Effect of Free Fatty Acids on Myocardial Function and Oxygen Consumption in Intact Dogs

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ABSTRACT Myocardial function and oxygen consumption ($M\dot{V}O_2$) were studied during increased myocardial uptake of free fatty acids (FFA) induced by intravenous infusion of a fat emulsion (Intralipid) after heparinization in anesthetized and intact dogs. During raised myocardial uptake of FFA, $M\dot{V}O_2$ increased in all experiments. On the average, $M\dot{V}O_2$ rose from 8.6 to 10.7 ml/min·100 g, or 26% ($P < 0.001$). This was mainly due to elevated myocardial oxygen extraction, as myocardial blood flow was unchanged, or increased slightly. In the recovery period, $M\dot{V}O_2$ returned to normal. Left ventricular pressure, the maximal rate of rise of left ventricular pressure (dP/dt), heart rate, and cardiac output remained unchanged during the raised myocardial uptake of FFA.

These experiments show that increased myocardial uptake of FFA in intact hearts was associated with augmented $M\dot{V}O_2$, despite unchanged mechanical activity.

INTRODUCTION

Studies in isolated aerobically perfused rat hearts (1, 2) have shown that high concentrations of free fatty acids (FFA)[1] increased myocardial oxygen consumption ($M\dot{V}O_2$). Since augmentation of $M\dot{V}O_2$ has generally been attributed to increased mechanical performance through raised myocardial contractility, preload, developed tension, and heart rate (HR) (3), it is unclear whether the observed rise in $M\dot{V}O_2$ was due to increased myocardial performance or to a direct effect of FFA on myocardial metabolism. The present study was therefore undertaken to examine the possible $M\dot{V}O_2$-stimulating effect of high plasma-FFA in intact hearts under controlled hemodynamic conditions.

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[1] *Abbreviations used in this paper:* CO, cardiac output; dP/dt, maximal rate of rise of left ventricular pressure; FFA, free fatty acids; HR, heart rate; LVP, left ventricular pressure; LVW, left ventricular work; MF, myocardial blood flow; $M\dot{V}O_2$, myocardial oxygen consumption.

METHODS

Experiments were carried out in intact mongrel dogs of either sex, ranging in weight from 14 to 23 kg. The dogs were fasted overnight, and anesthetized with sodium pentobarbital at a dosage of 25 mg/kg and ventilated through an endotracheal tube. Left ventricular pressure (LVP) was measured with a Statham P23Gb transducer (Statham Instruments, Inc., Oxnard, Calif.) connected to a radiopaque polyethylene tubing introduced into the left ventricle from a carotid artery. The transducer system was series damped to obtain a flat frequency response of up to 30 cycles per sec. The first derivative of LVP with regard to time (dP/dt), was recorded continuously by means of a differentiator connected to the output from the pressure channel. Calibration was performed by linear ramp voltage. The reliability of this system has previously been examined (4). Cardiac output (CO) was determined by thermal dilution technique. 0.9% saline of room temperature was injected as a bolus into right atrium. Temperature changes were recorded with a thermocouple made of copper-constantan advanced from a femoral artery to the thoracic aorta, and CO calculated according to Fegler (5). Myocardial blood flow (MF) was measured by the hydrogen desaturation technique introduced by Aukland, Bower, and Berliner (6). H_2-concentration was determined polarographically with a platinum electrode mounted on the tip of a cardiac catheter inserted under fluoroscopic guidance into the coronary sinus. The current obtained is linearly related to H_2-concentration in the coronary sinus. The blood drained through the coronary sinus was regarded as representative of blood passing through the left ventricle of myocardium (7). Blood flow in ml/min·100 g tissue was calculated from the half-times of the monoexponential desaturation curves, according to the formula

$$F = \frac{69.3}{t_{\frac{1}{2}}}$$

where $t_{\frac{1}{2}}$ is the half-time in minutes. H_2 tissue/blood partition coefficient and specific gravity were assumed to be 1.00. Hydrogen gas was given with respiratory air until stable concentrations were recorded in coronary sinus blood. Gas administration was then stopped, and 30–50 ml of 0.9% saline, saturated with hydrogen at 1 atmosphere, were injected into the left ventricle to maintain a constant hydrogen concentration in the myocardium for another 60 sec, ensuring immediate desaturation of arterial blood at the end of the infusion. Hydrogen oxidation current was measured

TABLE I

Hemodynamic Effects of Intralipid Infusion in Heparinized Dogs. Mean ±SE of Seven Experiments

		LVSP	LVEDP	dP/dt	HR	CO	LVW
	n	*mm Hg*	*mm Hg*	$\frac{mm\ Hg \cdot 10^{-3}}{sec}$	*beats/min*	*ml/min·kg*	*kg/min·kg*
Control	7	149	3.9	2.44	144	131	0.27
±SE		3	0.6	0.27	4	15	0.03
Intralipid infusion		149	3.8	2.56	140	131	0.27
±SE		3	0.6	0.25	4	15	0.03
P		NS	NS	NS	NS	NS	NS
Recovery		143	3.6	2.47	143	133	0.26
±SE		4	0.6	0.22	3	14	0.03

n, number of dogs; LVSP, left ventricular systolic pressure; LVEDP, left ventricular end-diastolic pressure; dP/dt, first derivative of left ventricular pressure; HR, heart rate; CO, cardiac output; LVW, left ventricular work; NS, not significant ($P > 0.05$).

with Keithley Instruments Microvolt Ammeter, 150A, (Keithley Instruments, Inc., Cleveland, Ohio) and the output signal recorded on a Honeywell Brown Electronics Recorder (Honeywell Inc., Industrial Div., Fort Washington, Pa.).

Arterial and coronary sinus blood samples were obtained simultaneously and analyzed for oxygen saturation (8). Hemoglobin was measured spectrophotometrically as cyanmethemoglobin.

The concentrations of FFA in arterial and coronary sinus blood were determined by the method of Dole (9), as modified by Trout, Estes, and Friedberg (10). To avoid in vitro lipolysis, blood was sampled in tubes precooled to 0°C and immediately centrifuged for 10 min at 2°C. Plasma was frozen until FFA analyses were performed. Plasma-FFA was extracted with a mixture containing 40 parts by volume of isopropanol, 10 parts heptane, and 1 part 1N H_2SO_4 (10).

The magnitude of in vitro lipolysis during hypertriglyceridemia was studied in separate experiments. Plasma-FFA was unchanged 1 hr after blood sampling. When FFA-extraction was delayed for 2 hr after blood sampling, plasma-FFA rose by 10–15%. This late effect was prevented when blood was sampled in tubes containing protamine sulphate. However, since extraction of FFA was performed within half-an-hour, pretreatment of tubes with protamine sulphate was not used in the routine procedure.

Experimental procedure. After control registrations and blood sampling, plasma-FFA was elevated by continuous intravenous (i.v.) infusion of a fat emulsion, Intralipid "Vitrum," after heparinization (sodium heparin, 3 mg/kg, i.v.). 10% Intralipid was infused at a rate of 1.5-3.0 ml/min for a period up to 45 min. Blood samples were obtained at 15, 30, and 45 min infusion. Arterial concentrations of FFA increased promptly and leveled off after 15 min. Tables I and II show mean values of hemodynamic and metabolic data obtained after 30 and 45 min infusion. 30 min after discontinuation of the Intralipid infusion, recovery registrations, and blood samples were obtained.

Calculations. Left ventricular work (LVW) (kg/min·kg) was calculated as $0.0136 \times CO \times LVSP$ (11), where CO is cardiac output (ml/min·kg) and LVSP left ventricular systolic pressure (mm Hg).

$M\dot{V}O_2$ (ml/min·100 g) was calculated from the oxygen extraction of the myocard (O_2-extr.) and MF.

Myocardial uptake of FFA (μEq/min·100 g) was calculated as the product of arterio-coronary sinus differences of FFA and myocardial plasma flow.

To evaluate differences, probability values were obtained utilizing Student's *t* test for paired data.

RESULTS

Mechanical performance before, during, and after Intralipid infusion, as measured by systolic and end-diastolic LVP, dP/dt, HR, CO, and LVW, remained unchanged (Table I).

In all experiments, increased myocardial uptake of FFA was associated with augmented $M\dot{V}O_2$ (Fig. 1).

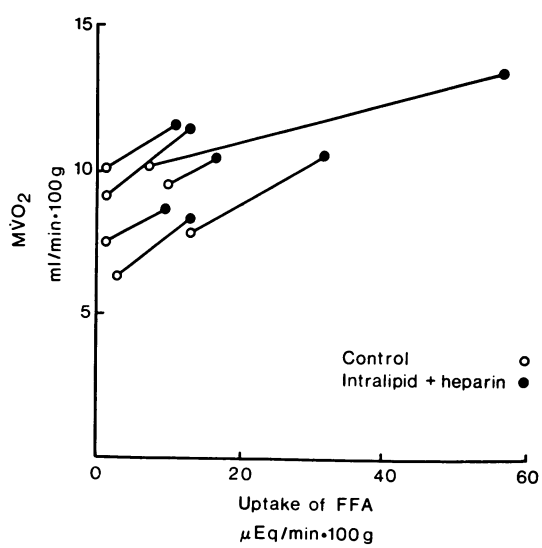

FIGURE 1 Myocardial oxygen consumption ($M\dot{V}O_2$) and myocardial uptake of free fatty acids (FFA) before and during administration of Intralipid + heparin.

Table II shows metabolic data before, during, and after Intralipid infusion. $M\dot{V}O_2$ increased on average from 8.6 to 10.7 ml/min·100 g (+26%, $P < 0.001$) when FFA uptake rose on average from 5.7 to 22.0 μEq/min·100 g. The augmentation $M\dot{V}O_2$ was mainly due to elevated myocardial oxygen extraction. MF was unchanged or increased slightly. In the recovery period, $M\dot{V}O_2$ returned to normal.

DISCUSSION

The major finding in the present investigation was that elevated myocardial uptake of FFA was associated with increased $M\dot{V}O_2$.

Raised $M\dot{V}O_2$ was principally based on increased myocardial oxygen extraction while MF was virtually unchanged. Previous data have shown that the H_2 desaturation technique is a sensitive and reliable method for measurements of MF (6). Thus it is unlikely that a decrease in MF reciprocal to increased O_2-extraction would not have been registered, had it occurred.

The chief determinants of $M\dot{V}O_2$ are myocardial contractility, preload, developed tension, and HR (3). Mechanical factors, however, can be ruled out as responsible for the elevated $M\dot{V}O_2$, as systolic and end-diastolic LVP, dP/dt, CO, and HR remained unchanged during raised plasma-FFA. These results are in accordance with Henderson, Most, Parmley, Gorlin, and Sonnenblick (12) who found that FFA did not alter developed force in well-oxygenated rat papillary muscle.

Whether high arterial concentrations and myocardial uptake of FFA actually increase $M\dot{V}O_2$ in vivo has not been clearly established, and the experimental evidence is conflicting (1, 13).

The $M\dot{V}O_2$-stimulating effect of FFA has been demonstrated by Challoner and Steinberg (1, 2) in isolated perfused rat hearts; however, mechanical activity was not controlled in their experiments. Moreover, marked elevation of $M\dot{V}O_2$ generally required higher concentrations of FFA than those found in vivo.

Henderson and Sonnenblick (13) have recently shown that high concentrations of FFA actually *decreased* $M\dot{V}O_2$ in isolated rat hearts perfused at constant flow and paced at constant rate. On the other hand, myocardial performance was also markedly depressed, thus invalidating the interpretation of the relationship between FFA-uptake and $M\dot{V}O_2$.

A mechanism for the $M\dot{V}O_2$-stimulating action of FFA in vivo remains to be definitely established. Challoner and Steinberg (1, 2) suggested an uncoupling effect of high concentrations of FFA within myocardial cells on oxidative phosphorylation, as also demonstrated in isolated liver mitochondria (14).

It is known that during increased myocardial uptake of FFA, a significant fraction of the FFA taken up is not immediately oxidized but stored in myocard as triglycerides. However, the amount of oxygen required for esterification of the additional amount of FFA taken up during high plasma-FFA is less than 10% of the observed increase in $M\dot{V}O_2$ (15).

The induced levels of plasma-FFA in the present experiments are comparable with those seen during increased catecholamine activity. Augmented $M\dot{V}O_2$ during catecholamine stimulation—which has been difficult to attribute to augmented mechanical activity alone—may therefore be partly explained by the effect of FFA (2, 16). The present results may also be rele-

TABLE II

Metabolic Effects of Intralipid Infusion in Heparinized Dogs. Mean ±SE of Seven Experiments

		FFA						
		a	a-cs	u	$M\dot{V}O_2$	O_2-extr.	MF	Hb
	n	*μEq/liter*	*μEq/liter*	*μEq/min ·100 g*	*ml/min ·100 g*	%	*ml/min ·100 g*	*g/100 ml*
Control	7	488	109	5.7	8.6	57.6	90	14.1
±SE		67	32	1.8	0.6	3.0	7	0.6
Intralipid infusion		3105	371	22.0	10.7	69.3	93	14.2
±SE		286	80	6.5	0.6	1.7	8	0.5
P		<0.001	<0.01	<0.05	<0.001	<0.005	NS	NS
Recovery		584	148	8.9	8.3	58.3	90	12.8
±SE		80	45	2.8	0.6	3.9	4	0.3

n, number of dogs; a, arterial concentrations; a-cs, arterio-coronary sinus differences; u, myocardial uptake of free fatty acids (FFA); $M\dot{V}O_2$, myocardial oxygen consumption; O_2-extr., myocardial oxygen extraction; MF, myocardial blood flow; Hb, hemoglobin; the values for *P* are the highest probability that the observed differences are due to chance. NS, not significant ($P > 0.05$).

vant in other situations with high plasma-FFA, i.e., hyperthyroidism and diabetes mellitus.

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