BETA ADRENERGIC BLOCKERS AND LIPID METABOLISM. I. CHANGES OF SERUM FREE FATTY ACIDS AFTER SINGLE PROPRANOLOL APPLICATION IN RATS

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Key-words: propranolol — lipid metabolism — free fatty acids — lipolysis — gas-liquid chromatography

Changes of plasma lipid levels present one of the most essential coronary risk factors after hypertension and smoking. Some authors (11) note that the appearance of a myocardial infarction can be predicted by means of the analysis of the fatty-acid composition of serum lipids.

Beta adrenergic blockers which have occupied stable positions in antihypertensive therapy as well as in the treatment of the coronary disease and some arrhythmias could not demonstrate a definite preventive effect on ischemic accidents probably because of unfavourable metabolic effects especially concerning plasma lipids (10). A series of investigations revealed that both non-selective and selective (not rarely) beta blockers induced plasma triglyceride level increase (5, 14, 15) and reduced HDL-cholesterol level and HDL-cholesterol: LDL cholesterol ratio (8, 10, 15). It is found out that there is an antagonistic action upon isoprenaline-induced increase of plasma free fatty acids (FFA) in healthy volunteers (7) as well as an initial serum FFA reduction after prolonged beta blocker application (9). There are scanty data about beta blocker influence upon tissue or plasma individual FFA levels (12).

The oblect of the present communication are the first results from our investigation of the influence of propranolol, the most-widely used non-selective beta blocker, on lipid metabolism. Both dose- and time-dependences of propranolol action on serum levels of eight individual FFA in white male rats are studied

Material and methods

The study covered 72 white male rats with b. w. of 150.0 — 170.0 g. Propranolol was intraperitoneally applied at doses of 1 mg, 3 mg and 9 mg/kg b. w. Animals were killed by cutting of the sublingual veins on the 1st, 2nd and 4th hour after drug administration. Control animals injected with saline were also killed for corresponding dosages and intervals of drug injections. Serum was extracted after Folsh's et al. method (1957). Lipid fractioning was performed by means of thin-layer chromatography in the system of hexane: aether: acetic acid (ratio 35 : 15 : 1) (1). FFAs were fluorescently visualized in ultraviolet light and quantitatively isolated. Then they were methylated with diazomethane and gas-chromatographically analyzed. The method was adapted on Chrom-4 apparatus with phase DEDS and chromosorb — W carrier, with flame-ionization detector. Quantities of single FFA: myristic acid (C_{14} : 0). palmitic acid

(C_{16} : 0), palmitoleic acid (C_{16} : 1), stearic acid (C_{18} : 0); oleic acid ($_{18}$: 1), linolic acid (C_{18} : 2), eicosantrienic acid (C_{20} : 3), and arachidonic acid (20 : 4): were calculated on the basis of intrinsic standard as arachidic acid (C_{20} : 0) added to the serum prior to extraction.

Results and discussion

Our results obtained demonstrated a linear dose- and time-dependent reduction of total FFA concentrations (fig. 1). At dose of 9 mg/kg b. w., total FFAs decrease linearly during the period from the first to the fourth hour after injection. On the 4th hour, this reduction is by 43.4 per cent and statistically significant in comparison with that of the control values. However, at dose of 3 mg-kg b. w., reduction is by 20.3 per cent only.

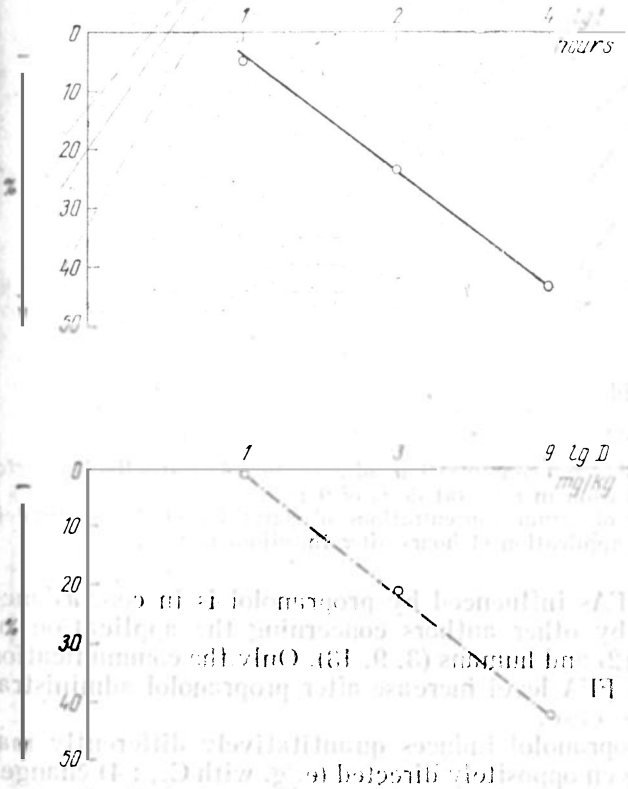

Fig. 1. — a. Time-dependent changes of serum free fatty acid concentrations after single i. p. propranolol application (at dosis of 9 mgk/g) in rats.
Fig. 1. — b. Dose-dependent changes of serum free fatty acid concentrationsa after single i. p. propranolol application (4 hours after injection) in rats.

Quantitatively, individual FFA decrease is differently expressed. It varies between 17.5 per cent (for C_{16} : 1) and 57.3 per cent (for C_{18} : 2) at propranolol dose of 9 mg/kg b. w. and 4-hour interval. Reduction of concentrations of the most important acids in FFA pool, namely C_{16} : 0 and C_{18} : 1, is close to the decrease of total FFAs — by 38.5 and 45.1 per cent, respectively ($p < 0.001$) (fig. 2). Similarly to the case of total FFAs, propranolol effect on the levels of these two FFAs demonstrates a linear time- and dose-dependence. C_{18} :2 submits to the same dependence, too (fig. 2). Its maximal reduction is by 57.3 per cent at doses of 9 mg/kg b. w. and 4-hour interval($p <$ F.00). C_{14} : 0 and C_{16} : 0 both show a linear dose-dependence (fig. 3). The rest three dFAs: C_{18} : 1, C_{20}: 4, and C_{20} : 3 do not strictly submit to these dependences. Arachitonic acid increases at the lowest propranolol dose used (fig. 4). This result is rather interesting on the background of total FFA reduction and it cannot be explain-

ed by propranolol blockade of beta-adrenoceptors. Besides as it is well-known, propranolol is deprived of intrinsic sympathicomimetic activity. At doses of 3 mg and 9 mg/kg b. w. of the beta blocker, arachidonic acid levels decrease (fig. 4), statistically significantly at dose of 9 mg/kg b. w. ($p < 0.01$).

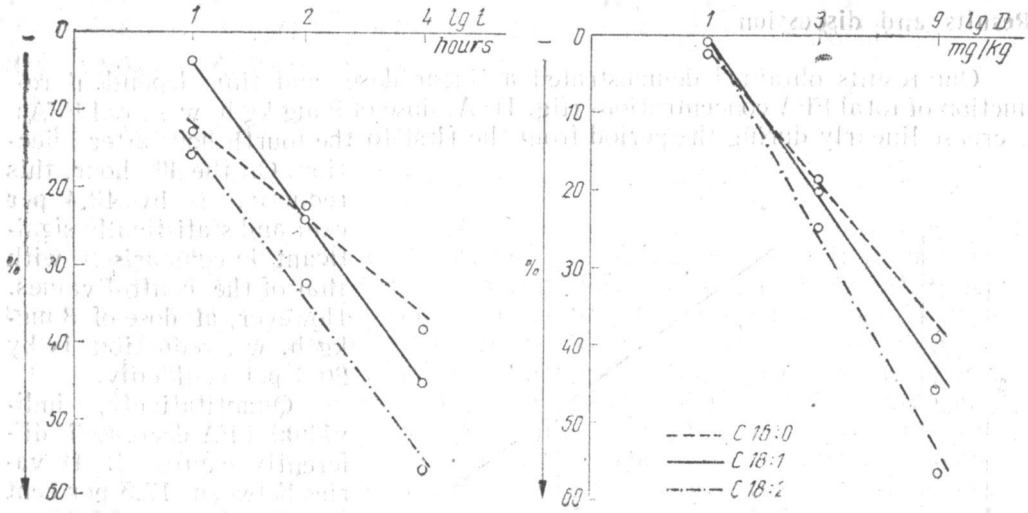

Fig. 2. Legend:

o Palmitic acid
x Oleic acid
O Linoleic acid

Fig. 2-a. Time-dependent changes of serum concentrations of palmitic, oleic and linoleic acids after single i. p. propranolol application in rats. (at dosis of 9 mg/kg)
Fig.. 2-b. Dose-dependent changes of serum concentrations of palmitic, oleic and linoleic acids after single i. p. propranolol application (4 hours after injection) in rats.

This reduction of total FFAs influenced by propranolol is in concordance with FFA decrease reported by other authors concerning the application of the same beta-blocker in dogs (2) and humans (3, 9, 13). Only the communication of Tanaka et al. (1976) about FFA level increase after propranolol administration is an exception in this case.

These data show that propranolol induces quantitatively differently manifested and in certain cases even oppositely directed (e. g. with C_{16} : 4) changes in individual serum FFA levels under the condition of our trial. It allows us to assume that beta-adrenergic control of lipolysis is of different importance for single FFAs.

The significant decrease of linoleic acid (C_{18} : 2) concentration seems to be particularly important when characterizing propranolol action because it is established that this acid reduces in plasma cholesterol esters of patients with ischemic heart disease and myocardial infarction (11).

The changes of serum arachidonic acid levels revealed in our study present a certain interest, too. Miura et al. (1988) report that under ischemic conditions of the myocardium in dogs on the background of total moderate FFA

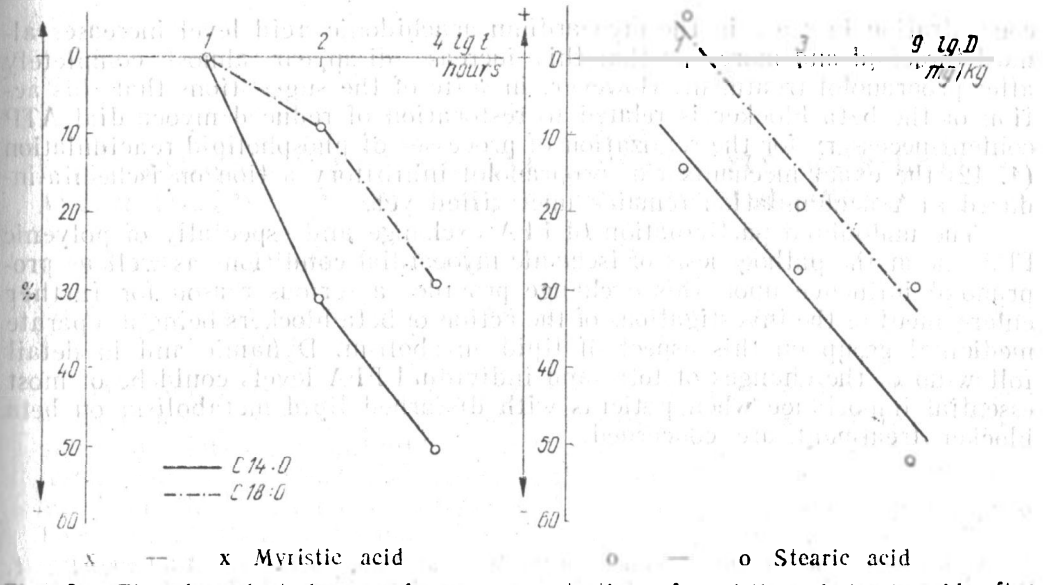

Fig.. 3-a. Time-dependent changes of serum concentrations of myristic and stearic acids after single i. p. propranolol application (at dosis 9 mg/kg) in rats.
Fig.. 3-b. Dose-dependent changes of serum concentrations of myristic and stearic acids after single i. p. propranolol application (4 hours after injection) in rats.

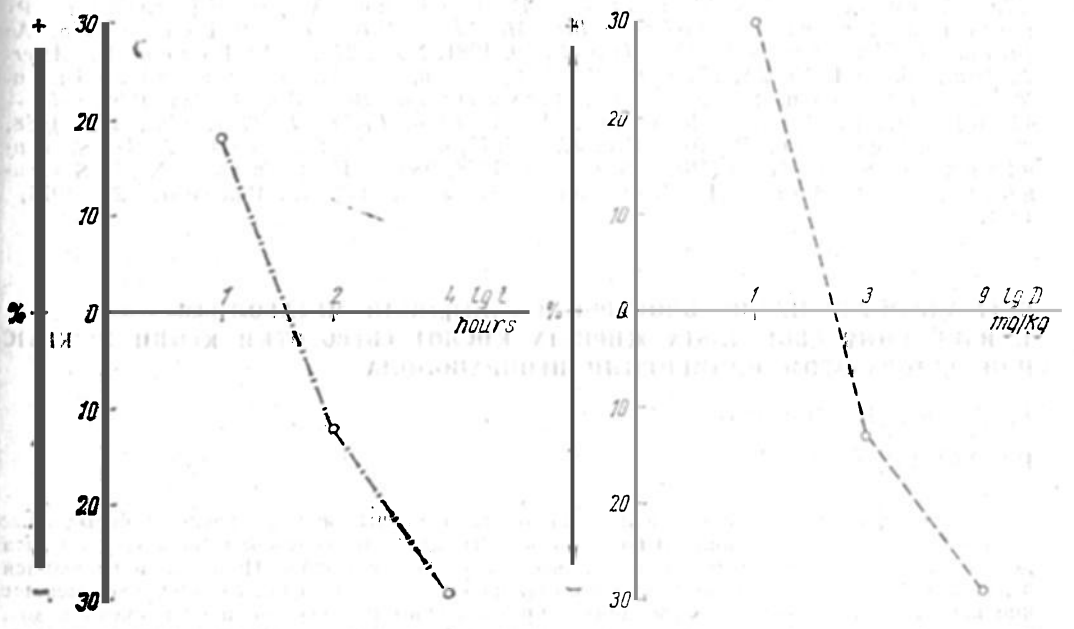

Fig. 4-a. Time-dependent changes of serum concentration of arachidonic acid after single i. p. propranolol application (at dosis of 9 mg/kg) in rats.
Fig. 4-b. Dose-dependent changes of serum concentration of arachidonic acid after single i. p. propranolol application (4 hours after injection) in rats.

concentration increase in the myocardium arachidonic acid level increases almost threefold and more and that this increase disappear almost completely after propranolol treatment. However, in spite of the suggestions that this action of the beta blocker is related to restoration of reduced myocardial ATP content necessary for the realization of processes of phospholipid reacidulation (4. 12) the exact mechanism of propranolol inhibitory action on ischemia-induced FFA accumulation remains unclarified yet.

The undoubted participation of FFA exchange and especially of polyenic FFA one at the pathogenesis of ischemic myocardial conditions as well as propranolol influence upon this exchange provides a serious reason for further enlargement of the investigations of the action of beta blockers being a separate medicinal group on this aspect of lipid mtabolism. Dynamic and in detail follow-up of the changes of total and individual FFA levels could be of most essential importance when patients with disturbed lipid metabolism on beta blocker treatment are concerned.

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БЕТА-АДРЭНЕРГИЧЕСКИЕ БЛОКЕРЫ И ЛИПИДНЫЙ МЕТАБОЛИЗМ I. ИЗМЕНЕНИЯ СВОБОДНЫХ ЖИРНЫХ КИСЛОТ СЫВОРОТКИ КРОВИ У КРЫС ПРИ ОДНОРАЗОВОМ ПРИМЕНЕНИИ ПРОПРАНОЛОЛА

В. Михова, М. Мангарова, Д. Желязков

РЕЗЮМЕ

Исследовано влияние пропранолола на восемь свободных жирных кислот у белых крыс самцов на сывороточном уровне. Свободные жирные кислоты исследовались за 1, 2 и 4 часа до введения пропранолола и столько же часов после его введения. Пропранолол вводился в дозах 1, 2 и 3 мг/кг с использованием газжидкой хроматогрифии. Установлено уменьшение концентрации тотальных свободных жирных кислот в зависимости от времени и доз. Эта зависимость линейного характера. Зависимости от дозы и времени вызывают количественно различные изменения уровня индивидуальных свободных жирных кислот.

Обсуждается различное значение адрэнергического контроля липолиза для отдельных свободных жирных кислот.