

TABLE I. Use of Sodium Thiocyanate in Eliminating the Interference by Silver during Magnesium Determination.

Group	No. of samples	Addition of		Mg in brain tissue (meq/kg fresh wt)	
		AgNO ₃ and Ag precipitation with HCl	NaSCN	Mean	Range
I	3	—	—	10.3	(9.9-10.8)
II	3	+	—	.0	(.0-.0)
III	6	+	+	10.0	(9.2-10.2)

eliminates this interference such that the results on Mg agree well with the control samples (Group I). Furthermore, the thiocyanate-treated-Ag-containing samples showed the characteristic spectral curves with intersection at 500 m μ , as those shown in Fig. 1A. This was not the case in the samples containing traces of Ag but not treated with thiocyanate.

Summary. The interference of Ag and Hg in determination of Mg by the Titan yellow

dye method is demonstrated. A method is described for prevention of Ag interference by use of sodium thiocyanate and illustrated with analysis on brain tissue.

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Received September 13, 1961. P.S.E.B.M., 1962, v110.

Plasma Free Fatty Acids During Exercise and the Effect of Lactic Acid. (27478)

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Investigations of recent years seem to focus attention on the possible role of unesterified free fatty acids (FFA) as an extremely rapidly moving fuel(1,2). Andres and co-workers(3) have shown that O₂ uptake of the skeletal muscle is so high that it cannot be explained by oxidation of glucose alone. Direct electrical stimulation induces a measurable uptake of FFA by skeletal muscle of the dog(4). Friedberg *et al.* reported(5) a decrease of plasma FFA and an increased disappearance rate of injected palmitic-1-C¹⁴ during exercise.

To obtain further information about the role of FFA in muscular work, we studied the effect of exercise on plasma FFA levels on dogs with indwelling arterial and venous catheters.

Methods. Experiments were carried out on healthy mongrel dogs 9-13 kg of body weight. In morphine-pentothal narcosis, polyethylene catheters were placed into the right carotid artery and jugular vein. After

2-3 days of recovery, the dog was put on the treadmill, working for 20-25 minutes at a speed of 100 m/min and at a grade of 15-19%. O₂ uptake and CO₂ output were measured as follows: A plastic hood with a rubber collar was placed over the head of the animal. An air flow at rest of 25 l/min and 120 l/min at work was maintained by a pump; samples of the diluted expired air were collected in Douglas bags for about 2 minutes. To assure a proper evaporation of water from the tongue of the dog, the air flow was increased 5-6-fold between sample collections. The diluted expired air was analyzed by means of a Noyons diaferometer, the calibration of which was carried out as previously described(6). Arterial blood samples were taken at rest, during exercise (usually at 1½ min, 10 minutes and during the last 2 minutes of work), as well as 10 and 40 minutes after work. In other experiments 10 or 20% lactic acid was infused in the resting dog into the jugular vein at a rate of 1.97

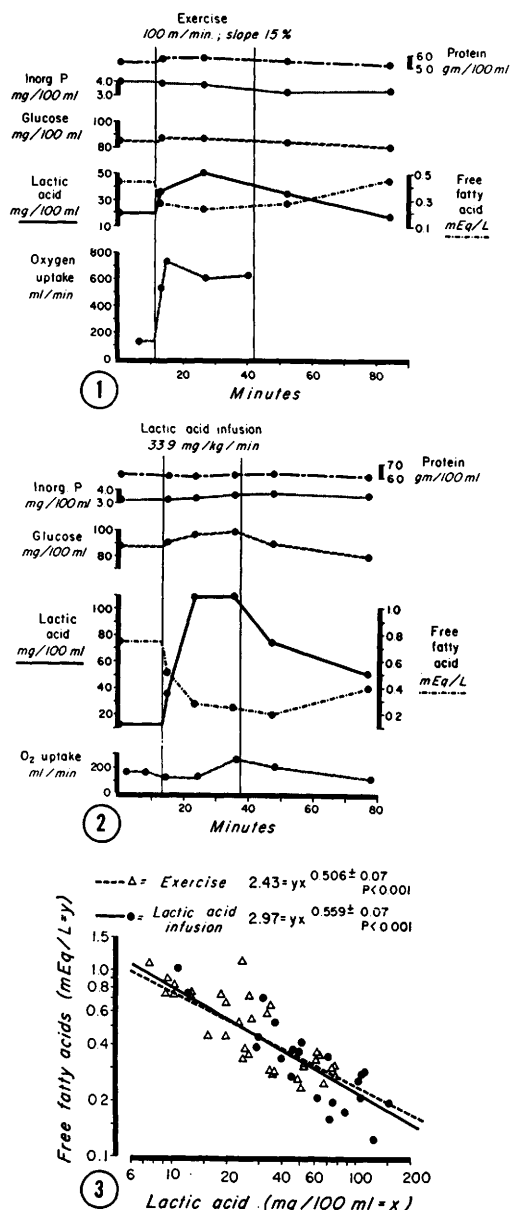


FIG. 1. Treadmill experiment on a dog (9.5 kg). Oxygen uptake expressed as ml O_2 STPD/min.

FIG. 2. Effect of lactic acid infusion on a resting dog (13.3 kg). Oxygen uptake expressed as ml O_2 STPD/min.

FIG. 3. Interrelationship between plasma FFA level and blood lactic acid concentration.

ml/min (16-34 mg/kg/min).

Plasma FFA was measured according to the method of Dole and Meinertz(7). To eliminate the interference due to lactic acid, the second extraction procedure was followed as recommended by these authors. The blood

lactic acid was determined by using the Ström modification(8) of the Summerson-Barker method. The following blood constituents were also measured: Plasma protein level according to Mehl(9), plasma inorganic P by using the method of Chen *et al.*(10) and blood glucose according to the glucose-oxidase* method.

Results. Fig. 1 shows the effect of exercise in a typical experiment. O_2 uptake rapidly increases to about 5 times that of the resting value. The blood lactate level quickly rises, the rate of increase being greater in the first 1½ minutes. Plasma FFA concentrations change inversely, rapidly declining in the first 1½ minutes and slowly approaching a final value of 0.2 meq/l. During recovery, as blood lactate decreases, FFA rises in the plasma. In general, the exercise-induced decrease of the FFA was greater when the initial FFA level was high (0.8-1.0 meq/l) and the resting lactate was low (around 10 mg/100 ml).

Considering the rapid changes in both lactic acid and FFA concentrations, it became necessary to study the effect of lactic acid infusions in the resting dog. Fig. 2 shows an experiment of this type. As the blood lactic acid level rises due to the intravenous infusion, the plasma FFA level drops correspondingly until it reaches its lowest value of 0.2 meq/l. After the cessation of infusion, like the exercise experiments when the lactate level decreases, the FFA starts to rise again.

A statistical evaluation of the 17 experiments (11 exercising dogs, and 6 lactate infusions) revealed a correlation between the logarithms of the blood lactate concentration and those of the plasma FFA concentration (Fig. 3). In both types of experiments, the regression coefficient proved to be significant at less than the .001 level. The difference between the 2 regression coefficients is less than their standard errors (± 0.07), consequently it is not significant ($0.7 < p < 0.6$). In other words, the decrease of FFA is not caused by the work itself but by the accumulation of lactic acid during exercise.

Discussion. The almost immediate drop of

* "Glucostat" reagent: Worthington Biochemical Corp., Freehold, N. J.

FFA at the start of the exercise, without significant change of the plasma protein, inorganic P and glucose levels, shows that the decrease of FFA cannot be explained by a dilution of the plasma, or by a sudden release of insulin. From the 2 metabolic parameters which change rapidly during work, namely O_2 uptake and lactate level, the effect of increased metabolism was excluded by the experiments carried out on resting dogs with lactate infusions.

In view of the demonstrated interrelationship between blood lactate and plasma FFA, it would be advisable, whenever a change of blood lactate is to be expected, to measure the concentration of lactic acid in the blood in addition to the FFA. At low resting levels of blood lactate a slight rise, for instance from 7 to 20 mg/100 ml, causes a considerable drop in FFA, from 1.0 to 0.55 meq/l. Whether this is the result of a competition between the lactate anion and FFA for the albumin carrier, or a direct metabolic effect on the release of FFA in the adipose tissue, remains to be shown.

Summary. Treadmill experiments were carried out on dogs with indwelling arterial

and venous catheters. The plasma free fatty acids level rapidly decreases during exercise, due to the rise of the blood lactic acid concentration, as was shown by lactic acid infusions in resting dogs. A logarithmic correlation was found between plasma free fatty acid and blood lactic acid concentration.

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Received February 12, 1962. P.S.E.B.M., 1962, v110.

Action of Plasma and of Plasma Albumin on Palmitate-1- C^{14} Oxidation by Blood Cells.* (27479)

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Plasma albumin appears to have an important regulatory function in cell metabolism. Thus, Fritz *et al.*(6) observed that plasma albumin stimulates the oxidation of palmitate-1- C^{14} by skeletal muscle *in vitro* in concentrations lower than one micromole of albumin per 7 micromoles of palmitate, but inhibits this oxidation at higher concentrations. Helinski and Cooper(8) observed that plasma albumin is an essential component for oxidative phosphorylation of sucrose, succinate,

beta-hydroxybutyrate and some other substrates by rat liver mitochondria, and Sacktor *et al.*(13) made similar observations with mitochondria from insect flight muscle. Glinos (4) observed that the albumin level in blood plasma of the rat and its changes after partial hepatectomy constitute an important regulatory factor for liver growth and regeneration.

The aim of the present investigation was to obtain quantitative data concerning the action of blood plasma and of plasma albumin on rate of tissue and cell metabolism *in vitro*. We chose to use blood cells and palmitate-1- C^{14} as cell-substrate system for this study

* This research was supported in part by grant Nos. H-3173 and H-4843 from U. S. Public Health Service.