

# Modification of Lipemic Responses to an Alcohol-Corn Oil Mixture<sup>1</sup>

JOSEPH J. BARBORIAK, ARTHUR S. WILSON AND ROBERT C. MEADE  
*Research, Psychology and Radioisotope Services, Wood Veterans Administration Center, and Departments of Pharmacology, Psychiatry and Medicine, Marquette School of Medicine, Milwaukee, Wisconsin*

**ABSTRACT** Chronic ingestion of 15% (w/v) alcohol in lieu of drinking water in rats fed ad libitum failed to modify the usual lipemic response to a single dose of corn oil-alcohol mixture. The rise in plasma triglycerides was associated with an increase in their <sup>14</sup>C-specific activity indicating that the additional plasma lipids were derived from the <sup>14</sup>C-labeled dietary fat. The lipemic response was, however, markedly reduced by restricting the food intake of the animals to 2 hours/day.

It has been shown in previous studies with both human and animal subjects that consumption of a single large dose of alcohol prior to or with a meal enhanced the usual postprandial lipemia (1-7). In animals, the hepatic triglyceride levels were also increased (5-7). Chronic ingestion of relatively large amounts of dilute alcohol, however, failed to affect the plasma and liver triglyceride levels of rats (8, 9), possibly because of the increased activity of alcohol dehydrogenase and faster metabolic breakdown of alcohol reported to occur during long-term administration of alcohol (10, 11).

We decided, therefore, to find out whether previous exposure of animals to dilute alcohol for an extended period of time can modify the hyperlipemic response to a subsequent single large dose of alcohol. In addition, we investigated the effect of a reduced caloric intake on the ethanol-induced rise in plasma and liver lipids.

## METHODS

Forty male rats of the Sprague-Dawley strain initially weighing approximately 250 g each were used in the study. Twenty animals received a commercial rat ration<sup>2</sup> ad libitum; the remaining 20 received the same ration restricted to only 2 hours feeding time during each 24-hour period (8 to 10 AM). Each group was further subdivided into two subgroups. One subgroup received drinking water, the other subgroup was offered a 15% ethanol (w/v) solution for 28 days. Weekly weight changes and daily food and liquid intakes were recorded for each animal.

On day 29 of the experiment, food was removed and only water was offered as a liquid to all the animals. One half the animals in each group was given 5 g corn oil and 5 g ethanol/kg body weight by stomach tube. The remaining five animals in each group received in the same manner, 5 g corn oil/kg body weight and an amount of dextrose isocaloric with alcohol. The corn oil contained trace amounts of triolein-1-<sup>14</sup>C.<sup>3</sup> Analysis by thin-layer chromatography (12) revealed that more than 95% of the radioactivity was associated with the triolein moiety. Sixteen hours later, the animals were bled by heart puncture; their livers were removed, blotted on a filter paper and weighed. The blood samples were immediately centrifugated in a refrigerated centrifuge, and the plasma and tissue samples were kept at -15° until processed. Plasma and liver triglyceride levels were determined by conventional procedures (13, 14) and <sup>14</sup>C-activity of the plasma liver extracts lipid was measured in a scintillation spectrometer after absorption of phospholipids (13).

## RESULTS

Information on daily weight gain and food and water intake is summarized in table 1. The animals in the group receiving food and water ad libitum consumed an average of 30.4 ± 3.2 g (mean ± standard error) of food and gained 5.3 ± 0.2 g/day

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<sup>2</sup> Purina Rat Chow, Ralston Purina Company, St. Louis, Mo.

<sup>3</sup> Tracerlab, Waltham, Mass.

TABLE 1  
Effect of chronic alcohol administration and caloric restriction on weight gain,  
and food and liquid intakes of rats<sup>1</sup>

Food	Alcohol	No. of rats	Food intake	Wt gain	Water intake
			<i>g/day</i>	<i>g/day</i>	<i>g/day</i>
Ad libitum	—	10	30.4 ± 3.2	5.3 ± 0.2	46.0 ± 0.5
	+	10	21.9 ± 0.3	3.4 ± 0.3	20.5 ± 0.3
Restricted	—	10	16.0 ± 0.1	1.6 ± 0.3	28.7 ± 0.5
	+	10	12.0 ± 0.2	2.0 ± 0.2	22.0 ± 0.3

<sup>1</sup> Fifteen percent (w/v) alcohol or water was offered for 28 days; animals on caloric restriction received food for 2 hours each day. All values mean ± SE.

during the 4-week experimental period. Replacement of drinking water by 15% ethanol reduced the food intake to  $21.9 \pm 0.3$  g/day and the daily weight gain to  $3.4 \pm 0.3$  g. The liquid intake was also significantly reduced in this group. Animals limited to food for 2 hours/day and offered drinking water consumed  $16.0 \pm 0.1$  g food and gained  $1.6 \pm 0.3$  g/day. Animals in the subgroup with 15% alcohol as the drinking solution ate less food,  $12.0 \pm 0.2$  g, but gained slightly more weight,  $2.0 \pm 0.2$  g/day, probably reflecting the utilization of alcohol calories. This group drank  $22.0 \pm 0.3$  ml of the 15% alcohol solution per day, slightly more than the animals receiving food ad libitum and 15% alcohol which consumed  $20.5 \pm 0.3$  ml of the alcohol solution per day.

The effect of the dietary regimen and chronic alcohol intake on the lipemic response to the alcohol-corn oil mixture is shown in figure 1. Animals fed ad libitum reacted to the alcohol-corn oil mixture with a pronounced rise in plasma triglyceride levels. This lipid rise did not seem to be modified by a previous 4-week intake of 15% alcohol in lieu of drinking water. The increase in plasma triglycerides was associated with a rise in <sup>14</sup>C-specific activity of the lipids, indicating that the additional triglycerides appearing in the blood were derived from the diet. When food intake was restricted to 2 hours/day, the lipemic reaction to the alcohol-corn oil mixture was less marked than in the animal fed ad libitum. Again, the administration of 15% alcohol in place of water for 4 weeks prior to the test did not seem to enhance or reduce to any great extent the response to the alcohol-corn oil mixture.

The changes in hepatic triglyceride levels and <sup>14</sup>C-specific activity of hepatic

lipids are shown in figure 2. The animals fed ad libitum and offered drinking water had an average liver triglyceride level of  $10.7 \pm 0.4$  mg/g fresh tissue when receiving corn oil with glucose, and  $20.1 \pm 1.0$  mg/g when the corn oil was administered with alcohol. This rise in hepatic triglycerides, however, was not reflected in corresponding changes of <sup>14</sup>C-specific activity. The animals fed ad libitum and receiving 15% alcohol for drinking also showed a marked increase of liver triglycerides following administration of corn oil plus alcohol. In this group, the rise in triglyceride level was associated with an increase in <sup>14</sup>C-specific activity.

The restriction of food intake to 2 hours/day was, in general, associated with lower hepatic triglyceride levels and higher <sup>14</sup>C-specific activities than seen in the groups fed ad libitum. Regardless of the type of drinking solutions, administration of the corn oil-ethanol mixture was followed by a rise in hepatic triglycerides.

#### DISCUSSION

Chronic ingestion of 15% alcohol as drinking solution by the rats fed ad libitum failed to modify the lipemic response to the alcohol-corn oil mixture. It would seem, therefore, that an increase in alcohol dehydrogenase activity, observed to occur in animals given alcohol for longer periods of time (10, 11) was not able to prevent the lipemia-enhancing effect of the alcohol-oil combination.

The lipemic response to the alcohol-oil combination was, however, markedly reduced by the restriction of food intake to 2 hours/day. The animals on the restricted food regimen were probably in a state of caloric deficiency. It is possible, therefore, that the administered corn oil was pri-

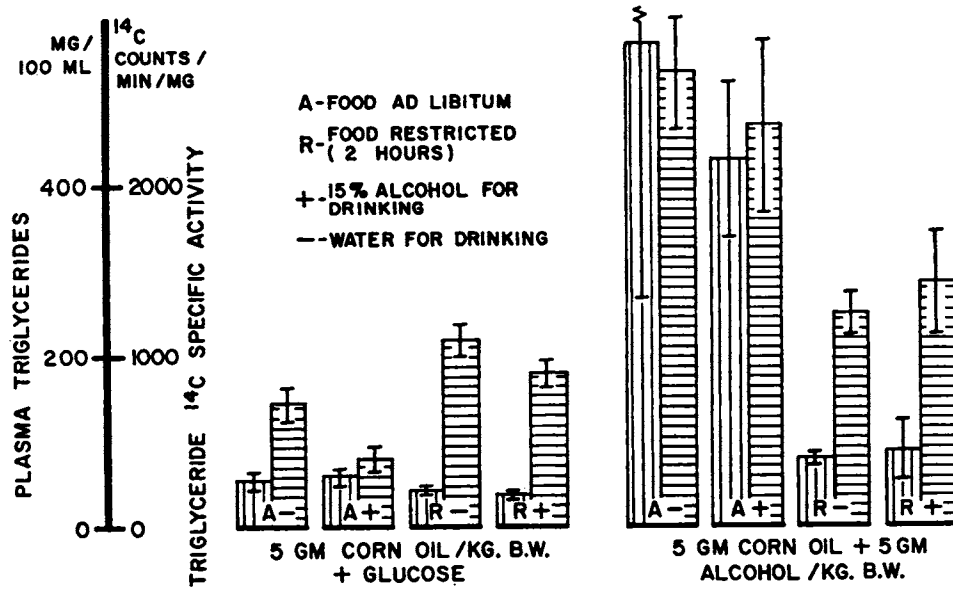


Fig. 1 Effect of chronic alcohol ingestion on plasma triglyceride levels (vertical stipling) and <sup>14</sup>C-specific activity of plasma lipids (horizontal stipling) in rats receiving a corn oil-alcohol mixture. The vertical lines represent standard error of the mean.

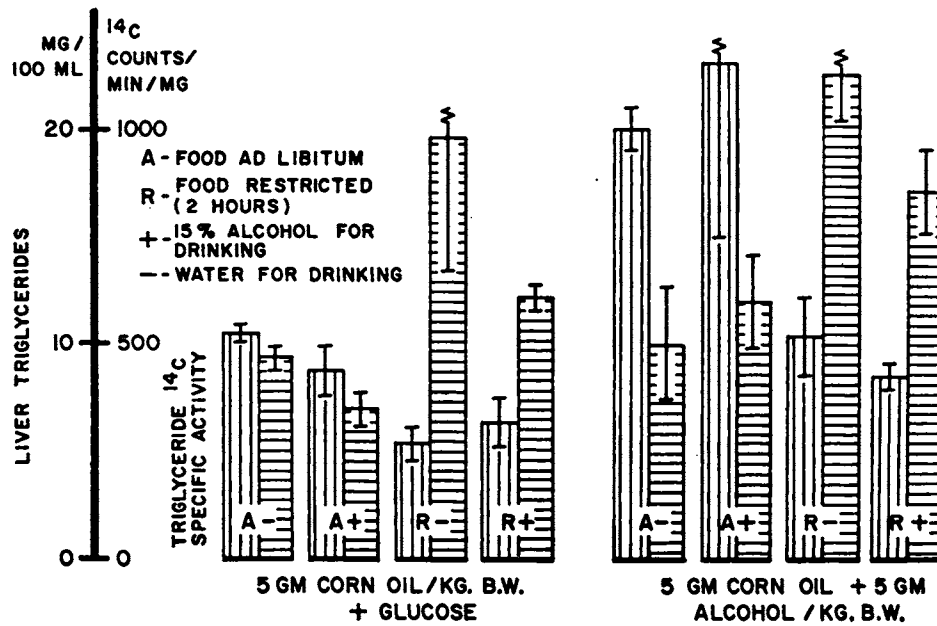


Fig. 2 Effect of chronic alcohol ingestion on hepatic triglyceride levels (vertical stipling) and <sup>14</sup>C-specific activity of hepatic lipids (horizontal stipling) in rats receiving a corn oil-alcohol mixture. The vertical lines represent standard error of the mean.

marily utilized as an immediate source of energy and only small portions remained to contribute to the build-up of plasma lipids. A rapid metabolic utilization of the dietary fat by the animals with restricted food intake is also suggested by the higher  $^{14}\text{C}$ -activity of their hepatic triglycerides. Furthermore, periodic food restrictions, such as used in the present study have been reported to enhance utilization of dietary substrates (15, 16) and this might have decreased the availability of the administered fat as a source of blood lipids.

The definite mechanism of the alcohol-induced enhancement of postprandial lipemia seen in rats fed ad libitum is not well understood at this time. The increase in  $^{14}\text{C}$ -specific activity of plasma lipids indicates that the triglycerides were mainly derived from the diet. The presence of high concentration of the  $^{14}\text{C}$ -label in blood lipids as late as 16 hours after administration of the fat suggests a delay in the metabolic processing of the absorbed corn oil (7).

Replacing the drinking water with the 15% alcohol solution in the rats fed ad libitum led to a reduced food intake and lower weight gains. Similar growth-retarding effects of chronic alcohol administration have been observed even in studies in which the energy deficit, due to low food intake was counterbalanced by the caloric content of the ingested alcohol (8, 17). It is not clear whether the observed impairment of growth is due to a direct effect of alcohol (18) or to a reduced intake of dietary factors needed for optimal growth, as suggested by Lucas et al. (19). Animals receiving restricted amounts of food and offered alcohol grew, however, at a slightly faster rate than animals receiving the restricted diet and water. Furthermore, the former group consumed more alcohol than the animals fed ad libitum and given alcohol.

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#### LITERATURE CITED

1. Talbott, G. D., and B. M. Keating 1962 Effects of preprandial whiskey on postalimentary lipemia. *Geriatrics*, 17: 802.
2. Brewster, A. C., H. G. Lankford, M. G. Schwartz and J. F. Sullivan 1966 Ethanol and alimentary lipemia. *Amer. J. Clin. Nutr.*, 19: 255.
3. Verdy, M., and A. Gattereau 1967 Ethanol, lipase activity and serum lipid level. *Amer. J. Clin. Nutr.*, 20: 997.
4. Barboriak, J. J., and R. C. Meade 1968 Enhancement of alimentary lipemia by preprandial alcohol. *Amer. J. Med. Sci.*, 255: 245.
5. DiLuzio, N. R., and M. Poggi 1963 Abnormal lipid tolerance and hyperlipemia in acute ethanol-treated rats. *Life Sci.*, 2: 751.
6. Barboriak, J. J. 1966 Effect of ethanol on lipoprotein lipase activity. *Life Sci.*, 5: 237.
7. Barboriak, J. J., and R. C. Meade 1968 Post-alcoholic lipemia in the rat. *Quart. J. Stud. Alc.*, 29: 283.
8. Barboriak, J. J., and A. S. Wilson 1966 Interaction of ethanol and feeding schedules. *Proc. 7th Int. Congr. Nutr.*, Hamburg, 2: 194.
9. Scheig, R., N. A. Alexander and G. Klatskin 1966 Effects of prolonged ingestion of glucose or ethanol on tissue lipid composition and lipid biosynthesis in rat. *J. Lipid Res.*, 7: 188.
10. Dajani, R. M., J. Danielski and J. M. Orten 1963 Utilization of ethanol. II. The alcohol-acetaldehyde dehydrogenase systems in the livers of alcohol-treated rats. *J. Nutr.*, 80: 196.
11. Hawkins, R. D., H. Kalant and J. M. Khanna 1966 Effect of chronic intake of ethanol on rate of ethanol metabolism. *Can. J. Physiol. Pharmacol.*, 44: 241.
12. Tuna, N., H. K. Mangold and D. G. Mosser 1963 Re-evaluation of the  $\text{I}^{131}$ -triolein absorption test. Analysis and purification of commercial radioiodinated triolein and clinical studies with pure preparations. *J. Lab. Clin. Med.*, 61: 620.
13. Van Handel, E., and D. B. Zilversmit 1957 Micromethod for the direct determination of serum triglycerides. *J. Lab. Clin. Med.*, 50: 152.
14. Butler, W. M., Jr., H. M. Maling, M. G. Horning and B. B. Brodie 1961 The direct determination of liver triglycerides. *J. Lipid Res.*, 2: 95.
15. Tepperman, J., and H. M. Tepperman 1958 Effects of antecedent food intake pattern on hepatic lipogenesis. *Amer. J. Physiol.*, 193: 55.
16. Leveille, G. A., and R. W. Hanson 1965 Influence of periodicity of eating on adipose tissue metabolism in the rat. *Can. J. Physiol. Pharmacol.*, 43: 857.
17. Aebi, H., and J. P. von Wartburg 1960 Vergleichendbiologische Aspekte der experimentellen Erforschung chronischer Alkoholwirkungen. *Bull. Schweiz. Akad. Wiss.*, 16: 25.
18. Mallov, S. 1955 Effect of chronic ethanol intoxication on liver lipid content of rats. *Proc. Soc. Exp. Biol. Med.*, 88: 246.
19. Lucas, C. C., J. H. Ridout and G. L. Lumchik 1968 Dietary protein and chronic intoxication with ethanol. *Can. J. Physiol. Pharmacol.*, 46: 475.