Effect of Inhibition of Lipolysis on Myocardial Oxygen Consumption in the Presence of Isoproterenol

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ABSTRACT The effect of intravenous infusion of isoproterenol on myocardial oxygen consumption ($M\dot{V}O_2$) was studied in 10 intact and anesthetized dogs before and after inhibition of lipolysis. In five dogs lipolysis was inhibited by nicotinic acid or beta pyridyl carbinol and in five other dogs by high plasma glucose concentrations. In spite of similar mechanical responses to isoproterenol, as evidenced by left ventricular pressure, maximal rate of rise of left ventricular pressure (dP/dt), heart rate and cardiac output, augmentation of $M\dot{V}O_2$ was larger before (on average 7.6 ml/min·100 g) than after inhibition of lipolysis either by antilipolytic drugs (on average 4.5 ml/min·100 g) ($P < 0.005$), or by high plasma glucose concentrations (on average 4.3 ml/min·100 g) ($P < 0.02$). As mechanical responses to isoproterenol were similar before and after inhibition of lipolysis, it is concluded that the additional rise in $M\dot{V}O_2$ with intact lipolysis was caused by a metabolic stimulation by high concentrations of free fatty acids.

INTRODUCTION

Stimulation of β -adrenergic receptors, e.g. by isoproterenol, increases the mechanical performance of the heart and has a marked effect on myocardial oxygen consumption ($M\dot{V}O_2$).¹ The augmented $M\dot{V}O_2$ has previously been related to increased mechanical activity of the heart (1, 2). In an earlier study it was found that increased myocardial uptake of free fatty acids (FFA) was accompanied by raised myocardial oxygen consumption

in intact dogs, despite unchanged mechanical activity (3). Since intravenous (i.v.) infusion of isoproterenol is associated with augmentation of plasma FFA, and hence raised myocardial uptake of FFA, it is conceivable that part of the large increase in $M\dot{V}O_2$ is due to increased myocardial uptake of FFA. Lipolysis induced by catecholamines may be inhibited by nicotinic acid (4-6), beta pyridyl carbinol (Ronicol; F. Hoffmann-La Roche & Co. A. G., Basel, Switzerland) (7), or high plasma glucose concentrations (8). The present investigation was undertaken to examine the effect of isoproterenol on $M\dot{V}O_2$ and its relation to high concentrations of FFA.

METHODS

After a fasting period of 12-15 hr, experiments were carried out on 10 mongrel dogs of either sex (18-33 kg body weight [b.w.]). The dogs were anesthetized with sodium pentobarbital, 25 mg/kg b.w., as initial dose, followed by maintenance doses. Sodium heparin, 3 mg/kg b.w., given i.v., was used as anticoagulant.

Left ventricular pressure (LVP) was measured with a Statham P23Gb transducer connected to a radiopaque polyethylene tubing introduced into the left ventricle from a carotid artery. 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 (9). Mean aortic blood pressure ($\overline{AP}$) was recorded from a femoral artery with a Statham P23Gb transducer. Cardiac output (CO) was determined by thermal dilution technique (3), and calculated according to Fegler (10). Myocardial blood flow (MF) was measured by the hydrogen desaturation technique, introduced by Aukland, Bower, and Berliner (11), described in detail earlier (3). Arterial and coronary sinus blood samples were obtained simultaneously and analyzed for oxygen saturation (12). Hemoglobin was measured spectrophotometrically as cyanmethemoglobin. The concentrations of FFA in arterial and coronary sinus blood were determined by the method of Dole (13), as modified by Trout, Estes, and Friedberg (14). In vitro lipolysis was avoided by sampling blood in tubes precooled to 0°C and immediately centrifuging for 10 min at 2°C (3). Plasma was frozen until FFA analyses were performed.

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¹Abbreviations used in this paper: $\overline{AP}$, mean aortic blood pressure; CO, cardiac output; dP/dt , maximal rate of rise of left ventricular pressure; FFA, free fatty acids; HR, heart rate; LVEDP, left ventricular end-diastolic pressure; LVP, left ventricular pressure; LVSP, left ventricular systolic pressure; LVW, left ventricular work; MF, myocardial blood flow; $M\dot{V}O_2$, myocardial oxygen consumption.

The dogs were divided into two experimental groups. In the first series, lipolysis was inhibited by nicotinic acid or beta pyridyl carbinol. The results have been pooled, as the action of beta pyridyl carbinol is mediated through nicotinic acid (7), which is a metabolic product of beta pyridyl carbinol. In the second series, lipolysis was inhibited by high plasma glucose concentrations.

Group I consisted of five dogs. After control registrations, constant i.v. infusion of isoproterenol was started at a rate which doubled dP/dt. Infusion rates of 2–3 µg/min were usually required. Stable conditions were reached in about 10 min, and new registrations and blood samples were obtained. 30 min after discontinuing isoproterenol infusion, lipolysis was inhibited by continuous i.v. infusion of nicotinic acid (5–20 mg/min) in three experiments, and by i.v. infusion of beta pyridyl carbinol (5–20 mg/min) in the other two dogs. Intravenous infusion of isoproterenol began 15 min later. The infusion rate was adjusted so as to reach the same dP/dt as in the control run. This led to comparable changes in all hemodynamic parameters measured. The dose of isoproterenol needed was similar to that used in the control run. In about 10 min, stable conditions were reached, and registrations and blood samples were obtained.

Group II also consisted of five dogs. Registrations and blood sampling were performed before and after 10 min constant i.v. infusion of isoproterenol at a rate of 2–3 µg/min. 30 min after discontinuing isoproterenol infusion, lipolysis was inhibited by plasma glucose concentrations of about 250 mg/100 ml. Plasma glucose was raised by i.v. injection of 40–50 ml 10–20% glucose solution followed by infusion at a constant rate of 1 ml/min. Control samples were obtained 30 min later; i.v. infusion of isoproterenol was started at a similar infusion rate as in the control run. Stable conditions were reached after 10 min infusion of isoproterenol, and new blood samples were obtained for metabolic studies.

Control studies with two repetitive i.v. infusions of isoproterenol at similar rates, without inhibition of lipolysis, showed a similar rise in mechanical performance, $\dot{M}\dot{V}O_2$,

arterial concentration, and myocardial uptake of FFA in both experimental settings.

Calculations. Left ventricular work (LVW) (kg-m/min·kg) was calculated as $0.0136 \times CO \times LVSP$ (15), where CO is cardiac output (ml/min·kg), and LVSP (left ventricular systolic pressure) (mm Hg). $\dot{M}\dot{V}O_2$ (ml/min·100 g) was calculated from the oxygen extraction of the myocardium and MF. Myocardial uptake of FFA (µEq/min·100 g) was calculated as the product of arteriocoronary sinus differences of FFA and myocardial plasma flow.

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

RESULTS

Group I. Hemodynamic data before and during i.v. infusion of isoproterenol, without and with inhibition of lipolysis by nicotinic acid or beta pyridyl carbinol, are presented in Table I. The isoproterenol-induced changes in mechanical activity, as evidenced by $\bar{A}P$, LVSP, left ventricular end-diastolic pressure (LVEDP), dP/dt, heart rate (HR), CO, and LVW, were similar before and after inhibition of lipolysis.

Table II shows metabolic data before and after inhibition of lipolysis. While isoproterenol infusion raised plasma FFA and myocardial uptake of FFA two- to threefold in control experiments, these increases were abolished by nicotinic acid or beta pyridyl carbinol. Infusion of isoproterenol raised $\dot{M}\dot{V}O_2$ in all experiments. However, the augmentation was larger ($P < 0.005$) before (on average 7.7 ml/min·100 g) than after (on average 4.5 ml/min·100 g) inhibition of lipolysis. MF increased similarly with isoproterenol before (on average 63 ml/min·100 g) and after (on average 68 ml/min·100 g) inhibition of lipolysis. The higher $\dot{M}\dot{V}O_2$ during

TABLE I
Hemodynamic Effects of Isoproterenol Infusion (2–3 µg/min) before and after Inhibition of Lipolysis by Nicotinic Acid or β-Pyridyl Carbinol, Mean ±SE of Five Dogs

	$\bar{A}P$	LVSP	LVEDP	dP/dt	HR	CO	LVW
	mm Hg	mm Hg	mm Hg	(mm Hg·10 ⁻³)/sec	beats/min	ml/min·kg	kg-m/min·kg
Control	132	144	4.4	2.5	160	131	0.25
±SE	6	6	1.3	0.3	14	11	0.02
Isoproterenol	121	137	3.2	5.4	185	225	0.42
±SE	7	6	1.2	0.5	13	13	0.03
<i>P</i>	NS	NS	<0.05	<0.001	<0.02	<0.005	<0.01
Inhibition of lipolysis							
Control	132	144	4.4	2.5	160	130	0.25
±SE	6	6	1.3	0.3	15	11	0.02
Isoproterenol	122	138	3.1	5.5	185	226	0.42
±SE	4	6	1.2	0.5	13	14	0.02
<i>P</i>	<0.05	NS	<0.05	<0.001	<0.02	<0.001	<0.005

$\bar{A}P$, mean aortic blood pressure; LVSP, left ventricular systolic pressure; LVEDP, left ventricular end-diastolic pressure; dP/dt, maximal rate of rise of left ventricular pressure; HR, heart rate; CO, cardiac output; LVW, left ventricular work; NS, not significant ($P > 0.05$).

TABLE II
*Metabolic Effects of Isoproterenol Infusion (2–3 µg/min) before and after Inhibition of Lipolysis
by Nicotinic Acid or β-Pyridyl Carbinol, Mean ±SE of Five Dogs*

	FFA			M $\dot{V}$ O ₂	O ₂ -extr.	MF	Hb
	a	a-cs	u				
	µEq/liter	µEq/liter	µEq/min·100g	ml/min·100g	%	ml/min·100g	g/100 ml
Control	449	145	9.5	9.9	72.1	100	11.1
±SE	95	37	2.8	0.8	4.8	8	0.6
Isoproterenol	1094	209	21.6	17.6	72.7	163	12.1
±SE	160	25	4.3	1.2	4.8	26	0.8
P	<0.02	<0.05	<0.05	<0.001	NS	<0.05	NS
Inhibition of lipolysis							
Control	350	143	9.4	9.7	70.2	100	11.1
±SE	60	37	2.9	1.0	5.5	8	0.5
Isoproterenol	340	62	6.1	14.2	59.0	168	11.7
±SE	49	13	1.2	1.2	5.6	23	0.8
P	NS	NS	NS	<0.001	NS	<0.025	NS

a, arterial concentrations; a-cs, arteriocardiac sinus differences; u, myocardial uptake of free fatty acids (FFA); M $\dot{V}$ O₂, myocardial oxygen consumption; O₂-extr., myocardial oxygen extraction; MF, myocardial blood flow; Hb, hemoglobin; NS, not significant ($P > 0.05$).

isoproterenol infusion in control experiments was due to larger ($P < 0.01$) myocardial oxygen extraction, as opposed to increased coronary flow, than with inhibited lipolysis.

Group II. The isoproterenol-induced changes in mechanical activity, as evidenced by $\overline{AP}$, LVSP, LVEDP, dP/dt, HR, CO, and LVW, were similar before and after inhibition of lipolysis by high plasma glucose concentrations (Table III).

Table IV shows metabolic data before and after inhibition of lipolysis. In control experiments, i.v. infusion of isoproterenol raised plasma FFA three- to four-

fold and myocardial uptake of FFA 5–6 times. At high plasma glucose concentrations, the increments in plasma FFA and myocardial uptake of FFA were less than in control experiments, averaging only 56 and 21%, respectively, of the increments seen in the control experiments. Isoproterenol infusion raised M $\dot{V}$ O₂ in all experiments. However, the augmentation of M $\dot{V}$ O₂ was larger ($P < 0.02$) before (on average 7.5 ml/min·100 g) than after (on average 4.3 ml/min·100 g) inhibition of lipolysis. MF increased similarly in control experiments (on average 57 ml/min·100 g) and by inhibition of lipolysis (on average 50 ml/min·100 g). Higher M $\dot{V}$ O₂

TABLE III
*Hemodynamic Effects of Isoproterenol Infusion (2–3 µg/min) before and after Inhibition of Lipolysis
by High Plasma Concentrations of Glucose, Mean ±SE of Five Dogs*

	$\overline{AP}$	LVSP	LVEDP	dP/dt	HR	CO	LVW
	mm Hg	mm Hg	mm Hg	(mm Hg·10 ⁻³)/sec	beats/min	ml/kg·min	kg-m/min·kg
Control	138	142	3.4	2.7	134	156	0.31
±SE	5	5	0.4	0.0	5	13	0.04
Isoproterenol	125	139	1.8	5.6	165	240	0.45
±SE	5	3	0.5	0.4	6	15	0.04
P	<0.05	NS	<0.025	<0.005	<0.001	<0.001	<0.001
Inhibition of lipolysis							
Control	138	142	3.4	2.7	134	155	0.31
±SE	5	5	0.4	0.0	5	14	0.04
Isoproterenol	126	141	2.2	5.8	161	250	0.47
±SE	3	5	0.4	0.4	5	15	0.05
P	<0.02	NS	NS	<0.005	<0.001	<0.005	<0.005

For abbreviations, see Table I.

TABLE IV
*Metabolic Effects of Isoproterenol Infusion (2–3 µg/min) before and after Inhibition of Lipolysis
by High Plasma Glucose Concentrations, Mean ±SE of Five Dogs*

	FFA			M $\dot{V}$ O ₂	O ₂ -extr.	MF	Hb
	a	a-cs	u				
	µEq/liter	µEq/liter	µEq/min·100g	ml/min·100g	%	ml/min·100g	g/100 ml
Control	532	70	4.9	8.2	60.5	96	11.6
±SE	97	32	2.4	1.7	5.8	7	0.7
Isoproterenol	1843	297	26.3	15.7	65.1	153	13.1
±SE	279	68	5.2	2.0	4.8	13	0.7
P	<0.005	=0.01	<0.005	=0.005	NS	<0.05	NS
Inhibition of lipolysis							
Control	452	69	4.6	8.3	61.9	96	11.1
±SE	104	32	2.5	1.7	5.5	7	0.9
Isoproterenol	1181	109	9.0	12.0	59.8	144	11.4
±SE	82	29	1.8	1.4	4.8	16	0.4
P	<0.005	NS	NS	<0.005	NS	<0.05	NS

For abbreviations, see Table II.

during isoproterenol infusion before than after high plasma glucose concentrations was primarily due to larger ($P < 0.02$) myocardial oxygen extraction.

DISCUSSION

Catecholamine-induced changes in M $\dot{V}$ O₂ in beating hearts have usually been attributed to inotropic and chronotropic stimulation of the mechanical activity of the heart (1, 2). However, the isoproterenol-induced rise in M $\dot{V}$ O₂ was significantly reduced after inhibition of lipolysis, despite similar effects on mechanical activity of the heart, as evidenced by LVSP, LVEDP, dP/dt, HR, and LVW. The present study therefore supports the concept that the calorogenic action of catecholamines in the beating heart is partly attributable to their FFA-mobilizing action as shown in the isolated heart (16, 17).

A similarly reduced isoproterenol-induced rise in M $\dot{V}$ O₂ with antilipolytic drugs or high plasma glucose concentrations indicates that this effect was not confined to the agents per se. The common cause was probably a largely abolished rise in myocardial uptake of FFA by isoproterenol in both groups, suggesting a causal relationship between myocardial uptake of FFA and oxygen consumption. In the presence of nicotinic acid or beta pyridyl carbinol, this can be solely explained by the abolished rise in plasma FFA as a consequence of complete inhibition of lipolysis in adipose tissue. However, as the isoproterenol-induced rise in plasma FFA was reduced by an average of only 44% in the glucose series, an additional suppressive effect of high glucose concentrations on myocardial uptake of FFA must be postulated in order to explain our findings.

The present data indicate that the larger rise in M $\dot{V}$ O₂ by isoproterenol before than after inhibition of lipolysis is related to the high myocardial uptake of circulatory FFA. However, previous in vitro studies have suggested increased endogenous lipolysis within the heart by catecholamines (16, 17). It is therefore conceivable that increased myocardial lipolysis would contribute to the rise in M $\dot{V}$ O₂ in addition to the effect of circulatory FFA. Thus M $\dot{V}$ O₂ would be expected to increase more for a given rise in myocardial uptake of FFA during isoproterenol infusion than for a similar exogenously induced rise in myocardial uptake of FFA. In accordance with this interpretation, the present study shows an almost doubled rise in M $\dot{V}$ O₂ for a similar increase in myocardial uptake of FFA with isoproterenol compared with that induced by i.v. infusion of Intralipid (Vitrium, Stockholm, Sweden) in heparinized dogs (3).

The higher M $\dot{V}$ O₂ with isoproterenol alone was principally due to larger myocardial oxygen extraction than after inhibition of lipolysis. If Po₂ were the only determinant of MF, it would be expected that with isoproterenol alone MF would be larger than after inhibition of lipolysis. However, as proposed by Berne (18), metabolites like adenosine probably play an important role in the regulation of MF. The present experiments appear to suggest that regulation of MF, and probably also the production of adenosine, is not completely dependent on myocardial metabolism, but seems to be better related to mechanical activity.

The results of this study imply that inhibition of lipolysis by nicotinic acid, beta pyridyl carbinol, or high plasma glucose concentrations can substantially reduce the catecholamine-induced rise in M $\dot{V}$ O₂ without affecting the supporting action of catecholamines on myocardial

performance. This observation might be utilized in the treatment of patients with high sympathetic activity, e.g., patients with myocardial infarction associated with high values of plasma FFA. Although the beneficial effect of glucose is known in isolated hearts during oxygen deprivation and subsequent recovery (19–22), clinical trials with glucose, potassium, and insulin in patients with acute myocardial infarction have yielded conflicting results (23–25). However, as pointed out by Owen, Thomas, and Opie (26), and by Henderson, Most, Parmley, Gorlin, and Sonnenblick (27), this treatment has been directed rather towards replacing intracellular potassium than depressing myocardial uptake of FFA through high plasma glucose concentrations. As a result, plasma FFA and myocardial uptake of FFA were not measured in these studies.

The exact mechanism for the metabolic stimulation of high concentrations of FFA is not known. It may be due to an uncoupling action of high intracellular concentrations of FFA on oxidative phosphorylation (16, 17, 28).

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