, 1968b, p. 99]." This possibility can be also excluded on the ground that 10 μ l of a hypotonic solution can hardly significantly alter the tonicity of the rats' cerebrospinal fluid. The brain ventricles of rats contain anywhere from 120-500 μ l of fluid (see Cicero & Myers, 1968; Reed et al, 1967); therefore, our 10 μ l of hypotonic solution seems to be an unlikely source of confoundment.

At this time, on the basis of data from our laboratory and the findings of Myers and his colleagues at Purdue, we are impelled to conclude that the notion of ventricular modification fails to account for carbachol-induced drinking.

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NOTE

1. Due to a family illness I have been abroad for the past few months and returned recently to the U.S. Therefore, my apologies for the delay in preparation and submission of this overdue note.

Table 1

Mean water intake in ml for predrug and drug tests are shown in Columns 2 and 3, respectively. Column 4 shows the water intake of each group of Ss under a given dose of carbachol, expressed in per cents of their intake under predrug saline.

Carbachol Dose (μ g/rat)	Intake in ml Predrug	Intake in ml Drug	Intake under Drug % Predrug	Rank
0.25	24.2	27.2	112	1
0.5	24.8	22.7	91	3
1.0	23.4	22.6	96	2
2.0	24.4	20.8	85	5
4.0	21.8	19.5	89	4