Plasma prostaglandin levels in fed and starved lean, normal and obese women

P. B. Curtis-Prior[1,2], M. Jenner, M. Smethurst and J. W. Woodward

Research Department, The Marie Curie Memorial Foundation, Oxted (Surrey) and General Practicioner Unit, Granville Road, Sidcup (Kent, England), 13 September 1978

Summary. The plasma obtained from fed and starved lean, normal and obese women was estimated, by a radio-immunoassay method, for prostaglandins, owing to their implication in the regulation of adipose tissue lipolysis and the development of obesity. No significant differences were found due to nutritional status or body-build. However, a significantly higher plasma concentration of prostaglandins of the E-type than of the F-type, was found consistently. The very low levels of prostaglandins observed (a range of 0.10–0.15 ng ml^{-1} for E-type and a range of 0.05–0.07 ng ml^{-1} for the F-type) may be due, in part, to the activity of a plasmatic prostaglandin metabolizing system.

It has been proposed that prostaglandins (PG) regulate, in part, lipolysis in adipose tissue[3,4] and that their activity may be a function of nutritional status[5,6]. Also prostaglandins have been credited with a role in the development of human obesity as physiological inhibitors of the release of free fatty acids from calorie stores in adipose tissue[7]. Thus, we compared the levels of PG in venous blood in lean, normal and obese women under fed conditions and following an overnight fast, to investigate a possible role of prostaglandins as mediators of intermediary lipid metabolism.

Methods. Apparently healthy women attending their General Practicioner for contraceptive advice/therapy or for other reasons, were invited to participate in this study concerned with body weight and prostaglandin levels. The 32 who gave their informed consent were divided into a lean (PI > 13), a normal (PI < 13) and obese group (PI ⩽ 12) on the basis of their ponderal index[8]. The mean values of each group are shown in table 1. Each subject attended the clinic on 2 occasions: once following her normal breakfast meal, and once after 12 h overnight fast. 20 ml of whole blood was withdrawn from a cubital vein and transferred into lithium-heparinized tubes, mixed thoroughly and placed on ice immediately, prior to transfer to the laboratory in a Dewar flask containing coolant to minimize prostaglandin degradation. Anticoagulated blood was centrifuged at 40,000 × g for 20 min at +4 °C. The plasma separated and deep-frozen at −15 °C until estimated for PG by a radio-immuno assay method[9,10].

Results. The results are summarized in table 2. There were no significant differences found in the levels of the same type of PG between fed and starved plasma samples within groups, nor were there any significant differences of PGE or PGF levels between the lean, normal and obese groups. The actual levels of each PG were very similar from one group to another. Because of these similarities the values for each type of prostaglandin from fed and starved plasma were pooled to give a single value for PGE for each group and a single value for PGF for each group. In all groups of

Table 1. Details of volunteer female subjects

Group type	n	Group mean age (years)	Body weights (kg)	Height (m)	Ponderal index*
			48.16	1.69	14.06
			50.00	1.69	13.88
			47.27	1.65	13.83
			55.00	1.73	13.79
			47.72	1.65	13.77
			50.45	1.68	13.72
			52.72	1.66	13.42
			51.36	1.64	13.35
			60.45	1.73	13.33
			50.91	1.63	13.28
Lean	10	39.3 ± 1.7**			13.64 ± 0.09
			59.55	1.68	12.99
			63.64	1.70	12.91
			62.27	1.69	12.89
			62.72	1.68	12.77
			63.64	1.68	12.72
			74.32	1.75	12.61
			59.55	1.63	12.60
			72.27	1.73	12.55
			57.95	1.60	12.52
			61.36	1.60	12.28
			81.82	1.75	12.21
			61.36	1.59	12.18
			70.23	1.65	12.10
Normal	13	43.5 ± 2.6			12.56 ± 0.08
			62.27	1.57	12.02
			76.36	1.68	11.96
			61.59	1.55	11.87
			88.18	1.73	11.74
			84.09	1.68	11.58
			80.91	1.64	11.46
			72.73	1.52	11.05
			100.91	1.68	10.89
			110.91	1.71	10.80
Obese	9	46.3 ± 3.9			11.49 ± 0.16

* Height in inches/$3\sqrt{\text{body weight in pounds}}$. ** $\bar{x} \pm$ SEM.

Table 2. Plasma levels (ng/ml) of prostaglandins (PGs) in human subjects

Group Type	Prostaglandins E			F		
	Nutritional status Fed	12-h-starved	Pooled*	Fed	12-h-starved	Pooled*
Lean	0.15 ± 0.03 (5)**	0.12 ± 0.02 (5)	0.14 ± 0.03 (10)	0.06 ± 0.01 (10)	0.06 ± 0.01 (10)	0.06 ± 0.0 (20)
Normal	0.12 ± 0.02 (12)	0.11 ± 0.01 (12)	0.12 ± 0.02 (24)	0.05 ± 0.00 (13)	0.06 ± 0.01 (13)	0.06 ± 0.0 (26)
Obese	0.10 ± 0.01 (5)	0.14 ± 0.03 (7)	0.12 ± 0.02 (12)	0.06 ± 0.00 (8)	0.07 ± 0.01 (9)	0.07 ± 0.0 (17)

* Combined fed and 12-h-starved values. ** Numbers in parenthesis indicate number of observations used in the calculation of the mean ± SEM. The discrepancy between these numbers and the numbers of subjects in the various groups shown in Table 1 is owing to elimination of plasmas which showed variation of PG levels with storage, as described in the text.

plasma (pooled values) there was significantly ($p < 0.01$) more PGE than PGF. Some variation in PG levels was found following storage of some of the plasmas for 7 weeks and in these cases, both values were discounted.

Discussion. The lack of differences in primary prostaglandin levels in the plasma in women of different body constitution and of different nutritional status lends little support, per se, to the prostaglandin hypothesis of obesity[7]. This result showing a significantly higher plasma concentration of PGE than PGF is at variance with a previous study when greater amounts of PGF than PGE were observed in human serum[11]. However, the absolute levels of PGE in our samples are compatible with previous observations[12]. Some of the discordant results of different studies may be explained by the presence of a PG-metabolizing system in plasma which reduces, significantly, plasma levels of PG within min in vitro[13]. This system of PG degradation plus that of the lungs and other vascular beds known to operate[14], indicate that elucidation of any role for prostaglandins, in vivo, must be accompanied by rapid removal of blood, and rapid 'fixing' of the blood sample in order to minimize PG degradation.

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PRO EXPERIMENTIS

A micro-electrode amplifier with an infinite resistance current source for intracellular measurements of membrane potential and resistance changes under current clamp

H. Muijser[1]

Department of Biophysics, Laboratorium voor Algemene Natuurkunde, Rijksuniversiteit Groningen, Groningen (The Netherlands), 23 March 1978

Summary. A microelectrode amplifier for intracellular electrophysiological research is described. It is equipped with an electronic infinite resistance constant current source for the injection of current into biological cells. With this amplifier the potential changes, the resistance changes and the dependence on extrinsic current of single cells can be measured independently and simultaneously.

Investigators of invertebrate visual sensory cells are repeatedly faced with the need to measure simultaneously the membrane potential of single cells, their resistance changes, and the dependence on extrinsic current (current clamp) of these membrane properties.

The latter types of measurements are carried out with a single intracellular microelectrode by extending the microelectrode amplifier for the membrane potential measurements with a current source in a bridge circuit arrangement. The drawbacks of the currently applied circuits and an improved design are discussed in this note.

The classical bridge circuit[2] for measuring resistance changes is given in figure 1. The membrane potential is measured through the microelectrode (with resistance R_e) and the follower amplifier A_1. Current is sent into the cell by feeding a large resistor R_s with a potential E_s (DC for current clamp and AC square waves for resistance measurements). The voltage drop over the electrode resistance (R_e) and the membrane resistance (R_m) is balanced by the output ($-E_sR_2/R_1$) of amplifier A_2. The bridge is in balance when

$$R_2/R_1 = (R_e + R_m)/R_s. \quad (1)$$

The changes in the membrane resistance (R_m) upon illumination appear as an imbalance of the bridge.

In the usual Wheatstone bridge the output is measured differentially, and the input of the bridge is fed single-ended whereas in the circuit of figure 1 the bridge output is measured single-ended, and the input is fed differentially.

The circuit of figure 1 suffers from several drawbacks.

1. The potential measured is not the membrane potential V_m but a potential V_o via the potential divider consisting of ($R_e + R_m$) and R_s, and the gain of amplifier A_2 which equals unity to a very good approximation:

$$V_o = \frac{R_s}{(R_s + R_e + R_m)} \cdot V_m \quad (2)$$

with $R_s = 1000$ MΩ and $R_e + R_m = 100$ MΩ (typical values); this means a systematic error of 9%.

2. The cell is loaded. With R_s grounded, $V_m = -60$ mV and $R_s + R_e + R_m = 1100$ MΩ a current of 55 pA is loading the cell which is much more than the input current of A_1 (< 1 pA).

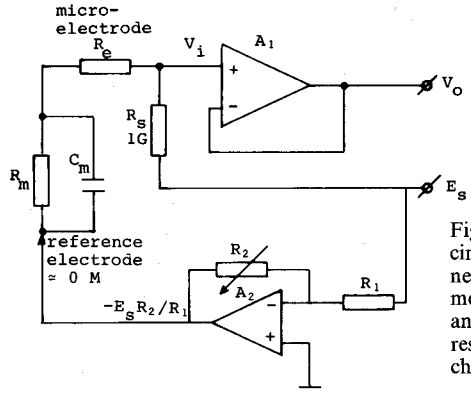

Fig. 1. Classical bridge circuit for simultaneously measuring the membrane potential and the membrane resistance (R_m) changes of single cells.