This work was supported in part by U.S.A.F.O.S.R. grant F 44620-69-C-0001 to Dr. R. E. Musty, investigator. We are grateful to Dr. A. Sahagian-Edwards, Ayerst Laboratories, New York, for supplies of propranolol.

Neuropsychology Laboratory,
Department of Psychology,
University of Vermont,
Burlington, Vermont, U.S.A.

J. STEVEN RICHARDSON
P. DAVID STACEY
PETER W. CERAUSKIS
RICHARD E. MUSTY

January 25, 1971

REFERENCES

HOROVITZ, Z. P. (1966). *Recent Adv. Biol. Psychiat.*, **8**, 21–31.
LAVERTY, R., & TAYLOR, K. M. (1968). *J. Pharm. Pharmac.*, **20**, 605–609.
PELLEGRINO, L. (1968). *J. comp. physiol. Psychol.*, **65**, 483–491.
SCHALLEK, W. & KUEHN, A. (1965). *Medna. Pharmac. exp.*, **12**, 204–208.
WAAL, H. J. (1967). *Br. Med. J.*, **2**, 50.
WHEATLEY, D. (1969). *Br. J. Psychiat.*, **115**, 1411–1412.

Influences of cholinergic mechanisms on the function and turnover of brain dopamine

Anticholinergic drugs have long been used in the treatment of both spontaneous and drug-induced parkinsonism. The dopamine receptor stimulating drug apomorphine has also been reported to have a beneficial effect on Parkinson's disease, though weaker than L-dopa (Cotzias, Papavasiliou & others, 1970). Furthermore, hallucinations can be evoked both by blockade of central acetylcholine receptors and by stimulation of central catecholamine receptors. In the present investigation we have compared the effects of anticholinergics and of apomorphine on the function and turnover of dopamine in the rat corpus striatum both after and without treatment with the neuroleptic drug haloperidol.

The following drugs were used: DL-α-methyltyrosine methylester HCl (H 44/68; *Hässle, Mölndal), haloperidol (*Leo, Hälsingborg), *N*-ethyl-2-pyrrolidylmethyl-cyclopentyl-phenyl-glycolate HCl plus *N*-ethyl-3-piperidyl-cyclopentyl-phenyl-glycolate HCl (70 + 30% = Ditran; *Lakeside, Milwaukee), trihexyphenidyl HCl (*Kabi, Stockholm), (±)-hyoscyamine sulphate (atropine; Sigma, St. Louis), (−)-hyoscine hydrobromide (scopolamine; Merck, Darmstadt), *N*-methylscopolamine nitrate (Pharmacia, Uppsala), apomorphine HCl (Sandoz, Basle). The doses given refer to the salts.

The corpus striatum of adult hooded rats (150–300 g) was removed on one side by suction during diethylether anaesthesia. All brains were examined after the experiment and only animals with correct lesions were considered. The drugs tested were given 2–5 h after the operation. As previously described (Andén, Dahlström & others, 1966), haloperidol (1 mg/kg, i.p.) produced a longlasting and marked turning of the head and the tail to the unoperated side. When the haloperidol-induced asymmetry was well established after about 2 h, hyoscine (20 mg/kg, i.p.) was administered to 16 rats. It caused a clearcut and long lasting change in the position in 13 of these rats: their position became almost symmetrical in 5 min and they could turn to the operated side. A larger dose of hyoscine did not modify this response. In the remaining 3 rats, no obvious change was observed. Hyoscine (20–100 mg/kg, i.p.) given alone did not cause any observable asymmetry in unilaterally treated rats. In contrast, apomorphine (1 mg/kg, i.p.) did not change the haloperidol-induced asymmetry but evoked by itself a strong turning of the head and the tail to the operated side for about 1½ h (Andén, Rubenson & others, 1967).

The rats were examined for catalepsy immediately before death by placing one foreleg on supports of different heights and by measuring the period during which the imposed posture was maintained (Morpurgo, 1962). After treatment with haloperidol (1 mg/kg, i.p., 4¼ h) plus H 44/68 (250 mg/kg, i.p., 4 h), all the rats showed a catalepsy of the highest degree. In agreement with Morpurgo (1962), atropine (100 mg/kg, i.p., 4½ h) and hyoscine (100 mg/kg, i.p., 4½ h) completely abolished this catalepsy. Ditran (10 mg/kg, i.p., 4½ h) and trihexyphenidyl (50 mg/kg, i.p., 4½ h) partly suppressed it whereas methylscopolamine (100 mg/kg, i.p., 4½ h) and apomorphine (1·25 mg/kg, s.c. for 4¼ h) were without effect.

The dopamine and noradrenaline were in each experiment determined in the pooled brains from two male Sprague-Dawley rats, 150–250 g, by spectrofluorometry after cation exchange chromatography of the amines and their oxidation (Bertler, Carlsson & Rosengren, 1958; Carlsson & Waldeck, 1958; Carlsson & Lindqvist, 1962). The results are in Tables 1 and 2. Inhibition of the enzyme tyrosine hydroxylase by H 44/68 (250 mg/kg, i.p.) caused in 4 h a decrease in brain dopamine and noradrenaline by about 70 and 50%, respectively (*cf.* Corrodi & Hanson, 1966). Pretreatment with

Table 1. *Influence of anticholinergics and apomorphine on the disappearance of dopamine in the rat brain induced by H* 44/68 (250 mg/kg, i.p., 4 h) *alone or together with haloperidol* (1 mg/kg, i.p., 4¼ h).

	Dopamine concentration*				Difference ± s.e.†		
Drug in *C* and *D*	*A* H 44/68	*B* Haloperidol + H 44/68	*C* Drug + haloperidol + H 44/68	*D* Drug + H 44/68	*B–A*	*C–B*	*D–A*
Ditran (10 mg/kg i.p.)	31·9 (5)	20·1 (5)	26·6 (5)	39·0 (5)	−11·8 ± 1·45 ($P < 0·005$)	6·4 ± 1·37 ($P < 0·01$)	7·1 ± 1·45 ($P < 0·01$)
Trihexyphenidyl (50 mg/kg i.p.)	30·9 (5)	21·2 (5)	25·8 (5)	37·7 (5)	−9·7 ± 3·48 ($P \approx 0·05$)	4·6 ± 1·49 ($P < 0·05$)	6·8 ± 2·40 ($P < 0·05$)
Atropine (100 mg/kg i.p.)	27·6 (5)	16·6 (5)	24·2 (5)	35·5 (5)	−11·0 ± 1·74 ($P < 0·005$)	7·6 ± 1·67 ($P < 0·02$)	7·8 ± 1·90 ($P < 0·02$)
Scopolamine (100 mg/kg i.p.)	27·6 (5)	16·6 (5)	25·1 (5)	30·3 (5)	−11·0 ± 1·74 ($P < 0·005$)	8·5 ± 2·64 ($P < 0·05$)	2·7 ± 1·66 ($P > 0·05$)
Methylscopolamine (100 mg/kg i.p.)	31·7 (5)	19·8 (5)	20·9 (5)	35·8 (5)	−11·9 ± 3·99 ($P < 0·05$)	1·1 ± 0·95 ($P > 0·05$)	4·1 ± 4·38 ($P > 0·05$)
Apomorphine (0·5+0·25× 3 mg/kg s.c.)	30·1 (6)	17·1 (6)	19·0 (6)	55·4 (6)	−13·1 ± 6·45 ($P < 0·001$)	1·9 ± 0·85 ($P > 0·05$)	25·2 ± 2·18 ($P < 0·001$)

* Mean values in per cent of untreated controls (0·72 μg/g = 100%). Number of experiments in parentheses.
† Statistical significance by Student's *t*-test after pairing of samples.

Table 2. *Influence of anticholinergics and apomorphine on the disappearance of noradrenaline in the rat brain induced by H* 44/68 (250 mg/kg, i.p., 4 h) *alone or together with haloperidol* (1 mg/kg, i.p., 4¼ h).

	Noradrenaline concentration*				Difference ± s.e.†		
Drug in *C* and *D*	*A* H 44/68	*B* Haloperidol + H 44/68	*C* Drug + haloperidol + H 44/68	*D* Drug + H 44/68	*B–A*	*C–B*	*D–A*
Ditran (10 mg/kg, i.p.)	60·1 (5)	47·0 (5)	40·2 (5)	51·1 (5)	−13·0 ± 2·77 ($P < 0·01$)	−6·8 ± 2·26 ($P < 0·05$)	−8·9 ± 3·16 ($P < 0·05$)
Trihexyphenidyl (50 mg/kg, i.p.)	53·4 (5)	45·8 (5)	39·7 (5)	46·3 (5)	−7·6 ± 2·12 ($P < 0·02$)	−6·0 ± 3·26 ($P > 0·05$)	−7·1 ± 3·02 ($P > 0·05$)
Atropine (100 mg/kg, i.p.)	53·7 (5)	42·6 (5)	36·3 (5)	40·2 (5)	−11·1 ± 4·22 ($P \approx 0·05$)	−6·4 ± 5·18 ($P > 0·05$)	−13·5 ± 3·37 ($P < 0·02$)
Hyoscine (100 mg/kg, i.p.)	53·7 (5)	42·6 (5)	41·7 (5)	40·4 (5)	−11·1 ± 4·22 ($P \approx 0·05$)	−0·9 ± 3·19 ($P > 0·05$)	−13·3 ± 3·16 ($P < 0·02$)
Methylscopolamine (100 mg/kg, i.p.)	52·5 (5)	40·7 (5)	39·3 (5)	44·3 (5)	−11·7 ± 3·20 ($P \approx 0·05$)	−1·4 ± 2·74 ($P > 0·05$)	−8·3 ± 4·89 ($P > 0·05$)
Apomorphine (0·5+0·25× 3mg/kg s.c.)	49·1 (6)	42·0 (6)	45·2 (6)	52·3 (6)	−7·1 ± 0·85 ($P < 0·001$)	3·1 ± 1·97 ($P > 0·05$)	3·3 ± 5·07 ($P > 0·05$)

* Mean values in per cent of untreated controls (0·36 μg/g = 100%). Number of experiments in parentheses.
† Statistical significance by Student's *t*-test after pairing of samples.

haloperidol (1 mg/kg, i.p. 15 min before H 44/68) significantly accelerated the disappearance of brain dopamine and noradrenaline induced by H 44/68.

If one of the centrally-active anticholinergics Ditran, trihexyphenidyl, atropine or hyoscine was given 15 min before haloperidol, the haloperidol-induced acceleration of the dopamine loss was significantly reduced. On the other hand, the effect of haloperidol on the noradrenaline turnover was, if anything, potentiated by these anticholinergics. Methylscopolamine did not change the acceleration of the brain dopamine or noradrenaline turnover observed after haloperidol, which agrees with the quaternary's difficulty in entering the brain. The ineffectiveness of methylscopolamine and the opposite effects on brain dopamine and noradrenaline of the centrally-active anticholinergics favour a specific action. Apomorphine did not influence the haloperidol-induced acceleration of the dopamine or noradrenaline turnover.

The anticholinergic drugs by themselves seemed to slightly reduce the H 44/68-induced rate of disappearance of brain dopamine in contrast to the slight acceleration observed for noradrenaline. Atropine has also been reported to slightly lower the level of homovanillic acid in the mouse corpus striatum and to reduce the increase in this dopamine metabolite after neuroleptics (O'Keeffe, Sharman & Vogt, 1970). In contrast to the anticholinergics, apomorphine caused a marked deceleration of the H 44/68-induced disappearance of brain dopamine whereas that of noradrenaline was largely unaffected. Apomorphine and the anticholinergics did not by themselves change the endogenous dopamine and noradrenaline levels significantly (data not shown).

The present study provides evidence for influence on the striatal function and the turnover of the striatal dopamine both of centrally active anticholinergics and of apomorphine. The effects are not the same, however. The actions of the anticholinergics were clearly observed only after pretreatment with haloperidol. On the other hand, the clearcut changes induced by apomorphine alone were virtually completely inhibited by haloperidol. Assuming that haloperidol and apomorphine act on the dopamine receptors (see Andén, Carlsson & Häggendal, 1969), the anticholinergics probably exert an effect on the striatal function beyond the dopamine receptors.

This work was supported by the Swedish Medical Research Council (B71-40X-2457-04). P.B. is a recipient of a fellowship from the Medical Research Council of Canada. For generous supplies of drugs we thank the companies indicated by* above. Skilful technical assistance was given by Mrs. Inger Oscarsson.

Department of Pharmacology,
University of Göteborg,
Sweden.

NILS-ERIK ANDÉN
PAUL BÉDARD

January 29, 1971

REFERENCES

ANDÉN, N.-E., CARLSSON, A. & HÄGGENDAL, J. (1969). *Ann. Rev. Pharmac.*, **9**, 119–134.
ANDÉN, N.-E., DAHLSTRÖM, A., FUXE, K. & LARSSON, K. (1966). *Acta pharmac. tox.*, **24**, 263–274.
ANDÉN, N.-E., RUBENSON, A., FUXE, K. & HÖKFELT, T. (1967). *J. Pharm. Pharmac.*, **19**, 627–629.
BERTLER, Å., CARLSSON, A. & ROSENGREN, E. (1958). *Acta physiol. scand.*, **44**, 273–292.
CARLSSON, A. & LINDQVIST, M. (1962). *Ibid.*, **54**, 87–94.
CARLSSON, A. & WALDECK, B. (1958). *Ibid.*, **44**, 293–298.
CORRODI, H. & HANSON, L. C. F. (1966). *Psychopharmacologia*, **10**, 116–125.
COTZIAS, G. C., PAPAVASILIOU, P. S., FEHLING, C., KAUFMAN, B. & MENA, I. (1970). *New Engl. J. Med.*, **282**, 31–33.
O'KEEFFE, R., SHARMAN, D. F. & VOGT, M. (1970). *Br. J. Pharmac.*, **38**, 287–305.
MORPURGO, C. (1962). *Archs int. Pharmacodyn. Thér.*, **137**, 84–90.