Effects of Ethanol on Carbohydrate Metabolism in the Elderly

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We have previously reported that in young men, ethanol caused acute insulin resistance, but compensatory insulin secretion prevented deterioration of glucose tolerance (1). In this study, we tested the hypothesis that elderly men, because of their pre-existing insulin resistance and compromised insulin secretory capacity, may experience worsening of their glucose tolerance after ethanol. Nine elderly men (65.7 ± 0.8 yr, BMI 25.8 ± 1.4 kg/m^2) received ethanol (13 mmol/kg for 30 min i.v.) or saline followed 30 min later by i.v. glucose (2.8 mmol/kg for 5 min). To determine the mechanism of the ethanol effect, six of the men underwent euglycemic-hyperinsulinemic (~350 pM) clamping with simultaneous infusion of ethanol or saline. Muscle biopsies were obtained before and 1 and 4 h after insulin infusion. In all nine men, glucose concentrations after i.v. glucose were higher after ethanol than after saline, whereas insulin was the same and glucose tolerance decreased by 23% (K_g 2.41 ± 0.2 vs. 1.86 ± 0.1%/min, $P < 0.01$). Ethanol reduced insulin-stimulated glucose uptake from 40.6 ± 3.1 to 25.6 ± 1.9 μmol · kg^{-1} · min^{-1} (−37%, $P < 0.05$), glucose oxidation from 11.7 ± 1.1 to 7.0 ± 0.7 μmol · kg^{-1} · min^{-1} (−33%, $P < 0.01$), and glucose storage from 28.7 ± 2.4 to 18.6 ± 1.7 μmol · kg^{-1} · min^{-1} (−35%, $P < 0.01$). Ethanol increased muscle lactate concentration from 0.49 ± 0.14 to 1.99 ± 0.99 μmol/mg protein ($P < 0.05$), but had no effects on muscle concentration of free glucose, G-6-P, and citrate concentrations, nor did it affect muscle GS activity. We concluded that modest amounts of ethanol in elderly men impaired glucose oxidation and caused insulin resistance, which because of lack of compensatory insulin secretion, resulted in deterioration of glucose tolerance. *Diabetes* 42:28–34, 1993

We and others (1,2) recently demonstrated that modest amounts of ethanol infused intravenously into healthy young men caused acute insulin resistance. These young subjects were able to maintain normal glucose tolerance by increasing their insulin release to compensate for the ethanol-induced insulin resistance (1). These results suggested that in individuals with impaired insulin secretory reserves, ethanol consumption may result in deterioration of glucose tolerance and occasionally overt diabetes mellitus. Such a group of individuals may be the elderly. Some evidence suggests that the insulin secretory capacity is diminished in elderly human subjects (3,4) and in aging experimental animals (5). In addition, insulin resistance has been demonstrated to increase with aging (6–8). Taken together, it appears likely that compromised insulin secretory capacity and pre-existing insulin resistance may make it difficult for elderly subjects to cope with the additional burden of ethanol-induced insulin resistance. Hence, glucose intolerance or frank diabetes mellitus may ensue after alcohol consumption.

The mechanisms by which ethanol produces acute insulin resistance are not well understood. We have previously shown that ethanol prevented oxidation of an intravenously administered glucose load (1). The inhibitory action of ethanol on CHO oxidation presumably takes place in muscle, where >80% of an intravenous glucose load is metabolized (9). As shown previously, ethanol was rapidly oxidized to acetate (1), which can be taken up by muscle, activated to acetyl-CoA, and oxi-

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CHO, carbohydrate; BMI, body mass index; GS, glycogen synthase; G-6-P, glucose-6-phosphate; IVGTT, intravenous glucose tolerance test; K_g, log of glucose disappearance; G_{Ra}, rate of glucose appearance; G_{Rd}, rate of glucose disappearance; npRQ, nonprotein respiratory quotient; LBF, leg blood flow; UDPG, uridine diphosphate glucose; MANOVA, multiple analysis of variance; NS, no significance; CV, coefficient of variation; HGO, hepatic glucose output; NIDDM, non-insulin-dependent diabetes; ETOH, ethyl alcohol.

TABLE 1
Clinical characteristics of study subjects

Study	*n*	Age (yr)	Height (cm)	Weight (kg)	BMI (kg/m^2)
Clamp plus ETOH	6	67.0 ± 1.8	1.68 ± 0.04	72.03 ± 3.06	25.65 ± 1.04
Clamp plus saline	6	70.3 ± 2.3	1.74 ± 0.04	74.17 ± 3.69	24.41 ± 0.84
IVGTT	9	65.7 ± 0.8	1.70 ± 0.03	73.75 ± 3.74	25.75 ± 1.41

dized in the tricarboxylic acid cycle. An increase in acetyl-CoA is known to inhibit pyruvate dehydrogenase and thus glucose oxidation (10).

It is, however, unlikely that inhibition of glucose oxidation alone could fully explain ethanol-induced insulin resistance, because glucose, which cannot be oxidized, could be shunted into the formation of glycogen or lactate. We suspect, therefore, that ethanol inhibits glycogen synthesis and perhaps also glycolysis at a locus above pyruvate dehydrogenase.

To test whether modest amounts of ethanol will cause insulin resistance and deterioration of glucose tolerance, we compared the effects of ethanol and saline on intravenous glucose tolerance and insulin sensitivity (measured with the euglycemic-hyperinsulinemic clamp technique) in elderly men. To investigate the mechanisms of ethanol action, we determined glucose turnover rates (with 6,6-D_2-glucose), rates of whole body CHO oxidation (with indirect calorimetry), and concentrations of key substrates including free glucose, G-6-P, lactate, and citrate and the activity of GS, the key enzyme of glycogen formation, in muscle biopsy samples obtained at different times from these elderly men.

RESEARCH DESIGN AND METHODS

Twelve healthy, normal-weight, elderly men were studied. Their ages, weights, and heights are shown in Table 1. None of the subjects had a family history of diabetes or other endocrine disorders, and none were taking any medications. Their weights were stable for at least 2 mo, and their diets contained a minimum of 250 g/day of CHO for at least 2 days before the study. Informed written consent was obtained after explanation of the nature, purpose, and potential risks of each study. The study protocol was approved by the Temple University Institutional Review Board for Human Research. Studies were performed in the General Clinical Research Center of Temple University Hospital and began between 0700 and 0800 after an overnight fast. The subjects were studied reclining in bed. A short polyethylene catheter was inserted into an antecubital vein for infusion of all test substances. A second catheter was inserted into a contralateral forearm vein for blood sampling. This arm was kept at 70°C with a heating blanket to arterialize venous blood.

Study 1: IVGTTs. Nine elderly subjects were studied twice in random order with and without ethanol. A minimum of 10 days was allowed between ethanol and control (saline) studies. Ethanol and control studies were identical except for the substitution of normal saline for ethanol in control studies. After a 30-min baseline period, alcohol (13 mmol/kg, USP, Abbott, North Chicago, IL) diluted in normal saline to make a 10% solution, was infused intravenously for 30 min (−60 to −30 min). Glucose (2.8 mmol/kg) was infused intravenously for 5 min starting at 0 min. Arterialized venous blood was collected for determination of plasma glucose concentration every 10 min for the next hour and at 30-min intervals thereafter. Arterialized venous blood samples for determination of glucose, insulin, and ethanol concentrations were obtained at 30-min intervals. K_g was calculated by least-squares analysis from the decline of plasma glucose levels between 10 and 60 min after the glucose infusion.

Study 2: hyperinsulinemic-euglycemic clamps. Six subjects were studied with and six without ethanol in random order. Regular human insulin (Humulin R, Lilly, Indianapolis, IN) was infused intravenously (6 pmol · kg^{-1} · min^{-1}) for 4 h. Glucose concentrations were clamped at ~5 mM by a feedback-controlled glucose infusion (1). At 0 min, ethanol (8 mmol/kg) or saline was infused intravenously over a 15-min period. Thereafter, an infusion of ethanol (2.17 mmol/kg for 3.75 h) or saline was delivered to maintain steady-state ethanol concentrations during ethanol infusions until the completion of the study. Arterialized venous blood samples for determination of ethanol and insulin were obtained at 30-min intervals.

Glucose turnover. Glucose turnover was determined with 6,6-D_2-glucose. The tracer infusion was started 2 h before initiation of the clamp to assure isotope equilibration. Plasma glucose was isolated from blood for determination of isotope enrichment as described (11) with a gas chromatograph–mass spectrometer (model 4610-B, Finnigan MAT, San Jose, CA). G_{Ra} and G_{Rd} were calculated from the isotope enrichment before (−30–0 min) and during the 4th h of the clamp (180–240 min) by using Steele's equation for non–steady-state conditions (12). Underestimation of G_{Ra} during hyperinsulinemia was avoided by adding 6,6-D_2-glucose (6.9 mmol/100 ml of a 50% dextrose solution) to the unlabeled glucose infused to maintain euglycemia (13).

Indirect calorimetry. Respiratory gas exchange rates were determined, as previously described, before (−30 and 0 min) and at the end of the 4-h ethanol or saline infusions (210 and 240 min) with a metabolic measurement cart (Beckman, Palo Alto, CA; 14). Rates of protein oxidation were estimated from urinary nitrogen excretion after correction for changes in urea nitrogen pool size (15). Rates of protein oxidation were used to determine npRQ. Rates of CHO and fat oxidation were determined with the npRQ tables of Lusk, which are based on an npRQ of 0.707 for 100% fat and 1.000 for 100% CHO oxidation. Based on our previous finding of complete

oxidation of metabolized ethanol to CO_2 and H_2O (1), these respiratory exchange data were corrected by subtracting the volumes of O_2 consumed and CO_2 produced during ethanol oxidation (complete oxidation of 1 mol of ethanol consumes 3 mol of O_2 and produces 2 mol of CO_2).

Rates of CHO storage were obtained by subtracting rates of CHO oxidation from G_{Rd}.

LBF. LBF was determined every 30 min by venous occlusion plethysmography with a mercury strain-gauge apparatus (Model EC-5R, Hokanson, Issaquah, WA; 16). Two minutes before blood-flow determination, circulation to the foot was interrupted by inflating a cuff around the ankle to a suprasystolic pressure.

Muscle biopsies and extractions. Biopsies were obtained before and 1 and 4 h after the start of the clamp, from the lateral aspect of the vastus lateralis muscle, ~15 cm above the patella, from all 12 subjects in study 2. The skin was cleaned with Betadine and anesthetized with 1% lidocaine without epinephrine. After an incision (~2.5 cm) was made through the skin, subcutaneous tissue, and fascia; ~150 mg of muscle was mobilized and excised. The muscle was dropped immediately into isopentane and kept at its freezing point (−160°C) by liquid nitrogen. The frozen muscle was stored at −80°C until it was aliquoted into two separate portions. One portion was extracted with fluoride buffer according to Hagg et al. (17) for measurement of glycogen synthase. The second portion was extracted with perchloric acid according to Allred and Guy (18) for measurement of free glucose, lactate, G-6-P, and citrate.

GS assay. GS was assayed by a modification of the method of Thomas et al. (19). Reactions were started by addition of 30 μl aliquots of the muscle extract to 60 μl of a reaction mixture containing 20 mM EDTA, 25 mM sodium fluoride, 50 mM Tris-HCl, 1% glycogen, 0.7 μCi [U-^{14}C]UDPG, 0.3 mM UDPG, and 0–10 mM G-6-P. The reaction was terminated after 15 min by precipitating 75 μl aliquots of the reaction mixture on 2- × 2-cm squares of filter paper, which were dropped into cold 66% ethanol, washed, dried, and counted. GS activity was calculated as micromoles of UDPG incorporated into glycogen per minute per milligram of protein. Results are expressed as V_{max} of GS determined with 10 mM G-6-P and 0.3 mM UDPG and as GS fractional velocity, i.e., the activity of GS at 0.1 mM G-6-P divided by the activity at 10 mM G-6-P. This is an indicator of the active form of GS and believed to be a sensitive parameter of in vivo GS activity (20).

Metabolite assays. Citrate was assayed in the perchloric acid muscle extract in a coupled end-point assay with citrate lyase and malate dehydrogenase (21). G-6-P was assayed in a coupled end-point assay with hexokinase and G-6-P dehydrogenase (22). Glucose and lactate were measured enzymatically (23,24).

Analytical procedures. Plasma glucose was measured with a glucose analyzer (Beckman Instruments, Palo Alto, CA). Serum insulin (25) was determined by radioimmunoassay. Plasma calecholamines were measured with a radiometric assay (26). Blood urea nitrogen (27) was measured colorimetrically. Urinary nitrogen was measured by the method of Kjeldahl (28). Plasma and urine ethanol were determined with a kit (Sigma, St. Louis, MO), as was blood acetate (Boehringer Mannheim, Elkhart, IN). For the determination of acetate (29) by enzymatic analysis, blood samples were immediately deproteinized and neutralized with perchloric acid/KOH and metaphosphoric acid/KOH, respectively, before assay.

Ethanol disappearance and metabolism. The rate of ethanol disappearance from the blood was calculated with the following formula: ethanol disappearance = $B \times P \times \gamma$, where B is the change in plasma ethanol concentration (mM) obtained from the linear part of the plasma ethanol curve; P is the body weight (kg) × 1.06 to correct for differences in density between blood and water; and γ is the coefficient of distribution for ethanol (and acetate; 30).

Ethanol metabolism was determined according to the following formula: ethanol metabolized = ethanol disappeared from blood − ethanol excreted in urine and breath − ethanol present in plasma as acetate.

Statistical analysis. All data are means ± SE. Statistical significance was assessed using MANOVA and two-tailed paired Student's *t* test when appropriate.

RESULTS

Effects of ethanol on intravenous glucose tolerance. Ethanol was infused at a rate of 13 mmol/kg for 30 min starting 1 h before the glucose infusion. At the beginning of the glucose infusion (2.8 mmol/kg over 5 min), plasma ethanol concentration was 17.6 ± 1.6 mM (Fig. 1). Subsequently, it declined at a constant rate and reached 10.5 ± 0.8 mM 2 h later. By comparison, impairment of physical coordination, slurring of speech, and loss of judgment are usually seen only with concentrations >22 mM.

Plasma glucose concentrations were significantly higher with than without ethanol. The K_g decreased from 2.41 ± 0.2 without ethanol to 1.86 ± 0.1 with ethanol (−23%, $P < 0.01$).

Serum insulin concentrations rose from 52 ± 10 and 46 ± 9 pM before, to peaks of 281 ± 62 and 250 ± 103 pM (NS) 10 min after intravenous glucose in saline and ethanol experiments, respectively. A second insulin peak occurred ~1 h later, when insulin concentrations were significantly higher during ethanol than during saline infusions (254 ± 50 vs. 160 ± 24 pM, $P < 0.05$). Neither insulin nor ethanol had statistically significant effects on plasma catecholamine concentrations. Epinephrine concentrations were 180 ± 44 and 175 ± 27 pM before and 240 ± 147 and 360 ± 109 pM after 4 h of insulin and insulin plus ethanol, respectively. Corresponding values for norepinephrine were 1.52 ± 0.21 and 1.67 ± 0.21 nM before and 1.67 ± 0.38 and 1.83 ± 0.25 nM after insulin.

Effects of ethanol on glucose uptake, oxidation, storage, and HGO. To determine the cause for the ethanol-induced deterioration of intravenous glucose tolerance, we performed euglycemic-hyperinsulinemic clamps with and without ethanol in these elderly men (Fig. 2). Plasma insulin concentrations were raised approximately seven-

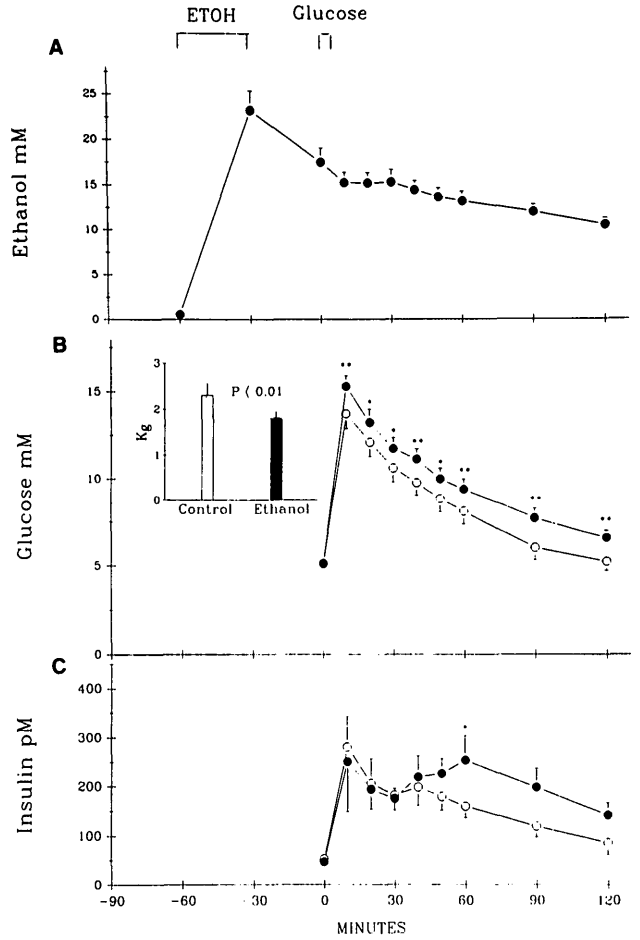

FIG. 1. Effects of ethanol on i.v. glucose tolerance. Plasma ethanol concentrations after ethanol infusion (13 mmol/kg for 30 min) in 9 elderly men (*A*). Effects of ethanol (●) and saline (○) on glucose concentration after intravenous glucose (2.8 mmol/kg for 5 min). *$P < 0.05$, **$P < 0.01$, $n = 9$. Insert shows rates of glucose disappearance from plasma (K_g, in %/min) (*B*). Effects of ethanol and saline on serum insulin concentrations after intravenous glucose. *$P < 0.05$, $n = 9$.

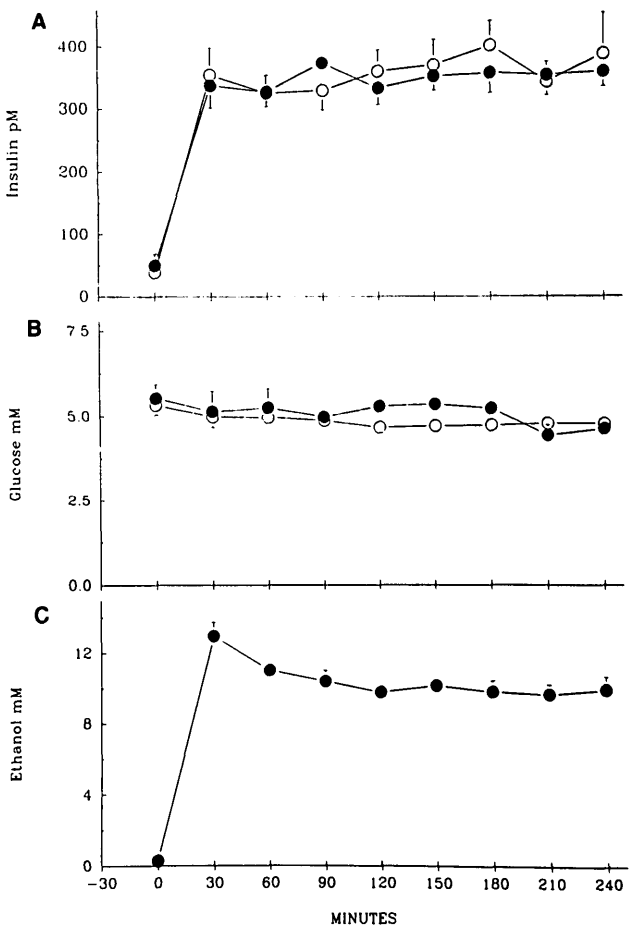

FIG. 2. Euglycemic-hyperinsulinemic clamps. Serum insulin concentrations (*A*), glucose concentrations (*B*), and ethanol concentrations (*C*) in elderly men during euglycemic-hyperinsulinemic clamping with infusion of ethanol (8 mmol/kg, ●), $n = 6$, or saline (○, $n = 6$).

fold from ~47 pM to a plateau of ~350 pM by infusion of insulin (6 pmol · kg^{-1} · min^{-1}). Plasma glucose concentrations were clamped at ~4.9 ± 0.7 mM (CV 10.2%). Plasma ethanol concentrations plateaued at ~10 mM.

Insulin increased G_{Rd} from 9.5 ± 0.4 (preclamp) to 40.6 ± 3.1 μmol · kg^{-1} · min^{-1} during the 4th h of the clamp (427%; $P < 0.001$). Ethanol inhibited this increase by 37% (from 40.6 ± 3.1 to 25.6 ± 1.9 μmol · kg^{-1} · min^{-1}, $P < 0.05$; Fig. 3).

Insulin increased glucose oxidation from 4.6 ± 0.7 (at 0 min) to 11.7 ± 1.1 μmol · kg^{-1} · min^{-1} during the last hour of the clamp (253%; $P < 0.01$). Ethanol inhibited this increase by 33% (from 11.7 ± 1.1 to 7.0 ± 0.7 μmol · kg^{-1} · min^{-1}, $P < 0.01$).

Insulin increased glucose storage from 5.4 ± 0.7 to 28.6 ± 2.4 μmol · kg^{-1} · min^{-1} (531%; $P < 0.01$). Ethanol inhibited this increase by 35% (from 28.7 ± 2.4 to 18.6 ± 1.7 μmol · kg^{-1} · min^{-1}, $P < 0.01$). Insulin decreased HGO by 80% from 8.83 ± 0.3 to 1.77 ± 0.37 μmol · kg^{-1} · min^{-1} with ethanol ($P < 0.01$) and by 82% from 9.28 to 1.67 μmol · kg^{1} · min^{1} with saline ($P < 0.01$).

LBF. During insulin infusion, LBF rose from 4.0 ± 0.2 to 5.7 ± 1.1 ml · 100 ml of leg tissue^{-1} · min^{-1} ($P < 0.05$). During insulin plus ethanol infusion, LBF increased from 3.7 ± 0.3 to 5.4 ± 0.4 ml · 100 ml of leg tissue^{-1} · min^{-1} ($P < 0.08$). These data show that ethanol had no statistically significant effect on the insulin-stimulated increase in LBF.

Effect of ethanol on muscle GS activity and metabolic intermediates. Neither insulin nor ethanol affected GS V_{max} (measured at 0.3 mM UDPG and 10 mM G-6-P; Table 2). GS fractional velocity (0.1/10 mM G-6-P), however, increased from 0.16 ± 0.03 to 0.255 ± 0.028 ($P < 0.05$) with insulin. This increase was unaffected by ethanol.

Neither insulin nor ethanol had significant effects on muscle concentrations of free glucose, G-6-P, and citrate, whereas ethanol significantly increased muscle lactate concentration from 0.30 ± 0.04 to 0.64 ± 0.14 μmol · mg protein^{-1} at 1 h and from 0.25 ± 0.11 to 1.99 ± 0.99 μmol · mg protein^{-1} at 4 h ($P < 0.05$).

DISCUSSION

Ethanol is the most widely used drug in western industrialized countries. In addition, it is an excellent metabolic

TABLE 2
Muscle biopsies

	Preclamp	Postclamp 1 h	Postclamp 4 h
Glycogen synthase			
V_{max} (nmol/mg protein)			
Control	0.054 ± 0.009	0.034 ± 0.003*	0.062 ± 0.014
		NS	NS
ETOH		0.031 ± 0.006	0.049 ± 0.010
Fractional velocity (0.1/10 mM G-6-P)			
Control	0.16 ± 0.03	0.275 ± 0.078	0.255 ± 0.028*
		NS	NS
ETOH		0.139 ± 0.034	0.313 ± 0.038†
Glucose (μmol/mg protein)			
Control	0.10 ± 0.03	0.21 ± 0.04	0.10 ± 0.03
		NS	NS
ETOH		0.12 ± 0.05	0.14 ± 0.05
G-6-P (μmol/mg protein)			
Control	0.018 ± 0.002	0.017 ± 0.007	0.019 ± 0.007
		NS	NS
ETOH		0.033 ± 0.019	0.022 ± 0.012
Lactate (μmol/mg protein)			
Control	0.49 ± 0.14	0.30 ± 0.04	0.25 ± 0.11
		$P < 0.05$	$P < 0.05$
ETOH		0.64 ± 0.14	1.99 ± 0.99*
Citrate (μmol/mg protein)			
Control	0.017 ± 0.002	0.013 ± 0.002	0.013 ± 0.002
		NS	NS
ETOH		0.017 ± 0.006	0.016 ± 0.003

*$P < 0.05$ compared with preclamp values.
†$P < 0.01$ compared with preclamp values.

fuel and can replace glucose as substrate for oxidation, causing acute insulin resistance even at relatively low blood levels (1). We have previously shown that young healthy men were able to compensate for the ethanol-induced insulin resistance by compensatory secretion of insulin, thus avoiding deterioration of their glucose tolerance (1). In this study, we found that elderly men, given comparable amounts of ethanol as young men, experienced a decrease in glucose tolerance. This decline in glucose tolerance had several causes: *1*) insulin resistance, as determined by G_{Rd}, was increased by ~50% in the elderly compared with young men (1); *2*) ethanol increased the pre-existing insulin resistance even more; and *3*) the elderly were unable to generate sufficient compensatory insulin responses. The diabetogenic effect of ethanol was acute and was seen in all nine elderly men tested. In none, however, did K_g decrease into the diabetic range. This suggests that occasional modest consumption of alcohol would probably remain metabolically inconsequential in most nonobese and active elderly men. On the other hand, many elderly men are obese to various degrees, relatively inactive, and known to be prone to developing NIDDM (6). Further depression of their glucose tolerance by repeated consumption of even modest amounts of alcohol may lead to glucose intolerance. It needs to be emphasized that we have only studied acute effects of ethanol and thus can only speculate about its long-term effects. On the other hand, the diabetogenic effects of chronic ethanol abuse are elicited by the development of alcoholic diabetes in some young and old subjects who consume alcohol excessively for prolonged periods of time and in whom the diabetes disappears with ethanol abstinence (31,32).

This anti-insulin action of ethanol presumably takes place in muscle, where >80% of intravenous infused glucose is metabolized (9). Of interest, in this respect, was the finding that ethanol did not interfere with suppression of HGO by insulin, which decreased ~80% in ethanol and control studies (Fig. 3). In this study, decreases in isotope enrichment during hyperinsulinemia, which in the past has resulted in serious underestimations of glucose turnover rates, were avoided by adding labeled glucose to the unlabeled glucose infused to maintain euglycemia (13).

We have attempted to determine the mechanism by which ethanol decreased insulin-stimulated glucose uptake in elderly men. Assuming that LBF was representative of blood flow to muscle in general, we could exclude differences in blood flow as a cause, because ethanol had no effect on insulin-stimulated blood flow. On the other hand, ethanol strongly inhibited (by 33%) insulin-stimulated rates of glucose oxidation, confirming similar observations in young men (1). Concerning the mechanism of this action, a >100-fold increase in plasma

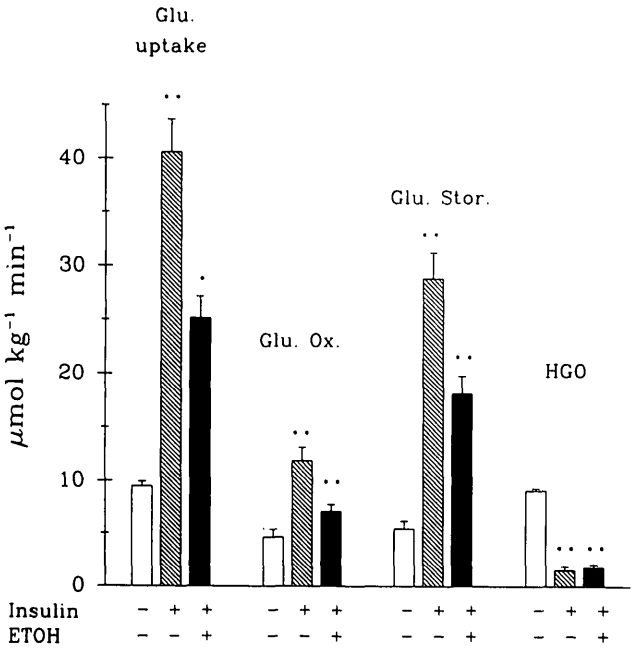

FIG. 3. Effects of ethanol on glucose metabolism. Glucose uptake (Glu. uptake), glucose oxidation (Glu. Ox.), glucose storage (Glu. Stor.), and HGO during basal state (□) in response to insulin (▨) and in response to insulin plus ethanol (■), $n = 6$. Statistical analysis: basal vs. insulin, $P < 0.01$ for glucose uptake, glucose oxidation, glucose storage, and HGO. Insulin vs. insulin plus ethanol, $P < 0.05$ for glucose uptake; $P < 0.01$ for glucose oxidation; and $P < 0.01$ for glucose storage. Basal vs. insulin plus ethanol, $P < 0.01$ for HGO.

acetate levels was observed (from 0.06 to 7.17 mM) after ethanol (this study). Acetate can be activated rapidly to acetyl-CoA, the accumulation of which is known to inhibit pyruvate dehydrogenase and thus pyruvate (glucose) oxidation (10). The observation that muscle content of lactate increased significantly after ethanol was entirely compatible with this concept.

Inhibition of glucose oxidation, however, cannot be the sole explanation for the ethanol-mediated insulin resistance, because glucose that cannot be oxidized can enter nonoxidative pathways such as glycogen synthesis or formation of three carbon compounds (essentially lactate). This, however, did not seem to have occurred in our studies. Muscle GS, the rate-controlling enzyme for glycogen formation, was unaffected by ethanol at the time when glucose uptake was significantly reduced (at 4 h; Table 2). Hence, ethanol did not appear to have interfered with glycogen synthesis. This conclusion, however, needs to be qualified for several reasons. First, muscle glycogen content itself was not measured because of the limited muscle biopsy sample size. Second, GS activity may not have correctly reflected in vivo rates of glycogen synthesis because it was measured at a UDPG concentration of 0.3 mM, which may have been higher than the actual UDPG concentration. Furthermore, the muscle biopsies provided no indication that glycolytic rates were significantly inhibited by ethanol. If a reduction in glycolytic rate had occurred, muscle lactate concentration should have decreased, whereas in fact, lactate increased significantly. Lastly, inhibition of glycolysis would probably have resulted in an increase in muscle G-6-P and free glucose concentrations, neither of which rose. Therefore, we feel that our data are most compatible with an ethanol-mediated defect involving glucose uptake into muscle, which developed during the later parts of the 4-h clamp studies. This interpretation was supported by the finding that ethanol reduced glucose uptake, oxidation, and storage rates to the same extent (37, 33, and 35%, respectively).

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