Lack of antilipolytic effect of lactate in subcutaneous abdominal adipose tissue during exercise

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Lack of antilipolytic effect of lactate in subcutaneous abdominal adipose tissue during exercise. J. Appl. Physiol. 86(6): 1800–1804, 1999.—The purpose of our study was to evaluate the potential inhibition of adipose tissue mobilization by lactate. Eight male subjects (age, 26.25 ± 1.75 yr) in good physical condition (maximal oxygen uptake, 59.87 ± 2.77 ml·kg⁻¹·min⁻¹; %body fat, 10.15 ± 0.89%) participated in this study. For each subject, two microdialysis probes were inserted into abdominal subcutaneous tissue. Lactate (16 mM) was perfused via one of the probes while physiological saline was only perfused via the other, both at a flow rate of 2.5 µl/min. In both probes, ethanol was also perfused for adipose tissue blood flow estimation. Dialysates were collected every 10 min during rest (30 min), exercise at 50% maximal oxygen consumption (120 min), and recovery (30 min) for the measurement of glycerol concentration. During exercise, glycerol increased significantly in both probes. However, no differences in glycerol level and ethanol extraction were observed between the lactate and control probes. These findings suggest that lactate does not impair subcutaneous abdominal adipose tissue mobilization during exercise.

adipose tissue lipolysis; lipid mobilization; microdialysis

DURING LOW-INTENSITY EXERCISE, lipids are the main source of energy for muscle contraction (21). However, as exercise intensity increases, the proportion of energy derived from lipid oxidation decreases as does free fatty acid (FFA) mobilization (6). Many factors are held responsible for this reduction of FFA mobilization. Lower plasma albumin availability for FFA transport and lower adipose tissue blood flow both favor FFA reesterification over mobilization (8, 9). Higher plasma lactate is also suspected to inhibit adipose tissue mobilization during high-intensity exercise (7). In the literature, this hypothesis is supported most often by three papers (5, 15, 16).

Fredholm (15) perfused dog subcutaneous abdominal fat pads with high lactate concentrations and found a decrease of FFA output from the fat pad, but this was not the case for glycerol. Lactate at a concentration as low as 3 mM was shown to inhibit norepinephrine-induced glycerol and FFA outflow in vitro from the rat epididymal adipocytes (3).

Isserukz and Miller (16) reported a significant inverse correlation between plasma FFA and lactate levels during exercise in the dog. In the same paper, arterial infusion at rest resulted in lower plasma FFA levels. These authors concluded that “the decrease of FFA is not caused by the work itself but by the accumulation of lactic acid during exercise.” In the pancreatectomized or normal dog at rest, lactate infusions raising plasma lactate to 6–12 mM were also reported to reduce FFA rate of appearance (20).

Finally a study performed in humans used “whole body” perfusion of lactate and plasma sampling of FFA or glycerol as an index of lipid mobilization from adipose tissue during exercise (5). Lactate infusion reduced the exercise-induced increase of FFA and glycerol (5).

On the basis of these often-cited papers (5, 15, 16), it seems unequivocal that lactate is a regulator of adipose tissue lipolysis at rest and during exercise. However, some papers do not support such a role or at the least modify it. One in vitro experiment showed no lactate-induced decrease of lipolysis with a lactate level of 3 mM in either rat parametrial fat or in chicken mesenteric white fat cells (13). Another in vitro study demonstrated a lower lipolysis only at a lactate level as high as 16 mM in isolated human superficial abdominal adipocytes (12). In vivo, such a lactate level would be observed only after supramaximal exercise (6).

Other rarely cited papers conclude that lactate infusions at rest (13, 17) and during exercise (1), resulting in plasma lactate between 2.91 and 5 mM, does not reduce FFA mobilization.

The purpose of our experiment was then to verify the hypothesis that lactate decreases exercise-induced lipid mobilization. We used microdialysis coupled with ethanol extraction to 1) study local lipolysis in superficial abdominal adipose tissue, 2) locally increase lactate levels, and 3) estimate local blood flow variations in the adipose tissue studied.

METHODS

Subjects. Eight male subjects (age, 26.25 ± 1.75 yr; weight, 73.8 ± 2 kg) participated in the study after giving their informed consent. The project was approved by the Université du Québec à Trois-Rivières institutional ethics committee. The subjects were in good physical condition (maximal oxygen consumption (VO₂max), 59.87 ± 2.77 ml·kg⁻¹·min⁻¹; %body fat, 10.15 ± 0.89%). They had to come twice to the laboratory, once for the direct measurement of VO₂max (Vacumed, Ventura, CA) on a cycle ergometer (Lode, Belgium). One week later, they came back for the experimental
Exercise protocol. They were submitted to a 120-min exercise protocol at 50% \(\dot{V}O_2\text{max}\), corresponding to a workload of \(150.00 \pm 4.23\) W on the cycle ergometer. They reported to the laboratory at 7:00 AM on the day of the experiment.

Adipose tissue microdialysis. Under sterile conditions and by using xylocaine without epinephrine for local anesthesia, two microdialysis probes with a dialysis membrane of 31.4 mm\(^2\) (length, 10 mm; diameter, 0.5 mm) and a 20,000-molecular weight cutoff (CMA/20 microdialysis probe, Carnegie, Stockholm, Sweden) were inserted bilaterally at 10 cm beside the umbilicus. One of the probes was perfused with sterile physiological saline and the other with a lactate solution (16 mM). Four subjects had the lactate probe inserted on the right side and four on the left side. The perfusion rate was 2.5 µl/min by using an infusion pump (model 22, Harvard, South Natick, MA). This rate was chosen following our in vivo studies of recovery rates (11). At a perfusion rate of 2.5 µl/min, the recovery rate is 41.5% (11).

The in vitro lactate delivery rate of the probe was evaluated at a perfusion rate of 2.5 µl/min in various volumes of saline (250, 500, 1,000, and 1,500 µl) after 1, 2, and 3 h of perfusion on the basis of an estimated interstitial volume of 250 µl in the vicinity of the probe (4).

The time course of dialysate sampling was as follows. After insertion of the probes, perfusion was performed for 60 min to reach equilibrium. After this stabilization period, dialysate fractions were collected every 10 min for 30 min of preexercise rest, 120 min of exercise, and 30 min of recovery. After collection, the fractions were kept at -20°C until analysis.

Blood sampling. From a catheter inserted in a right antecubital vein, blood was sampled after 20 and 30 min of rest; at 30, 60, 90, and 120 min of exercise; and 10 and 30 min of recovery. Immediately thereafter, the samples were centrifuged and plasma separated and kept at -80°C until assayed.

Assays. Glycerol from plasma and dialysates was measured with a bioluminescence technique (10, 19). Dialysate ethanol was measured by colorimetric assay. Plasma glucose and lactate were measured with colorimetric commercial kits (Sigma Chemical, St. Louis, MO), as was FFA (Boehringer-Mannheim). Plasma insulin was measured with a radioimmunoassay kit (Immunocorp, Montreal, PQ) and plasma catecholamine with a radioenzymatic kit (Amersham).

Statistics. Data are expressed as means \(\pm\) SE. Each unit of the dialysate ethanol is the percentage of ethanol recovered in the dialysate (outflow/inflow \(\times 100\)). Two-way ANOVA for repeated measures was used. When \(P < 0.05\) was obtained, the post hoc Student-Newman-Keuls test was applied to localize significant differences.

RESULTS

Lactate delivery. During the in vitro experiment, estimated interstitial lactate deliveries were 1.15 \(\pm\) 0.08, 3.77 \(\pm\) 0.37, and 6.39 \(\pm\) 0.51 mM after 1, 2, and 3 h of lactate perfusion corresponding, respectively, to the beginning of rest and 30 and 90 min of exercise.

Dialysate glycerol. At rest, the glycerol level was similar in the dialysate of both probes (40.2 \(\pm\) 6.1 vs. 43.1 \(\pm\) 5.0 µM for control and lactate, respectively). In both probes, it increased steadily from the beginning until the end of exercise. This increase was statistically significant after 40 min of exercise in the control (92.6 \(\pm\) 11.4 µM) and lactate probes (79.7 \(\pm\) 9.23 µM) (Fig. 1). Considering the relative rate of recovery for glycerol (41.5%), these values are 223.1 \(\pm\) 27.5 and 192.0 \(\pm\) 22.2 µM, respectively (11). During exercise recovery, glycerol in the dialysate decreased significantly after 20 and 10 min, respectively, for the control and lactate probes.

Ethanol extraction. At rest and during exercise, ethanol extraction was not different between the control and lactate probes (Fig. 1).

Plasma hormones. Plasma epinephrine and norepinephrine increased significantly from rest to exercise.
and returned to baseline at the end of recovery (Fig. 2). Plasma insulin decreased steadily during exercise (Fig. 2), reaching statistical significance after 30 min of exercise (Fig. 2).

Plasma substrates. Plasma FFAs rose gradually from rest to become significant after 60 min of exercise (0.19 ± 0.03 mM) (Fig. 3). However, the highest FFA levels were reached after 10 min of recovery (1.61 ± 0.15 mM). A similar pattern was observed for plasma glycerol, rising from 35.7 ± 3.11 mM at rest to 155.7 ± 34.4 mM after 60 min of exercise (Fig. 3). The highest plasma glycerol levels were recorded after 120 min of exercise.

Plasma lactate reached significantly higher levels only after 30 min of exercise. Otherwise, lactate levels were never significantly higher than at rest (Fig. 4). Plasma glucose levels were significantly lower after 30, 90, and 120 min of exercise (Fig. 4).

DISCUSSION

In the present study, we tested the hypothesis that lactate could exert a direct inhibitory effect on lipolysis in abdominal adipose tissue. We used two microdialysis probes to measure variations of interstitial glycerol as an index of adipose tissue mobilization. In parallel, one of the microdialysis probes was used to selectively
tissues each possess different physiological properties. Adipose tissue can be heterogenous in the same individual. Various adipose tissues can be confounding because it is quite difficult to standardize hormonal and metabolic influences on adipose tissue studied.

Our results indicate that lactate did not inhibit abdominal adipose tissue mobilization during exercise. We estimated that ~3.77 mM of lactate leaked around the adipocytes studied after 2 h of perfusion, i.e., at 30 min of the exercise period, and that 6.39 mM leaked 30 min before the end of exercise. If we add the endogenous plasma lactate level (1.92 and 1.54 mM at 30 and 90 min of exercise, respectively) to these values, we obtain some data points that are typical of intense exercise (6.92 and 7.93 mM) applied to adipocytes. We are thus confident that our negative results are not a consequence of failure to deliver lactate in adipose tissue.

Furthermore, the microdialysis technique we used has demonstrated its ability to detect inhibition of adipose tissue lipolysis in other circumstances such as propranolol-induced inhibition (2) or after sucrois ingestion (11).

In contrast, most other studies published on this problem seem to support an inhibitory effect of lactate on lipolysis. Of this group of studies, the only one performed in humans showed a decrease of plasma FFAs and glycerol (5) during infusion, resulting in a peak plasma lactate level of 8.8 mM during 90 min exercise performed at 40% VO2max. A major difference was their use of whole body glycerol and FFA measurements, whereas we conducted a localized investigation of lipolysis in a specific adipose tissue. Indeed, the term adipose tissue can be confounding because it is quite heterogenous in the same individual. Various adipose tissues each possess different physiological properties.

As an example, lipolytic activity is much higher in abdominal adipose tissue than in gluteal adipose tissue during exercise (2). It is possible that abdominal adipose tissue may not be as sensitive to lactate-induced inhibition. Furthermore, other investigations that used whole body approaches (i.e., glycerol turnover or plasma glycerol assays) could have averaged the responses of various regional adipose tissues to lactate, giving a weighted result. Lactate could inhibit lipolysis in some but not all regional depots.

One had positive results in the dog that differed slightly from the data of Boyd et al. (5). Lactate infusions resulted in either a decreased rate of FFA appearance but not a decline of glycerol release (17) or a fall in plasma FFA but with an elevation of glycerol level (16). This divergent response of glycerol under the influence of lactate led Issekutz et al. (17) to explain the inhibition of lipid mobilization by lactate by an increase of the NADH/NAD ratio in adipocytes. This lactate-induced reduction of NAD would lead to the production of α-glycerophosphate and, finally, of glycerol. The hypothesis would be plausible if lactic acid were infused, but, in their experiments, sodium lactate was infused in dogs (17). It is thus difficult to understand how sodium lactate could have been a H+ donor that would result in NAD reduction in adipocytes.

A second mechanism suggested for the lactate-induced decrease of lipolysis is a diminution of cAMP in adipocytes. However, despite such a lactate-induced decrease of cAMP in adipocytes, no inhibition of lipolysis could be observed in vitro, whereas inhibition was observed in the presence of various fatty acids and β-hydroxybutyrate (13). A third likely mechanism involves the lactate-induced reduction of β-adrenoceptor binding. Lactate decreases isoproterenol-induced lipolysis in a dose-response curve (12). However, this reduction becomes significant only at a lactate level of 16 mM, observed during exercise in humans, and is much higher than the lactate levels supposed to diminish lipid mobilization from adipose tissue.

Other studies indicate no effect of lactate infusion in humans at rest (14, 18). Moderate hyperlactatemia (2.91 mM), which is representative of moderate-exercise intensity, has no effect on both plasma FFA and glycerol levels (14). Jenssen et al. (18) did not observe such in vivo lactate-induced inhibition at a slightly higher lactate rate of infusion combined with exercise. They attributed the discrepancy of their result with that of Issekutz et al. (16, 17) to the utilization of [14C]palmitate as a tracer in the latter studies. The use of [14C]palmitate vs. [14C]oleate is believed to overestimate inhibition of FFA mobilization (1).
In conclusion, our results suggest that lactate applied locally to adipocytes in subcutaneous abdominal adipose tissue does not elicit decreased fat mobilization from this depot during exercise. However, given the specificity of this adipose tissue, it cannot be concluded that lactate-induced inhibition of lipolysis is not present in other regional fat depots. This specific hypothesis deserves further investigation.

This study was partially supported by a grant from Fonds d’Intégration de Recherche-Université du Québec à Trois-Rivières. Address for reprint requests: F. Trudeau, Département des Sciences de l’Activité Physique, Université du Québec à Trois-Rivières, 3351, boul. des Forges, C.P. 500, Trois-Rivières, Québec, Canada G9A 5H7 (E-mail: Francois_Trudeau@uqtr.uquebec.ca).

Received 6 July 1998; accepted in final form 21 January 1999.

REFERENCES