explain the first phenomenon; increased cardiac output in the presence of a sustained vasoconstriction, as first suggested by Lands & others (1950), would seem to explain the second phenomenon.

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Effects on lipomobilisation of the β-adrenergic blocking drugs, propranolol and INPEA

SIR,—An increase of plasma free fatty acids (FFA) occurred within 60 min after subcutaneous administration of propranolol [1-isopropylamino-3-(1-naphthyloxy)-2-propanol hydrochloride] (Black, Crowther, Shanks, Smith & Dornhorst, 1964) to rats. The rise was more evident with low doses and disappeared with increasing dosage (Table 1). In contrast, (±)-INPEA (*N*-isopropyl-*p*-nitrophenylethanolamine hydrochloride) (Somani & Lum, 1965) diminished plasma FFA at lower doses while, at greater doses, it did not induce significant changes in FFA level. The results obtained with the two optical isomers seem to indicate that a mild lipid-mobilising power is linked only to (−)-INPEA (Table 1).

The lipomobilising activity of propranolol *in vivo* was prevented by previous reserpinisation or treatment with dibenzyline (Table 2). Thus propranolol action on lipolysis *in vivo* is apparently an indirect adrenergic one.

Regarding the antagonistic action against the noradrenaline-induced lipomobilisation, propranol and (±)-INPEA are equally active *in vivo* (Table 3). The inhibitory power of INPEA appears to be greater in the (−)-isomer (Table 3).

In vitro propranolol and INPEA did not show any intrinsic lipomobilising activity on rat epididymal adipose tissue. On the contrary, at high concentrations (2 and 20 × 10^{-5}M) they depressed the basal lipolytic activity.

The antagonism of propranolol and INPEA against the FFA mobilisation stimulated by noradrenaline *in vitro* was studied according to a procedure previously described (Fassina, Tóth & Santi, 1965). The curves obtained by plotting the log concentration of noradrenaline against the amount of FFA released in the presence of increasing concentrations of propranolol and INPEA, indicate that the two β-adrenergic blocking drugs behave as competitive antagonists. The pA_2 values (Schild, 1947) (calculated when the effect of noradrenaline was 50% of the maximal) show that (−)-INPEA ($pA_2 = 6{\cdot}32$) is less active than propranolol ($pA_2 = 6{\cdot}75$) whilst (+)-INPEA ($pA_2 = 4{\cdot}20$) has a very small activity. From these values the affinity of (−)-INPEA for the lipid mobilising sites affected by noradrenaline gives results about 130 times higher than that of (+)-INPEA and 3 times lower than that of propranolol. This

striking quantitative dependence of the competitive antagonism on the steric configuration of the ethanolamine side-chain indicates that this part of the molecule of INPEA is involved in occupying the specific active sites for catecholamines in adipose tissue.

From the comparison of propranolol, (+)-INPEA and (−)-INPEA it seems that (*a*) propranolol has a greater lipomobilising action *in vivo* than INPEA, (*b*) propranolol is more active *in vitro* than INPEA, (*c*) the increase in FFA and the antagonistic action are both greater in the (−)-isomer of INPEA. These facts suggest that the lipomobilising and the antiadrenergic properties are related. The question now arises how the two actions may be connected. Other β-adrenergic

TABLE 1. EFFECT OF PROPRANOLOL AND INPEA ON PLASMA FREE FATTY ACID (FFA) LEVEL IN RATS

Drug	Dose and route	FFA % variation*	P†
Propranolol	2 mg/kg s.c.	+ 58 ± 9	<0·01
	5	+ 54 ± 8	<0·001
	40	+ 15 ± 6	n.s.
(±)-INPEA	1·65 mg/kg s.c.	− 30 ± 6	<0·01
	4·1	− 22 ± 3	<0·01
	33·0	+ 9 ± 5	n.s.
(−)-INPEA	2 mg/kg i.p.	+ 17 ± 3	<0·02
	10	+ 40 ± 6	<0·001
(+)-INPEA	2 mg/kg i.p.	− 8 ± 8	n.s.
	10	− 8 ± 3	n.s.

Male Sprague-Dawley fed rats (200 ● 30 g) were used. Animals were killed 60 min after treatment s.c. and 30 min after treatment i.p. Subcutaneous doses of propranolol and (±)-INPEA are equimolar, corresponding respectively to 7, 17 and 135 μM/kg. FFA were determined according to Dole (1956).
* Each value represents the mean ± s.e. of 5 to 12 rats.
† P = significance of the difference from the control (saline treated) group.

TABLE 2. EFFECT OF RESERPINE AND DIBENZYLINE ON THE FREE FATTY ACID (FFA) MOBILISATION INDUCED BY PROPRANOLOL IN RATS

Treatment	FFA % variation*	P†
Propranolol	+ 85 ± 4	<0·001
Reserpine + propranolol	− 18 ± 3	<0·05
Dibenzyline + propranolol	+ 1 ± 2	n.s.

Propranolol, 10 mg/kg i.p. 30 min before killing. Reserpine, 3 mg/kg i.p. repeated twice, 40 and 15 hours before propranolol. Dibenzyline, 10 mg/kg i.p. 2 hr before propranolol.
* Each value represents the mean ± s.e. of 8 rats.
† P = significance of the difference from the respective control group (treated with saline or with antagonistic drug + saline).

TABLE 3. ANTAGONISTIC EFFECT OF PROPRANOLOL AND INPEA ON THE INCREASE OF PLASMA FREE FATTY ACIDS (FFA) INDUCED BY NORADRENALINE IN RATS

Treatment	FFA % variation*	P†	Inhibition
Noradrenaline	+ 85 ± 2	<0·001	—
Propranolol + noradrenaline ..	+ 31 ± 2	<0·001	64%
(±)-INPEA + noradrenaline ..	+ 29 ± 5	<0·01	66%
Noradrenaline	+ 80 ± 7	<0·001	—
(−)-INPEA + noradrenaline ..	+ 36 ± 4	<0·001	55%
(+)-INPEA + noradrenaline ..	+140 ± 7	<0·001	0

Noradrenaline, 0·5 mg/kg i.p. 30 min before killing. Propranolol, 40 mg/kg s.c. 15 min before noradrenaline. (±)-, (+)- and (−)-INPEA, 33 mg/kg s.c. 15 min before noradrenaline. The doses of propranolol and INPEA are equimolar.
* Each value represents the mean ± s.e. of 8 rats.
† P = significance of the difference from the respective control group (treated with saline or with the antagonist + saline).

blockers (DCI and pronethalol) show a lipomobilising action together with a blocking effect against the catecholamine-induced FFA mobilisation. However, these compounds also stimulate lipolysis *in vitro*. These contrasting actions are generally ascribed to a direct dual effect on adipose tissue—a lipolytic and an antiadrenergic one—depending on the dose (Fröberg & Orö, 1963; Love, Carr & Ashmore, 1963; Westermann & Stock, 1963; Schusterová, Krčíková, Mühlbachová, Hynie & Wenke, 1964; Kvam, Riggilo & Lish, 1965; Love, Carr & Ashmore, 1965). On the other hand, propranolol is completely devoid of a direct lipolytic action *in vitro*, while its lipomobilising action *in vivo* is an indirect adrenergic one. The low content of endogenous catecholamines in adipose tissue (Paoletti, Smith, Maickel & Brodie, 1961; Sidman, Perkins & Weiner, 1962; Stock & Westermann, 1963) could explain the lack of FFA-releasing effect of propranolol *in vitro*. A similar condition exists with some indirect sympathomimetic amines, such as tyramine and amphetamine, which show a noticeable lipomobilising activity *in vivo* consequent to catecholamine release from nerve ending stores (Westermann & Stock, 1963; Fassina, 1964), but fail to manifest any lipomobilising action *in vitro*. Furthermore, they also antagonise the lipid mobilising effect of noradrenaline *in vitro* (Mühlbachová, Wenke, Schusterová, Krčíková & Elisová, 1964). The last fact seems to indicate that these drugs have some affinity for the receptor sites for catecholamines in adipose tissue (Mühlbachová & others, 1964). This affinity, being low, is completely masked *in vivo* by the catecholamine releasing action. From this comparison I am led to consider that propranolol behaves in a similar way to indirect acting sympathomimetic amines, but that it differs in intensity of lipomobilising effect *in vivo* and antiadrenergic action *in vitro*, probably because of a greater distribution coefficient and affinity towards the adrenergic receptors in adipose tissue than to the catecholamine stores. This difference appears to be further emphasized in (−)-INPEA.

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