Ethanol-Induced Hyperlacticacidemia: Inhibition of Lactate Utilization

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ABSTRACT The effects of oral ethanol administration on blood glucose and lactate concentrations, lactate inflow and outflow rates, and lactate incorporation into glucose were investigated in eight human volunteers. Lactate incorporation into glucose, lactate turnover, and lactate inflow and outflow rates were determined during an 8 hr constant infusion of 100 μCi of lactate-U-^{14}C. Ethanol was administered by mouth at hourly intervals, 60 ml of bonded whiskey initially and 30 ml/hr thereafter. Blood lactate concentrations increased precipitously after the administration of ethanol, reached a plateau within 120–180 min, and remained constant thereafter despite the continued administration of ethanol. Before ethanol, the lactate turnover rate was 0.76 mmoles/kg per hr ±0.05 (SEM) and lactate inflow and outflow rates were closely balanced. During the administration of ethanol, the lactate inflow rate was unchanged, but the lactate outflow rate was significantly inhibited, decreasing to 50% of the inflow rate. Despite the continued administration of ethanol, equilibrium between lactate inflow and outflow was restored within 120–180 min and coincided temporally with establishment of a constant blood lactate concentration. Lactate oxidation was unaltered by ethanol, but lactate incorporation into glucose was significantly inhibited. Lactate incorporation into glucose was reduced within 30 min of the administration of ethanol, and nadir values were reached within 120–180 min. Lactate incorporation into glucose remained constant thereafter at rates that were only 30% of those observed in the absence of ethanol. The results of these studies indicate that ethanol-induced hyperlacticacidemia is due to decreased lactate disposal rather than increased lactate production.

INTRODUCTION

Many alterations in intermediary metabolism are associated with the administration of ethanol (1–3). Although hypoglycemia is perhaps the most important clinical disorder of carbohydrate metabolism, produced by ethanol (4, 5), profound changes also occur in lactate metabolism (1–3). Elevated blood lactate concentrations are frequently observed in acute alcoholism (6, 7) and with the administration of ethanol to normal and alcoholic subjects (8–13). While recent reviews of the metabolic effects of ethanol attribute hyperlacticacidemia to increased hepatic production of lactate (1–3), there appears to be considerable confusion over the exact mechanism. Many of the studies in man (10, 11, 14) which are quoted in defense of the concept that ethanol increases splanchnic lactate production do not provide sufficient information to justify such a conclusion. In fact, the one study in which appropriate data are available indicates that ethanol decreases hepatic lactate removal rather than increasing production (15). Furthermore, studies with the isolated perfused rat liver (16–18) do not indicate that ethanol increases hepatic lactate production.

In general, hyperlacticacidemia may result from either increased lactate production, decreased utilization, or a combination of both. The possibility that the hyperlacticacidemia is due to decreased lactate removal must be seriously considered in view of the fact that the clearance of lactate from the blood after a lactate load (19) or exercise (20) is impaired by ethanol. Although little is known about lactate disposal, lactate is an important precursor of glucose in animals (21) and man (22), and inhibition of its incorporation into glucose could lead to its accumulation in blood.

The studies described in this paper were designed to evaluate the contribution of increased production and/or decreased utilization of lactate to the hyperlacticacidemia produced by ethanol. The results demonstrate that ethanol-induced hyperlacticacidemia is due to inhibition of lactate utilization rather than increased production.

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METHODS

A total of 10 lactate turnover studies were performed in six male and two female volunteers aged 23–30 yr, described in Table I. Subjects G. F. and D. S. received no ethanol, subjects G. S., J. G., L. G., and J. M. received ethanol only, and subjects H. B. and R. G. were studied before and after ethanol. Each study was begun between 7:30 and 8:00 a.m. after a 9–10 hr fast. 100 μCi of sodium L-(+) lactate-U-^{14}C (Nuclear-Chicago, Des Plaines, Ill.) was administered through an indwelling polyethylene venous cannula by a primed infusion technique in which the ratio of the priming dose (μCi) to infusion rate (μCi/hr) was 1:1. Blood was withdrawn at 15- to 30-min intervals through a similar cannula placed in the opposite arm that was kept patent with a slow infusion of isotonic saline or with a dilute heparin solution. 10 ml of whole blood was immediately deproteinized with 10 ml of 7% perchloric acid and stored in an ice bath until centrifuged at 4°C. The supernatant was neutralized to pH 7.0–7.4 with 5 N potassium hydroxide and centrifuged at 4°C to remove the potassium perchlorate precipitate. An aliquot (5.0 ml) of the neutralized extract was passed through a column (0.8 × 25 cm) containing 2.5 g of Dowex 1-X8 resin (formate form) to remove labeled anionic metabolic intermediates (22, 23). The column was rinsed with 20 ml of deionized distilled water and the combined eluates were counted and analyzed for glucose (24). Although this method does not separate alanine or glycerol from glucose, the contribution of these substrates to the radioactivity in this fraction, in both the control and ethanol studies, was relatively minor. Isolation of glucose as the potassium gluconate derivative (25) from the eluates of the 480-min samples revealed that 92.5 ±3.4% (SEM; n = 4) of the radioactivity in the control studies and 93.6 ±3.8% (n = 6) of the radioactivity in the ethanol studies was actually in glucose. The column was then washed with 25 ml of 0.05 N formic acid to remove intermediates which were less strongly bound to the resin than lactate. This fraction, which contained such compounds as acetoacetate, aspartate, and beta-hydroxybutyrate (23) was discarded. Lactate was removed from the column with 25 ml of 0.20 N formic acid and the eluate was counted and analyzed for lactate (26). Recovery of glucose-1-^{14}C and L-(+) lactate-U-^{14}C added to blood before precipitation by these techniques was 97–100%. 10-ml aliquots of the glucose and lactate-containing eluates were added to 13 ml of a toluene–Triton-X scintillation counting fluid (27). All samples were assayed for radioactivity in a Nuclear-Chicago Liquid Scintillation spectrophotometer and corrections for efficiency of counting were made by the chemicals ratio technique. Blood lactate concentrations were determined by measurement of lactate in an aliquot (0.5 ml) of the neutralized blood extract. Blood glucose concentrations were determined by Auto Analyzer techniques on oxalated whole blood (28).

During the infusion of L-(+) lactate-1-^{14}C, expired air was collected in a Douglas bag at 30-min intervals for 2 min. The minute ventilatory volume was determined with a Wright physiologic respirometer. Samples were analyzed immediately for CO_2 content with a Godaart Pulmonanalyzer (Instrumentation Associates Inc., New York) and $^{14}CO_2$ was trapped in hyamine (29). Aliquots of the hyamine trap were added to scintillation counting fluid (2,5-diphenyloxozole, 5.0 g; toluene, 1 liter) and assayed for radioactivity in a Nuclear-Chicago or Beckman Ambient Temperature Liquid Scintillation spectrophotometer. Corrections for efficiency of counting were also made by the channels ratio technique or an automated external standard.

Lactate turnover and oxidation and the per cent of glucose derived from lactate were calculated from relationships described previously (22). Lactate inflow and outflow were calculated from the following equations, described by Steele for glucose (30), with the exception that the lactate space was considered to be equivalent to the total body water:

$$R_1 = F - \frac{(l_2 + l_1/2)}{l_1}(\text{tbw} \times l_1)\frac{(SA_2 - SA_1)}{t_2 - t_1} \Big/ \frac{SA_2 + SA_1}{2} \quad (1)$$

$$R_2 = R_1 - \frac{(l_2 - l_1)}{l_1}(l_1 \times \text{tbw}) \Big/ t_2 - t_1 \quad (2)$$

where R_1 and R_2 are the lactate inflow and outflow rates (mmoles/min); F is the infusion rate of lactate-^{14}C (dpm/min); l_1 and l_2 are the lactate concentrations (mmoles/liter)

TABLE I

Vital Statistics of Eight Subjects Studied

Subject	Age	Sex	Height	Weight*	Per cent‡ ideal body weight	Diabetes§ mellitus
			cm	*kg*		
H. B.	25	F	152	66.4	+20	0
G. F.	23	M	170	54.3	−15	0
R. G.	25	M	169	61.4	−7	0
D. S.	30	M	183	88.6	+13	0
J. G.	25	M	179	88.6	+20	0
G. S.	27	F	163	72.7	+24	0
J. M.	25	M	178	101.4	+30	0
L. F.	24	M	179	68.6	−3	0

* With clothes.

‡ Metropolitan Life Insurance Tables 1959.

§ On the basis of an oral glucose tolerance test (40 g/m^2) or a 2 hr postprandial blood sugar.

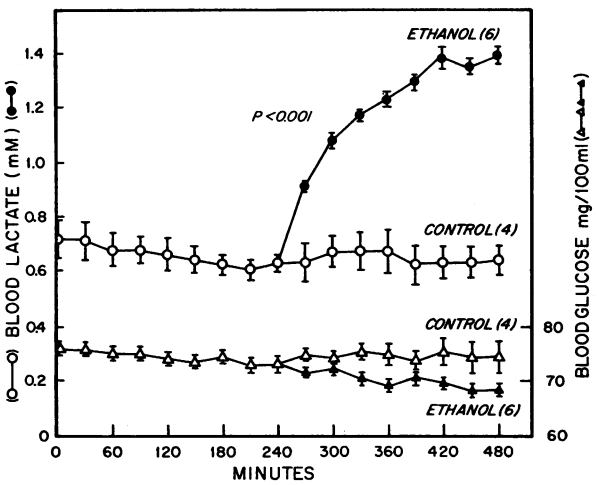

FIGURE 1 Blood lactate and glucose concentrations during the infusion of lactate-^{14}C. From 0 to 240 min, each value is the mean ±SEM for 10 subjects. From 270 to 480 min, the control group value is the mean ±SEM for 4 subjects and the ethanol-treated value is the mean ±SEM for 6 subjects. P = paired comparison analysis.

and SA_1 and SA_2 are the blood lactate specific activity values at times t_1 and t_2; tbw is the total body water and (tbw $\times$ l_1) is the lactate pool at t_1. R_1 and R_2 were corrected for body weight and are expressed in the manuscript as mmoles/kg per hr. Total body water (tbw) for males was derived from the equation tbw = 13.25 + 0.404 × (body weight in kg), and for females from the equation tbw = 11.63 + 0.318 × (body weight) (31).

Bonded alcohol (bourbon, blended whiskey, or Scotch) in water or dietary, carbohydrate-free beverages, was administered orally over a 15 min period from the 5th through the 8th hr of the lactate-U-^{14}C infusion. Each subject received 19.6 g of ethanol initially and 9.8 g hourly thereafter. Ethanol was measured enzymatically (32) and in no subject did its concentration exceed 75 mg/100 ml.

RESULTS

Blood lactate (Figs. 1 and 2). Blood lactate concentrations were determined at 30-min intervals during the infusion of lactate-^{14}C (Fig. 1). The mean blood lactate concentration declined by 20% during the period from 0 to 240 min. The higher initial blood lactate concentration was probably due to anxiety over the study and the stress associated with venipuncture and placement of the venous cannulas, while the subsequent decline was due to final achievement of the basal conditions. In those subjects who did not receive ethanol, the blood lactate concentration declined slightly over the period from 270 to 480 min, but as shown in Table II, the mean blood lactate concentration in those four subjects for the period 0–240 min (0.751 mmole/liter ±0.081) was not significantly different from the mean value for the period 390–480 min (0.637 mmole/liter ±0.054). In the six ethanol studies, the blood lactate concentrations

TABLE II

Comparison of Subjects Not Receiving Ethanol to the Ethanol-Treated Group

Subjects	Ethanol	Blood glucose		Blood lactate		Lactate turnover		Lactate oxidation		
		0–240*	390–480‡	0–240	390–480	150–240	390–480	0–240	270–360‡	390–480
		mg/100 ml		*mmoles/liter*		*mmoles/kg per hr*		*mmoles/kg per hr*		
H. B.	0	90.1	79.4	0.931	0.652	0.868	0.808	0.152	0.164	0.179
G. F.	0	75.2	80.8	0.871	0.788	1.251	0.974	0.199	0.145	0.202
R. G.	0	71.7	74.2	0.686	0.626	1.070	0.912	0.155	0.174	0.178
D. S.	0	76.3	76.0	0.517	0.483	0.582	0.521	0.062	0.072	0.076
Mean	0	78.3	77.6	0.751	0.637	0.942	0.803	0.142	0.139	0.159
±SEM		3.5	1.3	0.081	0.054	0.124	0.088	0.024	0.020	0.024
P§		NS		NS		NS		NS		NS
J. G.	+	75.3	66.2	0.549	1.167	0.804	0.624	0.175	0.166	0.152
G. S.	+	76.3	71.4	0.649	1.178	0.734	0.599	0.257	0.260	0.220
J. M.	+	70.9	69.2	0.563	1.580	0.676	0.621	0.205	0.206	—
L. F.	+	70.6	65.8	0.626	1.241	0.847	0.836	0.161	0.165	0.199
H. B.	+	93.6	84.8	0.526	1.404	0.581	0.503	0.102	0.112	0.112
R. G.	+	71.9	70.8	0.577	1.445	0.921	0.898	0.144	0.144	0.164
Mean		76.4	71.4	0.582	1.339	0.761	0.680	0.174	0.176	0.169
±SEM		3.3	2.6	0.018	0.052	0.046	0.057	0.020	0.019	0.017
P§		<0.02		<0.001		<0.05		NS		NS

* Mean of nine individual values obtained at 30-min intervals during the stated period.
‡ Mean of four individual values obtained at 30-min intervals during the stated period.
§ Paired comparison analysis.

rose promptly and despite the continued administration of ethanol reached a plateau between 360 and 420 min. Though not depicted in Fig. 1, ethanol significantly elevated the blood lactate concentration within 15 min. The mean lactate concentration during ethanol treatment increased approximately 2.5-fold, from 0.582 mmole/liter ±0.018 in the period from 0 to 240 min to 1.339 mmoles/liter ±0.062 in the period from 390 to 480 min ($P < 0.01$).

Blood glucose. The blood glucose concentrations during the study are also depicted in Fig. 1. In those subjects who did not receive ethanol, the glucose concentration during the period 270–480 min remained constant while it decreased slightly in those who received ethanol. There were no significant differences in the mean blood glucose concentrations for the periods 0–240 and 390–480 min in subjects not receiving ethanol Table II). The glucose concentration decreased by 5 mg/100 ml in the ethanol-treated group, from 76.4 ±3.3 in the 0–240 min period to 71.4 ±2.6 mg in the interval from 390 to 480 min ($P < 0.02$).

Lactate turnover. The specific activity-time course curves for lactate, glucose, and CO_2 and for lactate inflow and outflow in representative experiments with and without ethanol are shown in Figs. 2 and 3. The blood lactate concentration and specific activity in the absence of ethanol are shown in Fig. 2 A. The lactate specific activity increased in the period from 120 to 240 min, but in most subjects remained fairly constant thereafter. In those subjects with the highest initial lactate levels, more than 240 min was often required to achieve constant lactate concentrations, and the blood lactate specific activity continued to rise. In other subjects, isotopic and substrate steady state was achieved sooner with the result that it was possible to calculate lactate turnover at earlier intervals. Due to the higher initial lactate concentration, low specific activity values were seen routinely during the early hours of the infusion, despite administration of priming dose. The blood glucose specific activity (Fig. 2 B) increased throughout the entire 8 hr and was greater than could be attributed to the decrease in glucose concentration seen in this subject. The blood glucose concentration decreased by 12% during the period from 240 to 480 min while the glucose specific activity doubled. The same phenomenon was observed in subjects in whom there was no decline in blood glucose. As shown in panel C of Fig. 2, CO_2 specific activity reached a plateau between 240 and 300 min. A slight decline in CO_2 production was often observed during the studies and like the change in lactate concentration was probably due to finally reaching a basal state. Lactate inflow and outflow were always

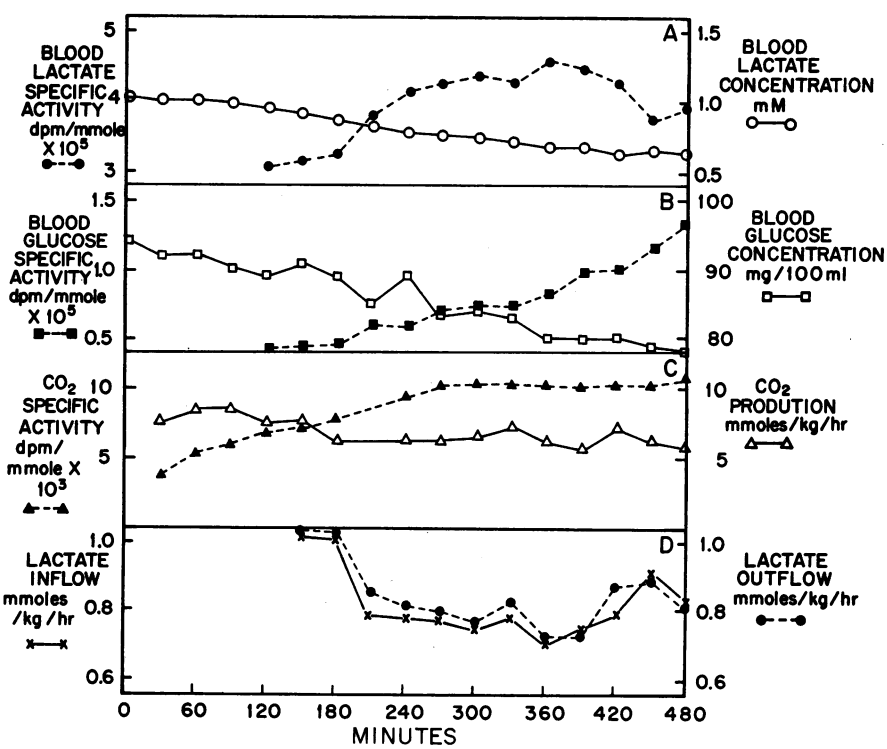

FIGURE 2 Specific activity-time course curves for blood lactate, glucose, and CO_2 and lactate and glucose concentrations during the infusion of lactate-^{14}C in subject H. B.

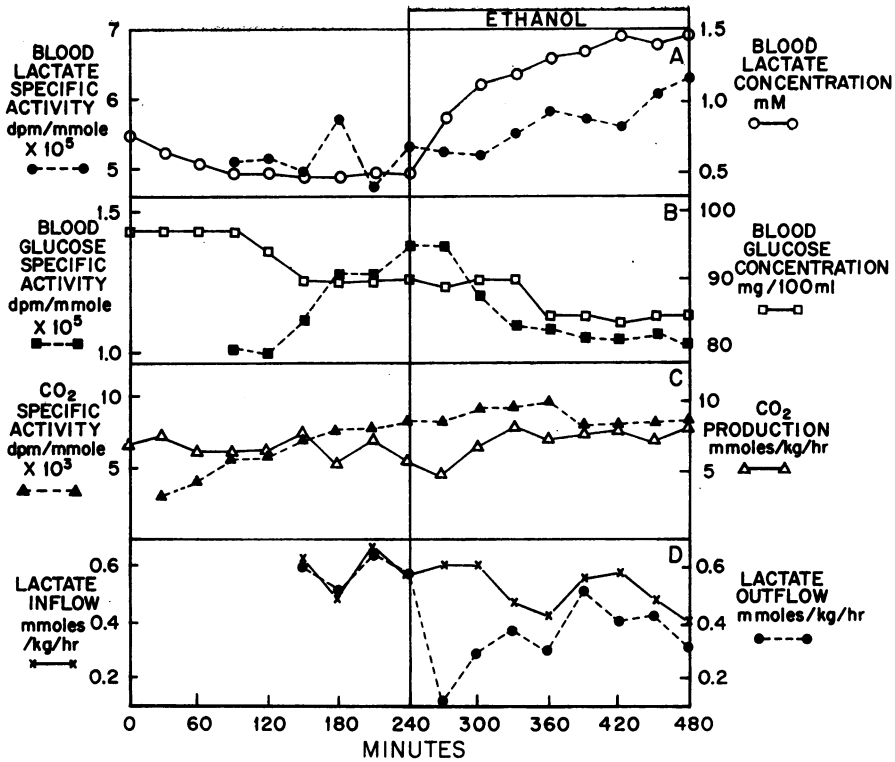

FIGURE 3 Same as Fig. 2. Effect of ethanol.

closely balanced (Fig. 2 D) although the rates were generally higher initially. In general, we found lactate specific activity values and inflow-outflow rates to be more variable than those observed in comparable studies with glucose.

The effect of ethanol on these same parameters is shown in Fig. 3. Despite the 2.5- to 3-fold increase in blood lactate concentration, the lactate specific activity also increased during the period of ethanol administration (Fig. 3 A). This finding occurred in all of the subjects receiving ethanol and indicated that the increase in lactate concentration could not be due to increased lactate production since this would dilute the lactate pool with nonradioactive lactate and lower the specific activity. Calculations of lactate turnover from the infusion rate of tracer and the lactate specific activity, both in the control and ethanol periods (Table II), indicate that ethanol produced a slight but significant decline ($P < 0.05$). As shown in Table II, this decline did not occur in the control group and therefore may be attributable to ethanol. Ethanol inhibited the incorporation of lactate into glucose, as indicated by the fall in blood glucose specific activity (Fig. 3 B). This contrasts with the continued increase observed in glucose specific activity in the absence of ethanol, shown in Fig. 2 B and discussed earlier. No significant changes in the contour of the CO_2 specific activity curve (Fig. 3 C) or in lactate oxidation were produced by ethanol. In Table II lactate oxidation during three different time intervals is shown. While it is possible that ethanol altered lactate oxidation, the changes are not striking. It should be emphasized that the recovery of ^{14}C in CO_2 occurs as a result of direct oxidation of lactate and by oxidation of glucose derived from lactate. Since lactate incorporation into glucose was inhibited by ethanol, the finding of an unchanged rate of $^{14}CO_2$ production could be interpreted to reflect increased direct oxidation of lactate. It is well known that the use of standard isotopic dilution techniques and equations for the circulation of lactate inflow and outflow only apply during steady-state conditions. In the nonsteady state, other relationships must be utilized. In sharp contrast to the close relationship observed between lactate inflow and outflow in the period before 240 min and in the study in which ethanol was not administered, ethanol produced a marked inhibition of lactate outflow, which reached a maximum within the first 30 min (Fig. 3 D). Thereafter, the lactate outflow rate gradually increased and within 360–420 min was again similar to lactate inflow. Both the inhibition of lactate incorporation into glucose and the blood lactate concentration reached plateau

values during the period when lactate inflow-outflow equilibrium was restored.

The mean lactate inflow and outflow rates for 10 studies performed are shown in Fig. 4. In the four studies in which ethanol was not administered (Fig. 4 A), lactate inflow and outflow were balanced throughout the period of infusion. In six studies (Fig. 4 B), lactate inflow and outflow rates were also closely balanced before the administration of ethanol. The mean lactate inflow rate during the interval from 150 to 240 min for these six subjects before ethanol was 0.76 mmoles/kg per hr ±0.07 (SEM), while outflow was 0.78 mmoles/kg per hr ±0.08. After the administration of ethanol, the mean lactate outflow decreased by 50% at 270 min to 0.398 mmoles/kg per hr ±0.150 ($P < 0.01$). Lactate outflow gradually increased over the next 90–120 min until it was again equivalent to inflow. The lactate outflow rates are significantly different from the inflow rates at 270, 300, and 330 min ($P < 0.01$).

The effect of ethanol on lactate incorporation into glucose is shown in Fig. 5. The "per cent glucose turnover derived from lactate" (% G^{lact}) was calculated from the relationship: $(G^{SA} \times 100)/(L^{SA} \times 2)$ in which G^{SA} and L^{SA} are the specific activities of glucose and lactate respectively in disintegrations per minute per millimole. The mean % G^{lact} calculated from the 240 min value in the 10 studies was 14.5 ±1.5 (range 7–19), a value comparable to those previously published (22). To accurately establish the degree of inhibition in % G^{lact} produced by ethanol, it is necessary to compare

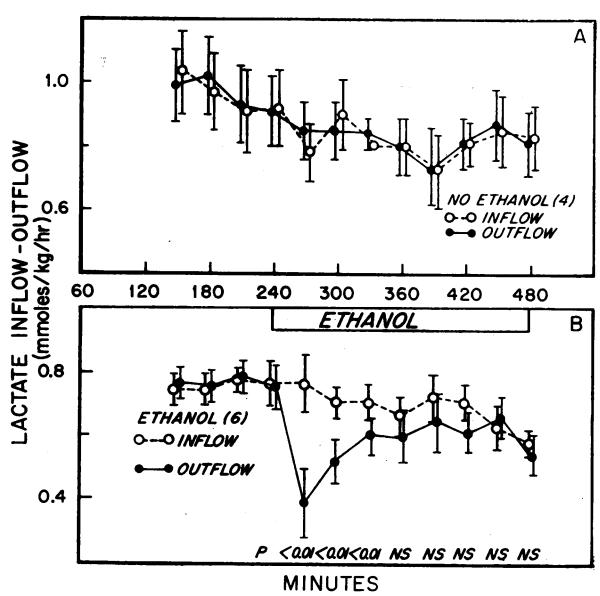

FIGURE 4 Effect of ethanol on lactate inflow-outflow rates. Each value is the mean ±SEM. P = paired comparison analysis.

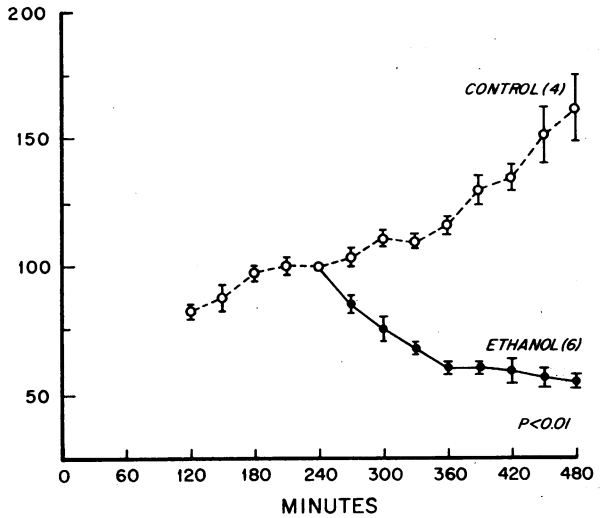

FIGURE 5 Per cent of glucose derived from lactate expressed as per cent of the 240 min value. Each value is the mean ±SEM.

% G^{lact} values obtained during the administration of ethanol with values obtained at comparable times in subjects who did not receive ethanol. To facilitate this comparison, the % G^{lact} values obtained at 30-min intervals during the 8 hr infusion period of lactate-^{14}C were expressed as a per cent of the 240 min value. In those subjects who did not receive ethanol, the glucose specific activity continued to increase and the % G^{lact} at 480 min was 180% of the calculated 240 min value. In those subjects who received ethanol, lactate incorporation into glucose was abruptly inhibited and glucose specific activity not only did not rise but actually declined. Maximum inhibition occurred at approximately 360–420 min and thereafter the glucose specific activity remained constant. The mean value for % G^{lact} at 480 min was 55% of the 240 min value and 30% of the 480 min value for the control group. We believe that the latter value more accurately reflects the true inhibitory effect of ethanol on lactate incorporation into glucose. Despite the restoration of the balance between lactate inflow and outflow between 360 and 420 min, inhibition of lactate incorporation into glucose persisted.

DISCUSSION

It has been suggested that ethanol-induced hyperlacticacidemia is due to the increased availability of NADH in liver which results from the metabolism of ethanol and a subsequent increase in pyruvate conversion to lactate (1–3). However, studies of the effect of ethanol on lactate production by the perfused rat liver (16–18) or on hepatic vein lactate in man (15) do not support the contention that the liver is the source of the lactate.

In isolated rat livers perfused with an artificial buffer containing alanine as substrate, the ratio of lactate to pyruvate in the perfusate is invariably increased by ethanol, but lactate production is not (17, 18). In fact, in these experiments, ethanol decreased lactate and pyruvate production by the isolated perfused rat liver (17, 18), as well as their concentration in liver tissue (18); and the increased lactate to pyruvate ratio was a result of greater decrease in pyruvate than in lactate. In similar studies in which rat livers were perfused with diluted whole blood, ethanol only increased hepatic lactate production by livers from starved rats (16), yet it is well known that ethanol induces hyperlacticacidemia in the fed and fasted state. Furthermore, ethanol not only decreases hepatic lactate production in this system but, as shown by Krebs (20), also inhibits lactate removal and its subsequent incorporation into glucose. Measurements of splanchnic lactate production in humans also indicate that ethanol decreases lactate removal by the liver rather than increasing its production. Tygstrup, Winkler, and Lundquist have shown that hepatic arteriovenous (A-V) differences for lactate become negative with the administration of ethanol (15). In fasting healthy subjects, net uptake of lactate by the liver was 0.16 μmoles/min before and −0.02 μmoles/min after ethanol. On the basis of increased hepatic vein concentrations of lactate, the studies of Mendeloff (10) and of Seligson, Stone, and Nemir (11) have been quoted as supporting the proposal that ethanol increases lactate production by the liver. In neither study, however, were there sufficient data to support such a conclusion. The study of Mendeloff deals with peripheral venous-hepatic venous differences and the results are limited in scope. Furthermore, while Seligson and his coworkers demonstrated that ethanol produced a doubling of hepatic vein lactate concentrations, they did not measure hepatic blood flow or A-V differences so that no conclusions can be reached concerning lactate production rates in either study.

The possibility that ethanol-induced hyperlacticacidemia is due to decreased lactate utilization rather than increased production has not previously been investigated. Measurements of splanchnic lactate production by Tygstrup and coworkers (15) were actually peripheral to their interest in the effect of fructose on ethanol metabolism, and they did not discuss their results in terms of the mechanism of ethanol-induced hyperlacticacidemia. In previous, less precise studies, Lundquist, Tygstrup, Winkler, Mellemgaard, and Munck-Petersen noted that the increase in blood lactate concentration after ethanol could not be caused by increased production of lactate by the liver (14). They suggested that acetate, produced by metabolism of ethanol in the liver, may compete with lactate for utilization in extrahepatic tissues. The recent demonstration that ethanol impairs the disappearance of lactate from the blood after a lactate load (19) or after severe exercise (20) also suggests that decreased lactate removal may be an important factor in the hyperlacticacidemia. The results of the current studies clearly indicate that the elevated blood lactate levels which accompany ethanol administration are due primarily to inhibition of lactate removal. The rise in blood lactate concentration induced by ethanol was not associated with any significant change in the rate of lactate production, but coincided with a decrease in lactate removal and inhibition of lactate incorporation into glucose. Since overall lactate oxidation was not significantly altered by ethanol, the temporal relationship between the change in blood lactate concentration and the inhibition of lactate utilization strongly suggests that decreased lactate incorporation into glucose is at least partially responsible for the hyperlacticacidemia.

It must be emphasized that isotopic techniques, such as those used in these studies, only reflect the net or integrated changes that have occurred in lactate metabolism in the intact organism as a result of ethanol. They do not necessarily identify the sites of these changes or quantitate the extent of the change at any site; however, the concomitant demonstration in these studies that lactate conversion to glucose was decreased obviously implicates the liver. The possibility that ethanol also alters the utilization of lactate by nonhepatic tissues, as previously suggested by Lundquist (14), must also be considered. Although negative A-V differences for lactate across resting limbs indicate net lactate production, there is considerable evidence that skeletal muscle both simultaneously removes lactate from and releases it into the circulation. In both rabbits and man, lactate can be oxidized directly without prior conversion to glucose (33, 34) and may be a significant source of fuel. In normal rabbits 80–90% of a single intravenous dose of lactate-^{14}C is metabolized by direct oxidation and may account for 25–50% of the respired CO_2 (33). Net uptake of lactate by resting skeletal muscle has also been demonstrated in humans during exercise (35). In one study, during leg exercise, lactate was released by the legs in greater quantities and removed by the motionless upper arm (35). As a result of this study, it has been suggested that inactive muscle is an important site of lactate disposal during exercise. The elegant studies of Jorfeldt with the human forearm preparation (34) clearly establish the importance of lactate as a metabolic fuel for skeletal muscle. By the use of regional lactate-^{14}C infusions and conventional catheter techniques, he has demonstrated that the fractional uptake of lactate-^{14}C by the working human forearm was 0.3 and that approximately 40% of the radioactivity of the

lactate-^{14}C extracted was recovered as $^{14}CO_2$. Jorfeldt has discussed the possibility that the concomitant uptake and release of lactate by skeletal muscle is due to the compositional and metabolic heterogeneity of the muscle units. Thus, lactate may be produced primarily by white muscle fibers, which have a large content of glycogen and high glycolytic enzyme activity and be utilized by red muscle fibers, which are structurally and metabolically similar to those of cardiac muscle. In this regard, acetate has been demonstrated to be actively metabolized by the isolated, perfused rat heart (36); and it is possible that inhibition of lactate extraction and/or utilization by skeletal muscle could result from direct competition between acetate, formed during metabolism of ethanol by the liver, and lactate for oxidative metabolism in red muscle fibers.

Certain questions can be raised concerning the accuracy of the lactate inflow-outflow data which relate to the use of peripheral venous blood from a single site for the determination of lactate concentration and specific activity. The calculations of lactate inflow-outflow rates in these experiments is dependent upon estimation of the lactate pool and upon the specific activity in lactate in blood. For the purpose of these studies, we have assumed that the concentrations and specific activity of lactate in peripheral venous blood from a single site is representative of that in the lactate compartment. We have not obtained simultaneous samples of arterial and venous blood from several sites to determine to what extent this assumption is correct. The use of resting forearm venous lactate rather than arterial lactate concentrations could produce a 10–15% overestimation of size of the lactate pool and a commensurate elevation of lactate inflow-outflow rates. Venous blood lactate specific activity would be expected to be lower than arterial values for several reasons. First, muscle iteslf and/or the red blood cells traversing the vascular bed in muscle is responsible for net production of lactate and the negative A-V difference for lactate across muscle. Secondly, as indicated by the studies of Jorfeldt (34) significant quantities of lactate-^{14}C may be extracted by the muscles of the forearm in the face of net lactate production. To the extent that ethanol would increase these processes, the lactate especific activity would be further diluted and calculated rates of lactate inflow would be increased. Thus in a sense, the experimental conditions would favor a mechanism opposite that which was actually observed. We believe, therefore, that the constancy of blood lactate specific activity in peripheral venous blood in the presence of ethanol is strong evidence for the qualitative validity of the observations made in these experiments. However, because of the problems inherent in the use of isotopic dilution techniques and in sampling, it must be emphasized that many of the conclusions reached concerning the quantitative aspects of this study must, at best, be considered tentative.

Although ethanol produced a decrease in lactate conversion to glucose that was significant within 30 min and maximum by 180 min, blood glucose concentrations declined only slightly. It is likely that the fall in blood glucose was masked by mobilization of glycogen since the subjects used in these studies had only fasted for 18 hr before receiving ethanol. These results indicate that ethanol inhibits gluconeogenesis in the nonstarved subject and suggests that clinical hypoglycemia occurs only when hepatic glycogen stores are depleted. Data bearing upon this point are discussed elsewhere (*27*).

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