Role of acetate in the reduction of plasma free fatty acids produced by ethanol in man

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ABSTRACT To investigate the mechanism by which ethanol lowers plasma free fatty acids, we tested the ability of two products of alcohol metabolism, acetate and lactate, to lower free fatty acids in man. Sodium acetate was given orally to five healthy fasting volunteers and caused a significant fall in plasma free fatty acids. After amounts of ethanol and acetate that produced similar reductions in free fatty acids, plasma acetate increased 3- to 4-fold within 20 min. In each of three subjects the fall of free fatty acids observed after acetate ingestion occurred at plasma acetate levels less than or equal to those reached after ethanol. In all studies plasma glucose remained stable. Oral administration of sodium lactate to another volunteer in amounts sufficient to raise plasma lactate concentrations to a level similar to that found after ethanol administration failed to lower plasma free fatty acids.

Thus acetate, a metabolite of ethanol, reduces plasma free fatty acids at plasma acetate levels comparable to those resulting from ethanol metabolism, which suggests that the lowering of plasma free fatty acids produced by ethanol is mediated, at least in part, by acetate.

KEY WORDS ethanol · free fatty acids · acetate · gas chromatography · lactate

ETHANOL HAS BEEN SHOWN to lower circulating free fatty acid (FFA) levels when given over a short period of time either intravenously (1) or orally (2, 3) to healthy volunteers. The depressant effect, which has been confirmed by other groups (4, 5), is associated with a lowering of the peripheral venous–arterial FFA difference (1), a decreased FFA turnover (6), and a decline in arterial glycerol levels (3). These studies indicate that the fall in plasma FFA concentration is due to decreased release from peripheral depots.

Ethanol probably exerts this effect indirectly since it did not affect glycerol release from adipose tissue in vitro at concentrations from 20 to 200–500 mg/100 ml (7). This led us to inquire whether a metabolic product of alcohol might be responsible for the fall of FFA. We investigated the effects of two metabolites whose plasma concentrations increase after ethanol administration, namely acetate (8) and lactate (9–11). The former, which has been shown to increase in concentration as much as 20-fold after ethanol administration (12), was found to be capable of producing a fall in FFA. A preliminary account of this work was presented recently (13).

METHODS

Six healthy male volunteers were studied under metabolic ward conditions. They had a history of excessive alcohol consumption, but at the time of the study had not ingested alcohol for periods of 2–6 wk. They had no clinical or laboratory evidence of liver disease or disturbance in lipid metabolism. Subjects were studied after an overnight fast with strict abstention from smoking. Five subjects were studied for periods of 1.5 hr, during each of which the subjects drank approximately 2 ml/kg of a noncaloric beverage (No-Cal) every 15 min. After a 30 min control period, either sodium acetate or ethanol was added to the carrier beverage. Sodium acetate was given in two doses to five subjects: 143 mg/kg initially and 71 mg/kg 30 min later. Three of the five subjects given acetate were also tested with ethanol 1 wk before

Abbreviation: FFA, free fatty acids.

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or after the day of the acetate test: ethanol was ingested in No-Cal in four doses of 250 mg/kg each, drunk every 15 min.

9-hr tests were conducted on one volunteer who was given either sodium lactate or ethanol for 6.5 hr (after a 2.5 hr control period) on two separate days. FFA values were also obtained during a comparable fasting period on a third day. The subject drank a solution of sodium lactate (5 g/100 ml) in water flavored with grapefruit juice at doses of 45 mg/kg every 15 min for the first 3 hr followed by 22 mg/kg every 15 min for the final 3.5 hr. He drank ethanol (as a 12.5 g/100 ml solution in the same carrier beverage) every 15 min in doses of 113 mg/kg for the first 4 hr and 50 mg/kg for the final 2.5 hr. The subjects consumed all beverages within 2 min of the time of administration.

Blood samples were drawn without stasis in all subjects during both control and test periods. Venous samples were obtained through a polyethylene catheter or 20 gauge needle (with slow infusion of normal saline to prevent hemostasis), or from an indwelling Cournand needle in the antecubital vein.

Blood was immediately placed in heparinized tubes and centrifuged at 5°C. Measurements of FFA, ethanol, and glucose in the plasma (3) and lactate in the blood (1) were made as previously described and, as found previously (1), neither ethanol nor lactate interfered with the measurement of FFA. Acetate was determined in a perchloric acid filtrate of plasma as described by Lester (14) with the following modifications. An F&M model 400–5 Biomedical Analyzer gas chromatograph was used at $^1/_{10}$ full sensitivity. After vacuum distillation over anhydrous copper sulfate, samples were injected directly onto a 1500 × 6 mm I.D. glass column packed with uncoated porous polymer bead "Porapak Q", 50–80 mesh (Hewlett–Packard); we are indebted to Dr. David Lester for information about this method. The column was maintained at 130°C and the injector at 180°C. Nitrogen carrier gas was used and the effluents were ionized in a hydrogen–oxygen flame. The retention time for acetate on this column was 10 min. No preliminary column (14) was found to be necessary under the conditions described. The conversion constant K = 15.7 (14) was verified with standards run simultaneously with unknown samples. Recovery of sodium acetate added to plasma samples was 97%.

Significance of observed differences was determined by Student's "t" test (15).

RESULTS

In each of the five volunteers given sodium acetate (143 mg/kg in 30 min) there was a striking fall in plasma FFA (Fig. 1), about 0.75 as great as the FFA fall produced by

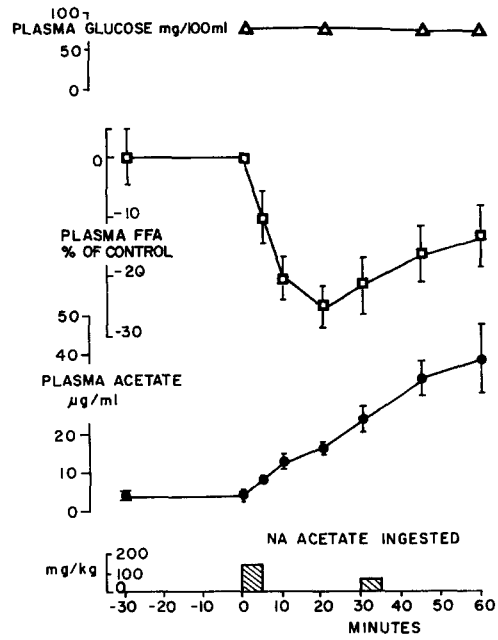

FIG. 1. Effect of oral administration of sodium acetate on plasma glucose (△), FFA (□), and acetate (●). Points represent average values for five volunteers. Variation is expressed as SEM. Plasma FFA at zero time averaged 572 ± 91 μeq/liter.

ethanol (500 mg/kg in 30 min) in three of these subjects (Fig. 2). Both ethanol and acetate caused a drop of FFA within 5 min, with a maximum decrease of FFA (25% of control or more) within 20 min. In each subject, the difference in FFA concentration between the point of maximum decrease of FFA and the corresponding control value (at zero time) was calculated; the mean difference was significantly greater than zero both after acetate ($P < 0.01$) and after ethanol ($P < 0.02$).

Acetate and ethanol both produced a rise in plasma acetate levels, from average baseline concentrations of 5 μg/ml to 16 and 35 μg/ml, respectively, at the time of maximum depression of FFA. Plasma acetate levels after ingestion of ethanol were at no time lower than those after administration of acetate. Plasma acetate levels increased within 5 min after either ethanol or acetate ingestion (Figs. 1 and 2), which indicates rapid absorption from the gastrointestinal tract.

One subject was given ethanol and sodium lactate orally (Fig. 3) on two separate occasions. After ethanol, the FFA dropped to an extent similar to that described previously (3), and blood lactate increased. Ingestion of sodium lactate produced a similar increase in blood lactate but failed to depress FFA. No marked change occurred during a comparable period of fasting. Intravenous administration of lactate has previously been shown to be similarly ineffectual in lowering plasma FFA concentrations in three individuals tested (1).

Plasma glucose levels stayed constant throughout the

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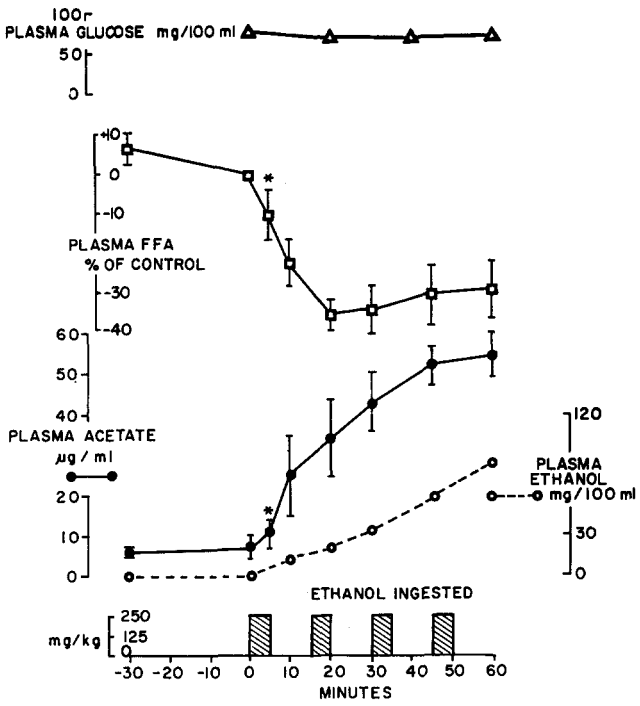

FIG. 2. Effect of oral administration of ethanol on plasma glucose (△), FFA (□), acetate (●), and ethanol (○). Points represent average values for three of the five volunteers studied in Fig. 1. Variation is expressed as SEM, except for the 5 min point (*) which is the average of two values ± range. Plasma FFA at zero time averaged 472 ± 48 µeq/liter.

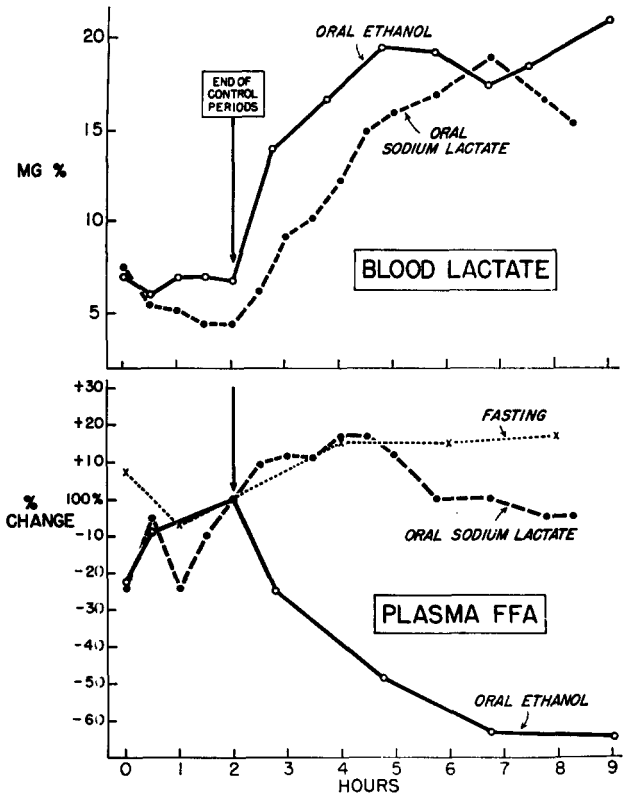

FIG. 3. Effect of ethanol (○) and sodium lactate (●) administered orally on two different occasions to the same subject. Fasting values (×---×) were obtained on a third experimental day. Above, blood lactate concentration; below, plasma FFA changes.

tests (Figs. 1 and 2) and plasma ethanol levels increased steadily after its ingestion (Fig. 2).

Some subjects became slightly euphoric, but not intoxicated, after the amounts of ethanol used.

DISCUSSION

Preliminary results of tests with intravenously administered sodium acetate (13, 16) demonstrated the ability of this compound to lower plasma FFA in man. Since ingestion of ethanol is known to increase plasma acetate levels as well as decrease the concentration of plasma FFA (1–5), we wondered whether the increase of plasma acetate could be responsible for the FFA-lowering effect of ethanol. The preliminary studies which we carried out with intravenous administration of acetate (13, 16) did not answer this question since levels of plasma acetate were not determined; in the absence of such measurements we could not know whether the concentrations of acetate reached in the plasma after intravenous acetate administration were comparable to those reached after ethanol ingestion. The present study demonstrates that ingestion of acetate can cause a decrease in plasma FFA in man comparable to that obtained with ethanol in the same subjects. Since acetate ingestion produced plasma acetate levels equal to or lower than those reached after administration of ethanol, acetate could be responsible for the FFA-lowering effect of ethanol.

The mechanism whereby acetate lowers FFA has not been studied in the present investigation, and no FFA turnover studies were carried out after the administration of acetate. The reduction in FFA following acetate administration was, however, of the same order of magnitude as the decrease produced by ethanol in both the present and previous (1–3) studies, and the FFA fall after ethanol ingestion has been shown to be associated with decreased FFA turnover (6). These results, as well as the reduction in plasma glycerol induced by ethanol ingestion (3), strongly suggest that the decrease of FFA after ethanol results from decreased FFA release from depots; a similar mechanism could play a role after acetate ingestion.

The metabolism of ethanol results not only in increased plasma acetate but also in acidosis and hyperlactacidemia (9–11, 17), which in principle could play a role in the FFA drop. In the present study, however, it was found that sodium acetate, a known alkalinizing agent, reduces FFA at blood acetate levels comparable to those found after ingestion of ethanol. This suggests that the increase in blood acetate after ethanol is sufficient to explain the FFA fall even without acidosis. Administration of very

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large amounts of lactate has been reported to lower FFA (18). At blood lactate levels comparable to those observed following ethanol ingestion, however, lactate (administered intravenously to three individuals in a previous study) had no such effect (1). This was confirmed in the present study in one subject given lactate by the oral route (Fig. 3). Both sodium lactate and sodium acetate are alkalinizing agents, but since the fall of plasma FFA was observed only after sodium acetate, acetate per se rather than alkalinization seems to be responsible, although pH was not measured in the present study.

Finally, it is evident from these as well as previous studies (1–3) that the lowering of plasma FFA is not due to a change in plasma glucose concentration, since the latter remained constant throughout the course of the experiments with both acetate and ethanol. Small changes in carbohydrate metabolism could, however, have gone undetected.

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