blocking activity of the competitive type and it was interesting to note that the pA_2 value was similar to that reported for practolol (Lumley & Broadley, 1975).

Roszkowski, Strosberg & others (1972) observed a tachyphylaxis to tazolol in anaesthetized dogs and proposed an indirect mechanism for the cardiac stimulant effects of the compound. More recently Strosberg (1976) suggested that β-adrenoceptor blocking activity possessed by tazolol may also account for this phenomenon. The results of the present studies tend to support this contention.

In conclusion, this limited *in vitro* study has confirmed the selectivity of tazolol to β_1-adrenoceptors, but has shown that the compound is a partial agonist, and may act as an antagonist, at these receptors.

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The roles of presynaptic function and hepatic drug metabolism in the hypothermic actions of two novel dopaminergic agonists

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Hypothermia in rats acclimated to a cold environment has been attributed to stimulation of dopamine receptors in the cns (Yehuda & Wurtman, 1972). Accordingly, measurements of changes in core temperature of cold-acclimated rats has been used to assay for dopaminergic agonist activity of pharmacological compounds (Fuxe, Agnati & others, 1975; Calne, Claveria & Reid, 1975).

We report here the results of temperature studies on two new compounds proposed to have the properties of dopaminergic agonists—lergotrile [(+)-2-chloro-6-methylergoline-8-actonitrile] and 25–397 (9,10-didehydro-6-methyl-8β-(2-pyridylthiomethyl ergoline). These drugs were compared with bromocriptine, an ergot derivative successfully used for the treatment of parkinsonism (Calne, Teychenne & others, 1974). Although the three compounds have been described as dopamine agonists (Fuxe & others, 1975; Lew, Ohashi & Goldstein, 1976; Jaton, Loew & Vigouret, 1976) little is known about their mechanisms of action on dopaminergic pathways or the role of metabolism in their activation or inactivation. The results of some studies suggest that the integrity of presynaptic dopamine terminals is important for the activity of bromocriptine in inducing stereotypy and rotation in rats with unilateral lesions of the nigrostriatal pathway, while interruption of dopamine synthesis does not interfere with lergotrile-induced stereotypy (Pfeiffer & Silbergeld, 1976; Fuxe, Corrodi & others, 1974). Comparable studies have not been done on 25–397. This compound is reported to induce rotation analogously to bromocriptine and apomorphine; however, in contrast to other dopamine agonists, it does not induce stereotypy (Jaton & others, 1976; Silbergeld & Kennedy, unpublished observations).

The relatively long latent period between administration of bromocriptine and 25–397 and induction of their behavioural effects raises the possibility that their

* Correspondence.

activity depends upon conversion to active metabolites. This hypothesis is further supported by the reported failure of bromocriptine to affect the activity of dopamine-sensitive adenylate cyclase *in vitro* (Sarau & Foley, 1976).

These two issues—(1) the relative importance of pre- and postsynaptic aspects of dopaminergic functions and (2) the role of metabolism in conferring activity—were explored in hypothermia experiments, through administration of α-methyl-*p*-tyrosine (α-MT), an inhibitor of tyrosine hydroxylase, and administration of SKF 525A, an inhibitor of microsomal hepatic drug metabolism.

The results suggest that presynaptic elements are important in the hypothermic effects of bromocriptine, and that both bromocriptine and 25–397 may be metabolized to active compounds. Lergotrile does not apparently depend on presynaptic elements and may be inactivated by hepatic metabolism.

Male, Sprague-Dawley rats (Zivic Miller), 240–270 g, were placed, 4 to a cage, in a cold room (4°) 1 h before beginning core temperature measurements. All experiments were begun at 9.00 a.m. Core temperature was measured by inserting a thermister probe 6 cm rectally; and read on a Yellow Springs Instruments telethermometer after equilibration (approximately 45 s after insertion). The first readings (T_0) were taken before drug administration and at 20 min intervals thereafter. For α-MT pretreatment, the drug was administered twice, 18 and 1 h before the experiment; for SKF 525A pretreatment, the drug was injected 30 min before the experiment. All drugs were given intraperitoneally in volumes of 1 ml per 100 g. Because of the aqueous insolubility of bromocriptine and 25–397, these drugs and lergotrile (which is water soluble), for consistency, were dissolved in 30% ethanol. Controls received 30% ethanol solutions only.

Administration of ethanol alone produced a slight but significant hypothermia (—0·9 ± 0·4°); thus, statistical analyses were performed by comparing the mean change in core temperature of each drug-treated group with the mean of the ethanol-treated group at the same time after treatment. Absolute core temperature in rats after 1 h acclimation to the cold was 37·20 ± 0·15°.

All three compounds produced hypothermia in cold-acclimated rats, which reached maximal effects 40 min after administration for 25–397 and 120 min after administration for lergotrile and bromocriptine (Fig. 1). The onset of hypothermic response was significantly delayed in animals treated with bromocriptine or 25–397 compared with lergotrile. The observed order of potency, at doses of 2, 5, 10 and 20 mg kg^{-1}, was lergotrile ≫ bromocriptine > 25–397 (Fig. 2). The effects of 25–397, lergotrile and bromocriptine were blocked by pretreatment with haloperidol (2 mg kg^{-1}) 30 min before administration of these drugs.

α-MT Pretreatment. α-MT pretreatment itself did not change core temperature in controls administered ethanol. As Fig. 3 shows, α-MT pretreatment com-

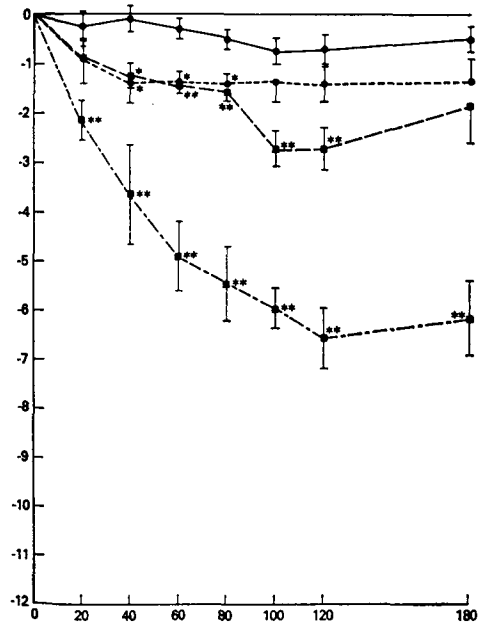

FIG. 1. Hypothermia in cold-acclimated rats administered 10 mg kg^{-1} 25–397 (●– –●), bromocriptine (■———■) or lergotrile (■———■). Controls (●—●) were given equivalent volumes of 30% ethanol. Points are means of 4 animals; vertical lines are s.e.m. Results of statistical analyses of sample means (*t*-test) are indicated on the figure. *P <0·01, **P <0·005. Ordinate: Δ in core temperature (°C). Abscissa: Time (min) after administration.

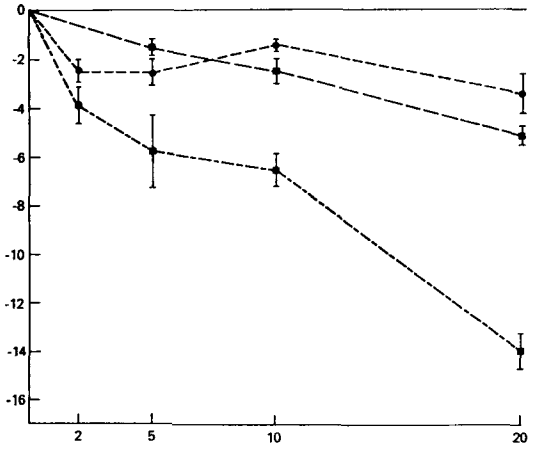

FIG. 2. Hypothermic responses in rats administered 2, 5, 10, 20 mg kg^{-1} of 25–397, bromocriptine or lergotrile. Points are means of 4 animals; vertical lines are s.e.m. 25–397 (●– –●), bromocriptine (■———■, data from Calne & others, 1975), lergotrile (■———■). Ordinate: Δ in core temperature (°C). Abscissa—Dose (μg kg^{-1}).

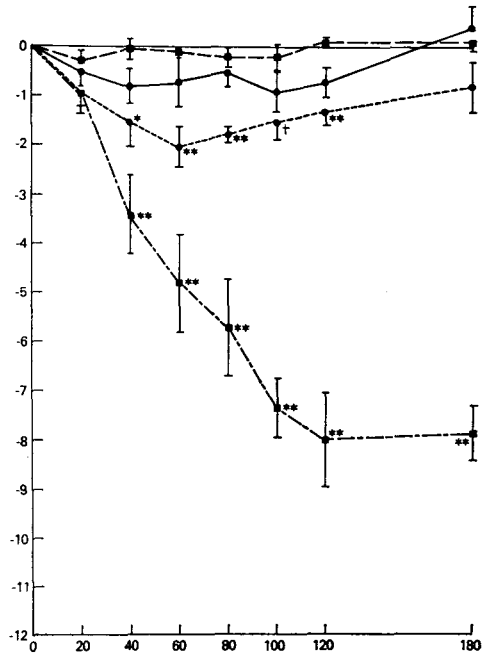

FIG. 3. Effect of α-MT pretreatment on hypothermia induced by 25–397, bromocriptine or lergotrile (designations are the same as Fig. 1). $*P < 0.05$, $+P < 0.025$, $**P < 0.005$. Ordinate: Δ in core temperature (°C). Abscissa: Time (min) after administration.

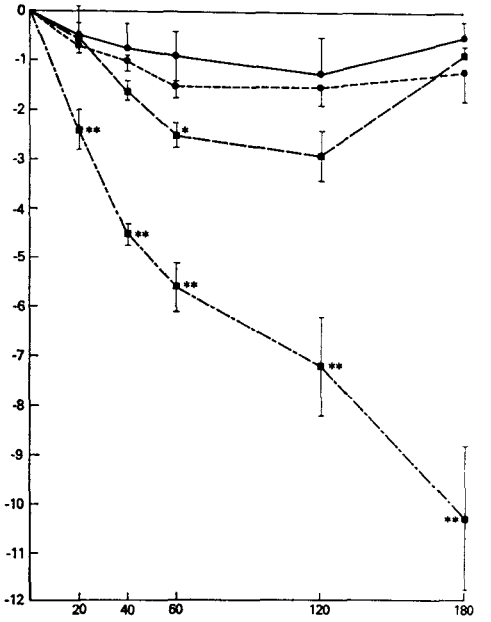

FIG. 4. Effect of SKF 525A pretreatment on hypothermia induced by 25–397, bromocriptine or lergotrile (designations are the same as Figs 1 and 3). $*P < 0.025$, $**P < 0.005$. Ordinate: Δ in core temperature (°C). Abscissa: Time (min) after administration.

pletely blocked the hypothermic effects of 10 mg kg^{-1} of bromocriptine, and slightly reduced those of 25–397. The response of α-MT-pretreated animals to lergotrile was not inhibited and may, in fact, have been somewhat potentiated (maximum temperature change without α-MT = —6·55 ± 0·6°; with α-MT, maximum temperature change = —8·0 ± 0·9°).

Pretreatment with SKF 525A. Pretreatment with 75 mg kg^{-1} SKF 525A produced significant hyperthermia, possibly as the result of reduced hepatic metabolism (in animals pretreated with SKF 525A, T_o = 33·4 ± 0·2°). After inhibition of drug metabolism, the hypothermic activity of bromocriptine and 25–397 were not significantly different from that of the vehicle in SKF 525A-pretreated rats (Fig. 4). Only at 60 min was there a significant bromocriptine-induced hypothermia. The hypothermia induced by lergotrile was significantly enhanced by SKF 525A pretreatment (maximum temperature change = —11·1 ± 0·6°).

These observations demonstrate that all 3 compounds, bromocriptine, lergotrile and 25–397, produce hypothermia in cold-acclimated rats. The results are in agreement with other studies in suggesting dopaminergic agonist activity for these agents; hypothermic action has been previously shown for bromocriptine (Calne & others, 1975; Fuxe & others, 1975); it has not been reported for lergotrile or 25–397. The ability of haloperidol, a specific dopamine receptor antagonist, to block their hypothermic effects further supports an involvement of dopaminergic mechanisms in this effect.

Interruption of presynaptic function by α-MT is reported to reduce significantly the rotation and stereotypy induced by bromocriptine and 25–397 (Fuxe & others, 1974; Jaton & others, 1976). Consistent with these observations, α-MT pretreatment blocked bromocriptine-induced hypothermia although the effects of 25–397 were not affected. Neurochemically, bromocriptine inhibits reuptake of dopamine (Silbergeld & Pfeiffer, 1977), which suggests that a component of its activity depends upon intact presynaptic function. Also, enhancement of its therapeutic action in parkinsonism is reported when the drug is administered in combination with levodopa (Kartzinel, Perlow & others, 1976).

The hypothermia resulting from bromocriptine and 25–397 after SKF 525A pretreatment was decreased. This result suggests that these compounds depend, at least in part, upon hepatic metabolism for their conversion into substances with hypothermic activity. In the case of bromocriptine, the parent compound rapidly disappears from the plasma of monkeys and of rats after intravenous injection, but its metabolites have not yet been identified (Markey & Colburn, personal communication). Since bromocriptine suppresses prolactin release from pituitary cells in tissue

culture (Pasteels, Danguy & others, 1971; Tashjian & Hoyt, 1972), the parent molecule appears to have the properties of a direct dopaminergic agonist.

The slight but significant enhancement of lergotrile-induced hypothermia by α-MT pretreatment is consistent with the results of studies on lergotrile-induced stereotypy (Pfeiffer & Silbergeld, 1976; Silbergeld & Pfeiffer, 1977). The observations suggest that lergotrile acts on other systems in addition to dopaminergic pathways (Perry & Fuller, 1976), and that such pathways play a part in the elicitation of stereotypy and hypothermia. The non-dopaminergic actions of lergotrile may be enhanced by inhibition of dopamine synthesis. The enhancement of lergotrile-induced hypothermia by SKF 525A pretreatment indicates that lergotrile may be inactivated, at least partly, by the hepatic microsomal drug metabolizing enzymes which are inhibited by SKF 525A. Lergotrile itself probably possesses dopaminergic agonist activity and it has been shown *in vitro* to release dopamine from presynaptic terminals at concentrations as low as 5×10^{-9} M (Silbergeld & Pfeiffer, 1977).

The findings reported here may have clinical implications since if an active metabolite of bromocriptine could be identified, it is possible that its administration might give an improved clinical response. Furthermore, metabolic transformation of bromocriptine raises the possibility that interactions might occur with medications which modify hepatic metabolism (by inducing increased microsomal enzyme activity, or by competitive binding to transformation pathways). Finally, these observations offer a possible explanation for the anomalous findings that 25–397 induces contralateral rotation in rats with unilateral nigral lesions (Jaton & others, 1976), a widely used experimental indicator of antiparkinsonian potency, but, in clinical trials, fails to elicit any therapeutic response when used in parkinsonism. Differences between rats and man in the metabolism of 25–397 could explain this discrepancy. Identification of the active metabolites of 25–397 in the rat might, therefore, have therapeutic implications.

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