The enhancement by benserazid of the L-dopa-induced decrease of noradrenaline in FLA63-treated animals was probably connected with the inhibition of the extracerebral decarboxylation of L-dopa. As a consequence, larger amounts of the amino-acid penetrated into the brain, leading to an enhanced formation of cerebral dopamine (Bartholini & Pletscher, 1968).

In conclusion, the present experiments indicate that L-dopa accelerates the turnover of noradrenaline *in vivo*: the endogenous amine is probably displaced by the dopamine newly formed and its loss is compensated by synthesis from the amino-acid. The displacement of endogenous noradrenaline may be of importance in enhancing central noradrenergic mechanisms, leading, for instance, to a decrease of blood pressure (Rubenson, 1971; Andén, Engel & Rubenson, 1972). In addition, the enhanced turnover of noradrenaline is possibly involved in the genesis of certain side effects of L-dopa (e.g. mental disturbances, involuntary movements) observed during the treatment of Parkinson's syndrome.

Department of Experimental Medicine,
F. Hoffmann-La Roche & Co. Ltd.,
4002 Basle, Switzerland.

H. H. KELLER
G. BARTHOLINI
A. PLETSCHER

February 25, 1974

REFERENCES

ANDÉN, N.-E., ENGEL, J. & RUBENSON, A. (1972). *Naunyn-Schmiedeberg's Arch. Pharmac.*, **273**, 1–10.

ANTON, A. & SAYRE, D. F. (1962). *J. Pharmac. exp. Ther.*, **138**, 360–375.

BARTHOLINI, G. & PLETSCHER, A. (1968). *Ibid.*, **161**, 14–20.

BARTHOLINI, G., KELLER, H. H. & PLETSCHER, A. (1973). *Neuropharmac.*, **12**, 751–756.

BERTLER, A., CARLSSON, A. & ROSENGREN, E. (1958). *Acta physiol. scand.*, **44**, 273–292.

CHALMERS, J. P., BALDESSARINI, R. J. & WURTMAN, R. J. (1971). *Proc. Nat. Acad. Sci.*, **68**, 662–666.

CONSTANTINIDIS, J., BARTHOLINI, G., TISSOT, R. & PLETSCHER, A. (1968). *Experientia (Basle)*, **24**, 130–131.

CORRODI, H., FUXE, K., HAMBERGER, B. & LJUNGDAHL, A. (1970). *Eur. J. Pharmac.*, **12**, 145–155.

LAVERTY, R. & TAYLOR, K. M. (1968). *Analyt. Biochem.*, **22**, 269–279.

MEEK, J. L. & NEFF, N. H. (1972). *Br. J. Pharmac.*, **45**, 435–441.

PERSSON, T. & WALDECK, B. (1971). *Acta pharmac. tox.*, **29**, 525–532.

ROMERO, J. A., CHALMERS, J. P., COTTMANN, K., LYTLE, L. D. & WURTMAN, R. J. (1972). *J. Pharmac. exp. Ther.*, **180**, 277–285.

RUBENSON, A. (1971). *J. Pharm. Pharmac.*, **23**, 228–230.

THOA, N. B., WEISE, V. K. & KOPIN, I. J. (1972). *Biochem. Pharmac.*, **21**, 2345–2350.

The involvement of noradrenergic systems in the locomotor activity stimulation in mice produced by β-phenethylamine

β-Phenethylamine (PE) on injection into mice produces an increase in locomotor activity (Mantegazza & Riva, 1963; Jackson, 1972). This response was shown to be biphasic (Jackson, 1972) with a first phase of increased activity occurring between 5–15 min after injection of PE 50 mg kg^{-1} (i.p.) and a second phase between 20–50 min after 100 mg kg^{-1}. The first phase was shown to be dependent on an intact dopamine and noradrenaline synthetic pathway, with the second phase apparently being produced by some metabolite of PE acting directly on dopamine receptors (Jackson, 1974). An involvement with a central cholinergic system has also been postulated, because centrally acting antimuscarinic agents potentiated the locomotor activity

produced by PE (Jackson, 1974a). The relative importance of dopaminergic and noradrenergic mechanisms both in spontaneous locomotor activity and stimulant induced activity (e.g., by amphetamine) is still in question. Most authors seem to concur with the hypothesis that both dopaminergic and noradrenergic mechanisms are involved in locomotor activity (D'Encarnacao, D'Encarnacao & Tapp, 1969; Svensson, 1970; Svensson & Waldeck, 1970; van Rossum, 1970) while Andén, Strömbom & Svensson (1973) concluded that although the dopamine receptor is of basic importance for increased motor activity, changes in noradrenaline receptor activity can quite markedly potentiate this activity. In contrast, Taylor & Snyder (1971) have suggested that locomotor activity is predominantly dependent on noradrenergic mechanisms.

Fuxe, Grobecker & Jonsson (1967) have postulated that PE may exert some of its effects by releasing extragranular amine stores in central noradrenaline and dopamine neurons. The increased locomotor activity in mice produced by PE 50 mg kg^{-1} does not disagree with this concept (Jackson, 1972, 1974b), since it was blocked by amine synthesis inhibitors such as α-methyltyrosine (α-MT) and FLA-63 (bis-(4-methyl-1-homoperazinyl thiocarbonyl) disulphide) (Spector, Sjoerdsma & Udenfriend, 1965; Corrodi, Fuxe & others, 1970). However the second phase of increased locomotor activity produced by the 100 mg kg^{-1} dose does not fit into this concept as it is inhibited only by specific dopamine receptor blocking agents. To further investigate the mechanism of action of PE on locomotor activity, groups of five mice were given various premedications of drugs known to modify synthesis and storage of noradrenaline, dopamine and 5-hydroxytryptamine (5-HT), these were: reserpine (4 mg kg^{-1}, 16–24 h premedication), FLA-63 (25·0 and 37·5 mg kg^{-1}, 2 h premedication) and α-MT (250 and 500 mg kg^{-1}, 4 h premedication) or various combinations of the three. After the appropriate premedication, the animals were then given water (as a control); clonidine (1·5 or 10 mg kg^{-1}), a noradrenaline receptor agonist (Andén, Corrodi & others, 1970); apomorphine (0·5 or 1·5 mg kg^{-1}), a dopamine receptor agonist (Ernst, 1967); or PE (100 mg kg^{-1}) and various combinations of these drugs. The animals were then immediately placed in a circular actophotometer (Jackson, 1974a) for 1 h, and activity measured as the number of times the light beams were cut between 20 and 60 min after injection. Reserpine was dissolved in a solution of 20% ascorbic acid and diluted as required. FLA-63 was suspended in a solution of Tween 80 and water, while the methyl ester HCl of α-MT was used. Control injections of the appropriate vehicle were given and all test drugs were dissolved in distilled water, which also served as a control. All drugs were administered intraperitoneally in a dose-volume of 1 ml 100 g^{-1} weight, and all animals were allowed free access to food and water up to the time of the experiment.

The results are presented in Table 1. Except where clonidine alone and PE alone were the test drugs, the increased motor activity was accompanied by stereotypies which were particularly marked with apomorphine + clonidine (all pretreatments), PE + clonidine (pretreatment reserpine + FLA-63) but less marked when the test combination was PE + clonidine (pretreatment reserpine + α-MT). The stereotypies present included a backward type of motion accompanied by licking of the forepaws sometimes so intense that the animal would roll over onto its back Stereotyped sniffing was also observed. Clonidine by itself produced no change in locomotor activity from control (water) values except when the pretreatment was reserpine + FLA-63 (37·5 mg kg^{-1}), when a significant potentiation occurred, or when the pretreatment was α-MT 250 mg kg^{-1} when a significant depression of activity occurred. Apomorphine in all cases, except where the pretreatment was α-MT 250 mg kg^{-1} caused a significant rise in locomotor activity. With clonidine and apomorphine together (each 1·5 mg kg^{-1}), a marked potentiation of the apomorphine-induced

Table 1. *The effect of pretreatment with reserpine, FLA-63 or α-MT and various combinations of these on locomotor activity produced by PE 100 mg kg^{-1}.* The PE was administered alone or with clonidine or apomorphine. In some cases apomorphine was combined with clonidine. Groups of 5 mice were used and the activity is expressed as the mean number of times the light beams were cut 20–60 min after PE injection. The figures in brackets are the number of experiments. The standard error of the mean is given. Comparisons given at the bottom of the table were made by Students *t* test.

Test drug and dose (mg kg^{-1})	Pretreatment*						
		Reserpine (4 mg kg^{-1})			α-MT		
	Alone	+ FLA-63 (25 mg kg^{-1})	+ α-MT (250 mg kg^{-1})	+ FLA-63 (37·5 mg kg^{-1})	250 mg kg^{-1}	500 mg kg^{-1}	
H_2O	458 ± 153 (16)	65 ± 42 (6)	89 ± 48 (6)		263 ± 90 (4)		a
Clonidine 1·5	815 ± 198 (16)	425 ± 79 (6)	214 ± 66 (6)		56 ± 22 (4)		b
Clonidine 10·0	416 ± 100 (8)		206 ± 98 (7)				c
Apomorphine 0·5			1377 ± 212 (6)				d
Apormorphine 1·5	1755 ± 128 (7)	1412 ± 102 (6)	1874 ± 92 (6)		391 ± 128 (4)		e
Apormorphine 1·5 + clonidine 1·5	4513 ± 320 (7)	5088 ± 488 (5)	4804 ± 283 (6)		1179 ± 239 (4)		f
PE 100	1673 ± 230 (6)	1357 ± 168 (6)	715 ± 167 (11)		1259 ± 166 (4)	1565 ± 141 (12)	g
PE 100 + Apomorphine 1·5	2188 ± 115 (6)	1522 ± 282 (5)	2220 ± 231 (7)	1252 ± 67 (4)	1486 ± 202 (4)		h
PE 100 + Clonidine 1·5	3364 ± 422 (5)	2122 ± 303 (6)	988 ± 244 (11)	2793 ± 381 (4)	2349 ± 202 (4)	2264 ± 285 (12)	i
PE 100 + Apomorphine 0·5			1508 ± 166 (9)				j
PE 100 + Clonidine 10	5686 ± 563 (6)		851 ± 232 (7)				k
	ab, ac, gh $P > 0.05$	gh, gi $P > 0.05$	gi, ab, ac, gk $P > 0.05$	gi $P < 0.01$	ae, gh, ab $P > 0.05$	gi $P < 0.05$	
	ae, ef, gk $P < 0.001$	ab $P < 0.01$	gh, ae, ef $P < 0.001$		ef $P < 0.05$		
	gi $P < 0.01$	ae, ef $P < 0.001$	gj $P < 0.01$		gi $P < 0.01$		

* Pretreatment times before test drug: Reserpine 24 h, FLA-63 2 h, α-MT 4 h.

activity was observed. This potentiation would appear to be independent of both newly synthesized noradrenaline and dopamine and reserpine sensitive amine stores since the pretreatments excluded them.

The data obtained with apomorphine and clonidine confirm those already reported by Andén & others (1973). PE (100 mg kg^{-1}) by itself produced a significant rise in locomotor activity above control values with all the pretreatments used confirming that neither intact reserpine sensitive stores of noradrenaline, dopamine or 5-HT nor an intact synthetic pathway for noradrenaline or dopamine is required for this increased locomotor activity (Jackson, 1974b). The increased locomotor activity induced by PE was potentiated by apomorphine only in those mice pretreated with reserpine + α-MT. The potentiation of locomotor activity with this particular pretreatment may be due to increased sensitivity of the dopamine receptors caused by both depletion and synthesis inhibition of dopamine.

Increased locomotor activity produced by PE was potentiated in a dose-dependent manner by clonidine in reserpine pretreated mice. Potentiation also occurred with clonidine in α-MT pretreated mice but not in those receiving reserpine + α-MT. With the latter pretreatment, even a dose of clonidine of 10 mg kg^{-1} did not potentiate PE-induced locomotor activity. These data would suggest that clonidine is able to potentiate the effect of PE when either an intact dopamine and/or noradrenaline synthetic pathway, or intact reserpine-sensitive stores, is present. However, if the reserpine-sensitive stores are depleted, and synthesis of both dopamine and noradrenaline is prevented with α-MT, the potentiation by clonidine no longer occurs. Some dopamine-β-oxidase activity is still present after vesicular store destruction by reserpine (Glowinski, Iversen & Axelrod, 1966; Wennmalm, 1968). Therefore, FLA-63 was combined with reserpine in an attempt to see whether the lack of potentiation by clonidine when pretreatment was with reserpine + α-MT was dependent on dopamine or noradrenaline. The FLA-63 + reserpine combination did not prevent clonidine

potentiation of PE, suggesting that dopamine and not noradrenaline is the critical amine. It is interesting here also to note again that clonidine by itself induced a significant increase in locomotor activity only when pretreatment was with reserpine + FLA-63, a state where one might expect an increase in newly synthesized dopamine.

Even though PE at a dose of 100 mg kg^{-1} has been shown to directly stimulate dopamine receptors 20–60 min after injection (Jackson, 1974b), this stimulation is clearly not via the same mechanism as that produced by apomorphine, since none of the pretreatments used prevented the clonidine-induced potentiation of apomorphine. It has previously been suggested that PE-induced stimulation of dopamine receptors at this time interval is due to a metabolite of PE, which is not phenylethanolamine, because dopamine-β-oxidase inhibitors do not block the locomotor activity (Jackson, 1972, 1974b). The fact that reserpine + α-MT pretreatment was required to prevent the clonidine potentiation of PE suggests that either reserpine-sensitive or newly synthesized dopamine is essential for the potentiation observed with clonidine. When both types of stores are depleted, potentiation does not occur. A presynaptic effect for clonidine, as far as I am aware, has not been reported. However, apomorphine, which is alleged to be a direct dopamine receptor stimulant (Ernst, 1967; Andén, Rubenson & others, 1967), was potentiated by clonidine in all pretreatment situations. Consequently, the lack of potentiation of PE-induced locomotor activity by clonidine after pretreatment with reserpine + α-MT is probably a consequence of the nature of the metabolite formed from PE.

I am grateful to Ms. M. Rutherford for her excellent technical assistance, to Professor R. H. Thorp for his continuing support and encouragement and to Dr. G. B. Chesher for his helpful discussions and comments.

Department of Pharmacology,
University of Sydney,
Sydney, N.S.W. 2006, Australia.

D. M. JACKSON

December 17, 1973

REEFRENCES

ANDÉN, N.-E., CORRODI, H., FUXE, K., HÖKFELT, B., HÖKFELT, T., RYDIN, C. & SVENSSON, T. (1970). *Life Sci.*, **9**, 513–523.

ANDÉN, N.-E., RUBENSON, A., FUXE, K. & HÖKFELT, T. (1967). *J. Pharm. Pharmac.*, **19**, 627–629.

ANDÉN, N.-E., STRÖMBOM, V. & SVENSSON, T. H. (1973). *Psychopharmacologia* (Berl.), **29**, 289–298.

CORRODI, H., FUXE, K., HAMBERGER, B. & LJUNGDAHL, A. (1970). *Eur. J. Pharmac.*, **12**, 145–155.

D'ENCARNACAO, P. S., D'ENCARNACAO, P. & TAPP, J. T. (1969). *Archs int. Pharmacodyn. Thér.*, **182**, 186–189.

ERNST, A. M. (1967). *Psychopharmacologia (Berl.)*, **10**, 316–323.

FUXE, K., GROBECKER, H. & JONSSON, J. (1967). *Eur. J. Pharmac.*, **2**, 202–207.

GLOWINSKI, J., IVERSEN, L. L. & AXELROD, J. (1966). *J. Pharmac. exp. Ther.*, **151**, 385–399.

JACKSON, D. M. (1972). *J. Pharm. Pharmac.*, **24**, 383–389.

JACKSON, D. M. (1974a). *Arzneimittel-Forsch.*, **24**, 24–27.

JACKSON, D. M. (1974b). *Ibid.*, in the press.

MANTEGAZZA, P. & RIVA, M. (1963). *J. Pharm. Pharmac.*, **15**, 472–478.

ROSSUM, J. M. VAN (1970). *Int. Rev. Neurobiol.*, **12**, 307–383.

SPECTOR, S., SJOERDSMA, A. & UDENFRIEND, S. (1965). *J. Pharmac. exp. Ther.*, **147**, 86–95.

SVENSSON, T. H. (1970). *Eur. J. Pharmac.*, **12**, 161–166.

SVENSSON, T. H. & WALDECK, B. (1970). *Psychopharmacologia* (Berl.), **18**, 357–365.

TAYLOR, K. M. & SNYDER, S. H. (1971). *Brain Res.*, **28**, 295–309.

WENNMALM, A. (1968). *Acta physiol. scand.*, **73**, 523–536.