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MYOCARDIAL METABOLISM IN MAN AT REST AND DURING PROLONGED EXERCISE

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The technique of coronary sinus catheterisation with measurement of arterio-venous differences across the heart has provided considerable information about the uptake and utilization of plasma substrates by the myocardium in resting man. There is general agreement that in the resting fasting state plasma free fatty acids (FFA) and glucose are the principle substrates for myocardial oxidation with smaller contributions being made by plasma lactate, pyruvate and ketone bodies (1,2,3,4,5,6). However, despite the finding of triglyceride uptake by the isolated animal heart (7,8, 9,10,11,12), and the demonstration of the presence of lipoprotein lipase activity in the human heart (13,14), coronary sinus catheterisation studies in man have failed to demonstrate satisfactorily the uptake of plasma triglycerides by individual hearts.

It is likely that this inability to demonstrate the disappearance of triglycerides across the heart by coronary sinus sampling in man has been due to the methodological difficulties of measuring the very small amounts of triglyceride which could provide a substantial proportion of the heart's energy supply. Thus, in an individual with a normal arterial triglyceride concentration of 1000 µmol/l plasma, there would need to be an arterio-venous difference of 150 µmol/l plasma in order for it to be detected using one of the current chemical methods of triglyceride estimation with a standard error of around five per cent for a determination. This difference is equal to about 90 µmol/l blood if the haematocrit is normal. Since the oxygen equivalent of triglyceride fatty acid is about 73.5, complete oxidation of 90 µmol of triglyceride fatty acid would require 6660 µmol of oxygen. However, the average arterio-venous difference of oxygen across the heart is about 5000 µmol/l

blood. Thus, oxidation of 90 µmol of triglyceride fatty acid would need 133 per cent of the heart's oxygen extraction per litre of blood: that is, triglyceride would have an oxygen extraction ratio (OER) of 133 per cent. Since it is known that plasma FFA and carbohydrate substrates already account for a substantial fraction of the myocardial oxygen uptake, this minimum arterio-venous difference of triglyceride detectable using a method of estimation with the usual error of around five per cent, is far greater than could be expected. Even using a very much more precise method with a standard error of only one per cent, an arterio-venous difference of 19 µmol/l blood would be required and oxidation of this amount of triglyceride fatty acid would produce an OER of 28 per cent. Thus, because of the very high oxygen equivalent of triglyceride fatty acid, a very precise method of triglyceride estimation is required in order to be able to detect the quantities of triglyceride which it might be reasonable to expect the heart to extract.

The first purpose of the present study was, therefore, to examine the problem of the uptake of plasma triglyceride by the myocardium in resting fasting man by using both a radio-isotope technique and a precise chemical method of triglyceride estimation. The second purpose was then to use these techniques to investigate the utilization of plasma triglycerides and other substrates in man during prolonged exercise in the fasting state. For, although myocardial substrate utilization has been studied in man performing exercise of four to 20 minutes duration (4,15,16), there have been no reports of the effects of prolonged exercise.

METHODS

Twenty-eight unsedated, healthy male subjects between the ages of 23 and 53 years were studied in the resting state after an overnight fast with catheters in the coronary sinus and a brachial artery. In nine of these subjects measurements were made both at rest and during the last five minutes of a two hour period of supine leg exercise on a bicycle ergometer at a load of approximately 50 per cent of the rate of work which the subject was able to perform at a heart rate of 170/min. Arterio-venous differences in concentration across the heart of FFA (17), triglyceride (18), glucose (19), lactate (20), pyruvate (21) and glycerol (22) were measured chemically. Plasma triglycerides were estimated in triplicate on 10 extracts from each blood sample. The average figure for the standard error of the mean value for the 10 extracts of each sample was 0.8 per cent.

In addition to the chemical measurements, arterio-venous differences in FFA and triglyceride were determined by using a continuous infusion of albumin-bound ^{3}H palmitate as a tracer for plasma FFA and to produce endogenous labelling of plasma triglycerides (23). FFA and triglyceride were separated by thin layer chromato-

graphy and then counted in a Packard 3375 liquid scintillation spectrometer (24).

The oxygen content of the blood samples was calculated from the haemoglobin concentration, the oxygen saturation measured spectrophotometrically (25), and the oxygen tension measured with a polarographic electrode.

ARTERIAL-CORONARY SINUS CONCENTRATION DIFFERENCES IN THE RESTING FASTING STATE

Table I presents the findings in the resting fasting state. The arterial concentrations of all substrates were within normal limits. The myocardial uptake of the substrates is presented in the table both as the arterial-coronary sinus difference in concentration and as the OER. Carbohydrate substrates accounted for

SUBSTRATE	ARTERIAL CONCENTRATION	ARTERIAL-CORONARY SINUS DIFFERENCE IN CONCENTRATION	OXYGEN EXTRACTION RATIO
GLUCOSE	3640±107	151±24	18±3
LACTATE	645±44	127±22	4±1
PYRUVATE	50±4	6±4	0±0.2
FFA	706±38		
Chemical		171±12	49±3
Isotope		243±10	69±3
TRIGLYCERIDE	1080±88		
Chemical		16±4	14±4
Isotope		24±7	23±6

Table I: Myocardial metabolism in resting fasting man. Arterial concentrations, arterial-coronary sinus differences and oxygen extraction ratios are presented as the mean ± SEM for 28 subjects. Concentrations and arterial-coronary sinus differences are in µmol/l plasma except for glucose which is in µmol/l blood. Arterial-coronary sinus differences in concentration (A-CS) for FFA and triglyceride were measured by both chemical and radio-isotope techniques.

a total OER of 22 per cent. There was a considerable difference in the uptake of FFA as estimated by chemical and isotope techniques. This has also been observed by other workers (6) and is due to the fact that the coronary sinus FFA specific activity is lower than the arterial specific activity.

In addition to the uptake of carbohydrate substrates and FFA, a significant arterio-venous difference across the heart of triglyceride was found by chemical estimation in 13 subjects and by isotope measurements in 17 subjects. The average OER for all 28 subjects was 14 per cent chemically and 23 per cent isotopically. A possible explanation for the higher value found by the isotope technique is that in calculating the arterio-venous difference in concentration from the arterio-venous difference in triglyceride radio-activity, it has been assumed that the labelled triglyceride is evenly distributed throughout all of the plasma lipoprotein fractions. It is probable, however, that under these conditions the major part of the labelled triglyceride was located in the very low density lipoprotein fraction which contains about 50 per cent of the total plasma triglycerides in normal man (J. Boberg, personal communication). If this is the case, then the arterio-venous difference in triglyceride concentration determined from the arterio-venous difference in radio-activity would be half of that which has been calculated. The myocardial uptake of triglyceride would then be about 12±4 (SEM) μmol/l plasma and the OER about 12±3 (SEM) per cent. The chemical and isotopic techniques would then agree much more closely.

ARTERIAL-CORONARY SINUS CONCENTRATION DIFFERENCES DURING PROLONGED EXERCISE

After two hours of exercise, changes were found in the coronary sinus oxygen saturation, the arterial concentrations of substrates and in the pattern of myocardial substrate uptake. In addition, a number of subjects showed a significant myocardial production of free glycerol.

Figure 1 shows the changes in oxygen saturation of the coronary sinus blood after two hours of exercise. The saturation fell in all nine subjects from an average resting value of 34.6±1.5 (SEM) per cent to 24.7±1.2 (SEM) per cent. This fall, equal to 28 per cent of the resting value, was highly significant ($p < 0.001$). Since the arterial oxygen saturation did not change with exercise, the reduced coronary sinus saturation indicates an increased myocardial extraction of oxygen from each litre of blood. The average arterial-coronary sinus difference in oxygen content was 4875±189 (SEM) μmol/l blood at rest and 5917±170 (SEM) μmol/l blood during exercise.

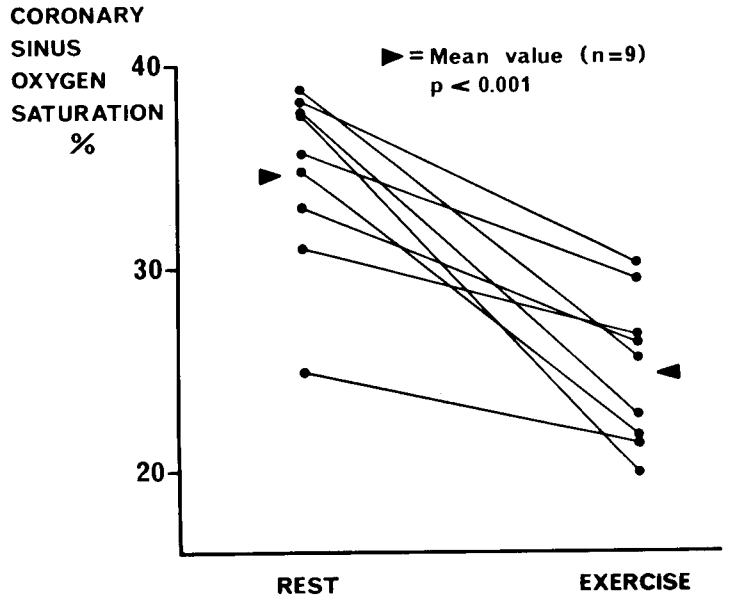

Figure 1: Changes in coronary sinus oxygen saturation after approximately two hours of supine leg exercise on a bicycle ergometer.

The effect of exercise on the arterial concentration of the various substrates is presented in Table II. Glucose concentration

SUBSTRATE	ARTERIAL CONCENTRATION	
	Rest	Exercise
GLUCOSE	3916±203	3258±167
LACTATE	603±91	1272±194
PYRUVATE	47±5	86±6
FREE FATTY ACIDS	614±49	1366±106
TRIGLYCERIDE	1127±157	1123±157

Table II: Effect of two hours of supine leg exercise on arterial concentrations of substrates. Concentrations are in µmol/l plasma except for glucose which is in µmol/l blood. Values are means±SEM for nine subjects.

fell to 83 per cent of the resting level while the concentrations of lactate and pyruvate doubled. The FFA turnover rate, which was estimated from the isotope data, increased an average of 2.8 times with exercise and the arterial concentration of FFA rose to 222 per cent of the resting level. There was no significant change in the arterial concentration of triglycerides with exercise.

Table III shows the effect of prolonged exercise on both the arterial-coronary sinus difference in concentration and the oxygen extraction ratios. The fall in arterial glucose concentration was

SUBSTRATE	ARTERIAL-CORONARY SINUS DIFFERENCES IN CONCENTRATION		OXYGEN EXTRACTION RATIOS PER CENT	
	Rest	Exercise	Rest	Exercise
GLUCOSE	191±59	97±32	24±8	9±3
LACTATE	84±32	293±113	3±1	9±3
PYRUVATE	-2±5	20±9	0±0.2	1±0.2
FFA				
Chemical	171±19	209±9	49±4	50±3
Isotope	242±18	229±13	70±4	55±4
TRIGLYCERIDE				
Chemical	15±10	17±10	14±9	12±7
Isotope	34±19	10±14	30±17	8±10

Table III: Effect of two hours of supine leg exercise on arterial-coronary sinus differences in substrate concentrations and on oxygen extraction ratios. Differences in concentration are in μmol/l plasma except for glucose which is in μmol/l blood. Values are means ±SEM for nine subjects except for the isotope estimate of triglyceride at rest where measurements were made in only five subjects.

accompanied by a fall in the OER from 24 per cent to nine per cent and the rise in arterial lactate concentration was accompanied by a rise in the OER of this substrate from three per cent to nine per cent. During exercise pyruvate had an OER of only about one per cent. Thus, during prolonged exercise in the fasting state the total OER for plasma carbohydrate substrates fell from 27 per cent to 19 per cent.

Although the arterial concentration of FFA more than doubled, there was only a small increase in FFA extraction as estimated

chemically and the OER did not change. The arterio-venous difference estimated isotopically fell and the OER decreased from 70 per cent to 55 per cent.

In the case of the chemical estimates of triglyceride uptake, there was a significant increase in uptake in one of the nine subjects and a significant fall in uptake in another. Two of the five subjects in whom isotope measurements of triglyceride uptake were made both at rest and during exercise showed a significant fall in uptake. However, the average triglyceride uptakes for the group as a whole, estimated either chemically or isotopically, did not change significantly.

Figure 2 shows the effect of exercise on the average arterial concentration of free glycerol and on the arterial-coronary sinus difference in free glycerol concentration. After two hours of exercise the glycerol concentration increased from 48±5 (SEM) μmol/l plasma to 278±36 (SEM) μmol/l plasma. With exercise five of the nine subjects showed a significant increase in the production of glycerol across the heart, suggesting increased hydrolysis of lipid. Possible sources of this glycerol are the plasma triglycerides and cardiac lipid stores. Since no increase in the arterial-coronary sinus difference in triglyceride was detected during exercise, it seems more likely that the glycerol arose from hydrolysis of cardiac lipid stores rather than from circulating triglyceride.

SUMMARY

Myocardial substrate utilization was studied in 19 normal, fasting men at rest and in nine others both at rest and during the last few minutes of a two hour period of submaximal exercise. Arterial-coronary sinus differences in concentration were measured chemically for glucose, lactate, pyruvate, glycerol, free fatty acid (FFA) and triglycerides, and radio-isotopically for FFA and triglycerides.

At rest the total oxygen extraction ratio (OER) for plasma carbohydrate substrates averaged 22 per cent, and that for FFA, 49 per cent chemically and 69 per cent isotopically. A statistically significant arterial-coronary sinus difference in triglycerides was detected in 13 of the 28 subjects chemically and in 17 subjects isotopically.

After two hours of exercise the nine subjects showed a significant fall in coronary sinus oxygen saturation and increase in myocardial oxygen extraction per litre of blood. The arterial con-

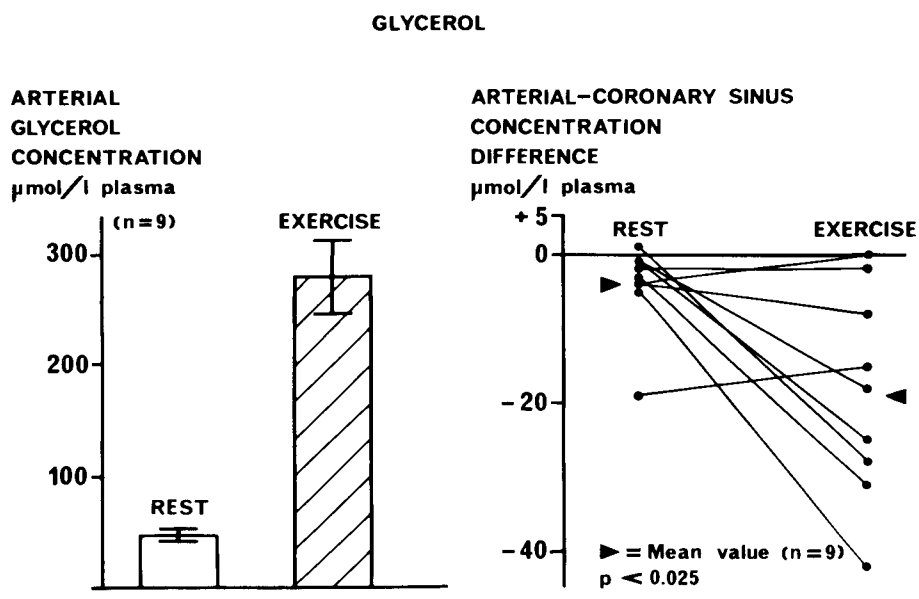

Figure 2: Effect of two hours of supine leg exercise on the average arterial concentration and the arterial-coronary sinus differences in concentration of free glycerol in nine subjects.

centration of glucose fell slightly and the concentrations of lactate, pyruvate and FFA approximately doubled, while the concentration of triglycerides did not change significantly. The total OER for plasma carbohydrate in these nine subjects fell from 27 per cent to 10 per cent during exercise. The OER for FFA estimated chemically did not change, but that estimated isotopically fell from 70 per cent to 55 per cent. No significant change in the average uptake of plasma triglycerides was detected either chemically or isotopically. During exercise five subjects showed a significant increase in the myocardial production of glycerol suggesting hydrolysis of cardiac lipid stores.

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