J. Pharm. Pharmacol. 1980, 32: 204–208 Received May 21, 1979

The effects of theophylline and caffeine on thermoregulatory functions of rats at different ambient temperatures

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Both systemic and central administration of theophylline and caffeine produced a dose-dependent rise in rectal temperature at ambient temperatures of 8, 22 and 30°C. The hyperthermia in response to either xanthine was brought about by an increase in metabolic heat production. In addition, their administration produced behavioral excitation, cutaneous vasodilation (as estimated by an increase in the foot and tail skin temperatures) and diuresis. There was no change in respiratory evaporative heat loss. Probably, the hyperthermia induced by the two drugs was due to behavioral excitation leading to an increased metabolism at the ambient temperatures studied. Furthermore, either destruction of central catecholaminergic nerve fibres (with 6-hydroxydopamine) or blockade of α-adrenergic and dopaminergic (with phentolamine and haloperidol) receptors antagonized the xanthine-induced hyperthermia. The data suggest that these xanthines elicit a central activation of both adrenergic and dopaminergic receptors via release of endogenous catecholamines that leads to behavioral excitation and hyperthermia in rats.

To our knowledge, little information is available on the effect of caffeine and theophylline on thermoregulation in rats. We have, therefore, assessed their effects on metabolic, respiratory and vasomotor activities as well as body temperature responses of conscious rats to ambient temperatures of 8, 22 and 30°C.

MATERIALS AND METHODS

Adult male Sprague-Dawley rats, 200–250 g were used. Measurements were obtained from conscious animals which were trained to sit quietly under minimal restraint in rat stocks (Lin et al 1979b). Between experiments the animals were housed individually in wire-mesh cages in a room maintained at 25 ± 1·0°C with a 12-h light 12-h dark cycle. The animals were given free access to tap water and granular chicken feed.

Surgical techniques. For intraventricular injection, the ventricular cannulae were chronically implanted in the animals under general anaesthesia (sodium pentobarbitone, 6 mg/100 g, i.p.). Implantation of ventricular cannulae was according to the De Groot coordinates: AP, 7·0; Lat., 1·0; Hor., 0·1 (De Groot 1959). A 27-gauge Hamilton syringe needle was connected via PE 10 tubing to a 50 μl Hamilton syringe. During surgery the correct positioning of each guide tube was verified by the rapid flow of 0·9% NaCl or drug solutions into the lateral cerebral ventricle under gravity. The animals were not used for at least two weeks after the operation.

Drug solutions. All drug solutions were prepared in pyrogen-free glassware which was baked at 180°C for 5 h before use. The drugs injected intraperitoneally included theophylline (5–30 mg kg^{-1}); caffeine (10–20 mg kg^{-1}) (Sigma); phentolamine (1 mg kg^{-1}); haloperidol (1 mg kg^{-1}); and propranolol (5 mg kg^{-1}). A 5 μl aliquot containing either theophylline (200 μg), caffeine (100 μg) or 6-hydroxydopamine (Sigma, 100 μg) was administered into the lateral cerebral ventricle. Dosage (as salts) were prepared on the day of testing.

Measurements of thermoregulatory variables. Metabolic rate (M), respiratory evaporative heat loss (E_{res}) and vasomotor activity were measured in a small calorimeter (Lin 1978; Lin et al 1979b); M was calculated from the animal's oxygen consumption and expressed as watts(W) kg^{-1} body weight; E_{res} was calculated by measuring the increase in water vapour content in the expired air. Evaporative heat loss expressed as W was calculated from evaporative water loss (Lin 1979; Lin et al 1979b). Rectal (T_r), back skin (T_{bsk}), foot skin (T_f) and tail skin (T_t) temperatures were measured using copper-constantan thermocouples. Rectal temperature was meas-

* Correspondence.

ured with a copper-constantan thermocouple enclosed in PE 200 tubing, sealed at one end, inserted 6 cm into the rectum.

All measurements were taken once a minute throughout the experiments, each variable being measured as a d.c. potential on a Hewlett-Packard digital voltmeter (DVM 3465) interfaced to an online CPU 9825 computer. At each minute, all temperatures, M and E_{res} were calculated instantaneously by the computer and displayed.

Data collection and analysis. Animals were permitted 120 min to attain thermal balance before each drug injection. Injections were carried out within 1 min of the door-opening and almost immediately following the injections and the door-closing, the calorimeter regained its equilibrated condition. The maximal changes in T_r, T_{bsk}, T_f, T_t, M and E_{res} produced within a 120 min period after theophylline or caffeine injection were expressed as ΔT_r, ΔT_{bsk}, ΔT_f, ΔT_t, ΔM and ΔE_{res}, respectively. These data were collected at 8, 22 and 30°C.

RESULTS

Effects of theophylline and caffeine treatment on thermoregulation in rats

Control injections of equivalent vehicle into either the peritoneal cavity or the lateral cerebral ventricle produced an insignificant changes in rectal temperature or other thermoregulatory variables in the rats. However, either intraperitoneal or intraventricular administration of theophylline or caffeine produced a dose-dependent rise in rectal temperature at the three ambient temperatures (Tables 1, 2). The hyperthermia in response to either drug was brought about solely by an increase in metabolic heat production (Tables 1, 2; Fig. 1). In addition, both drugs caused behavioral excitation, cutaneous vasodilation (as estimated by an increase in the foot and tail skin temperatures) and diuresis. Probably, the dose-related hyperthermia induced (Figs. 1, 2, 3) was due to behavioral excitation leading to increased metabolism at the temperatures used. There was no changes in respiratory evaporative heat loss.

Effects of 6-hydroxydopamine (6-OHDA), phentolamine, propranolol, and haloperidol on the hyperthermia induced by theophylline and caffeine at 22°C. Control administraton of 6-OHDA (100 μg, lateral cerebral ventricle, 7 days before), phentolamine (1 mg kg^{-1}, i.p., 30 min before), propranolol (5 mg kg^{-1}, i.p., 30 min before), and haloperidol (1 mg kg^{-1}, i.p., 60 min before) did not alter the basal level of rectal temperature. However, the hyperthermia

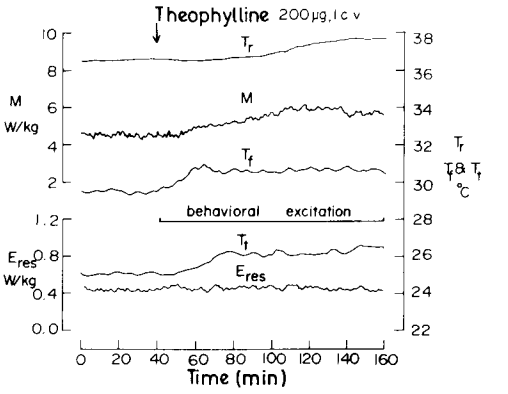

FIG. 1. The changes in thermoregulatory responses produced by an injection of 200 μg of theophylline into the lateral cerebral ventricle (i.v.c.) in a conscious rat at an ambient temperature of 22°C.

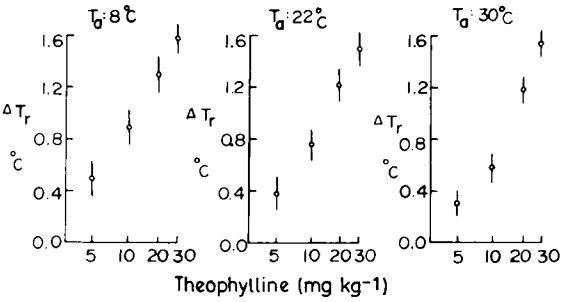

FIG. 2. Dose-response curve for theophylline (+ saline) injected into the peritoneal cavity in rats at various ambient temperatures (T_a). Each point contains five animals. The points represent the mean change in rectal temperature (T_r in degree Celsius) and the vertical bars denote ± s.e.m.

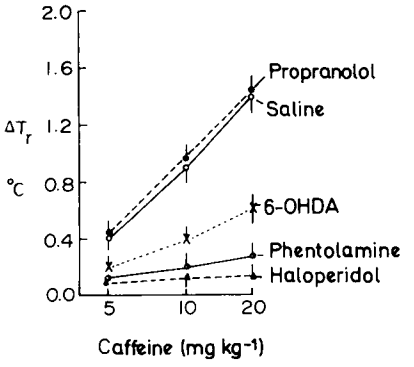

FIG. 3. Effects of 6-OHDA (100 μg, lateral cerebral ventricle, 7 days before), phentolamine (1 mg kg^{-1} i.p., 30 min before), haloperidol (1 mg kg^{-1} i.p., 60 min before), and propranolol (5 mg kg^{-1} i.p., 30 min before) on the hyperthermia induced by theophylline in rats at an ambient temperature of 22°C.

Table 1. The maximal changes in thermal responses produced by an injection of control vehicle or theophylline into the peritoneal cavity (i.p.) or lateral cerebral ventricle (l.c.v.) of conscious rats at various ambient temperatures (T_a).

Dosage and route of administration	T_a, °C	ΔT_r, °C	ΔT_{bsk}, °C	ΔT_f, °C	ΔT_t, °C	ΔE_{res}, W kg^{-1}	M, W kg^{-1}
Control vehicle, i.p.	8	−0·2±0·09	0·1±0·07	0·5±0·21	0·4±0·19	0·04±0·02	0·4±0·08
n = 4		(36·8–36·6)	(35·2–35·3)	(22·3–22·8)	(9·6–10·0)	(0·21–0·25)	(7·8–8·2)
Theophylline 20 mg kg^{-1}	8	+1·3±0·11*	1·2±0·09*	1·5±0·12*	1·2±0·13*	0·04±0·02	1·8±0·28*
i.p., n = 8		(37·0–38·3)	(35·0–36·2)	(22·6–24·1)	(9·2–10·4)	(0·19–0·23)	(7·6–9·4)
Control vehicle, l.c.v.	8	−0·1±0·06	0·2±0·07	0·6±0·25	0·4±0·23	0·05±0·03	0·3±0·09
n = 6		(36·6–36·5)	(35·3–35·5)	(22·0–22·6)	(9·4–9·8)	(0·17–0·22)	(7·7–8·0)
Theophylline 200 μg,	8	1·1±0·09*	1·2±0·08*	1·2±0·11*	1·5±0·14*	0·06±0·03	1·6±0·24*
l.c.v., n = 6		(36·8–37·9)	(34·9–36·1)	(22·4–23·6)	(9·8–11·3)	(0·18–0·24)	(7·9–9·5)
Control vehicle, i.p.	22	0·2±0·08	0·1±0·06	0·4±0·19	0·6±0·25	0·05±0·03	0·4±0·07
n = 6		(37·5–37·7)	(35·1–35·2)	(28·5–28·9)	(25·3–25·9)	(0·41–0·45)	(4·8–5·2)
Theophylline 20 mg kg^{-1},	22	1·2±0·12*	1·0±0·08*	1·1±0·10*	1·5±0·15*	0·05±0·02	2·0±0·26*
l.c.v., n = 8		(37·1–38·3)	(35·4–36·4)	(29·1–30·2)	(24·9–26·4)	(0·39–0·44)	(4·7–6·7)
Control vehicle, l.c.v.	22	−0·2±0·09	−0·1±0·08	−0·6±0·27	−0·5±0·21	−0·04±0·02	−0·5±0·10
n = 6		(37·6–37·4)	(35·6–35·5)	(28·7–28·1)	(25·3–24·8)	(0·38–0·34)	(4·9–4·4)
Theophylline 200 μg,	22	1·1±0·11*	1·0±0·10*	1·4±0·16*	1·1±0·14*	0·06±0·03	1·7±0·19*
l.c.v., n = 6		(37·4–38·5)	(35·7–36·7)	(29·3–30·7)	(25·6–26·7)	(0·37–0·43)	(4·6–6·3)
Control vehicle, i.p.	30	0·2±0·08	0·2±0·09	0·7±0·32	0·6±0·27	−0·05±0·03	−0·6±0·12
		(38·6–38·8)	(36·4–36·6)	(34·6–35·3)	(33·2–33·8)	(0·78–0·73)	(3·8–3·2)
Theophylline 20 mg kg^{-1}	30	1·2±0·10*	1·1±0·09*	1·2±0·15*	1·4±0·14*	0·04±0·02	2·1±0·22*
i.p., n = 8		(38·7–39·9)	(36·6–37·7)	(34·5–35·7)	(33·5–34·9)	(0·81–0·85)	(4·1–6·2)
Control vehicle, l.c.v.	30	−0·2±0·08	−0·2±0·09	−0·5±0·25	−0·4±0·23	0·05●0·03	−0·5±0·14
n = 6		(38·8–38·6)	(36·7–36·5)	(34·7–34·2)	(33·7–33·3)	(0·79–0·84)	(3·9–3·4)
Theophylline 200 μg,	30	1·0±0·08*	0·9±0·07*	1·0±0·12*	1·5±0·13*	0·05±0·03	1·9±0·25*
l.c.v., n = 6		(38·6–39·6)	(36·6–37·5)	(34·5–35·5)	(33·6–35·1)	(0·77–0·82)	(4·0–5·9)

* Significantly different from corresponding control value before the drug injection, $P < 0.05$ (Student's t-test). The values are expressed as the mean ± s.e.m. (range). n, numbers of animals tested.

Table 2. The maximal changes in thermal responses produced by an injection of caffeine or equivalent control vehicle into the peritoneal cavity (i.p.) or lateral cerebral ventricle (l.c.v.) of conscious rats at various ambient temperatures (T_a).

Dosage and route of administration	T_a, °C	ΔT_r, °C	ΔT_{bsk}, °C	ΔT_f, °C	ΔT_t, °C	ΔE_{res}, W kg^{-1}	ΔM, W kg^{-1}
Control vehicle, i.p.,	8	0·2±0·08	0·1±0·06	0·6±0·23	−0·4±0·19	−0·03±0·01	0·4±0·07
n = 4		(36·5–36·7)	(35·1–35·2)	(22·1–22·7)	(9·8–9·4)	(0·18–0·15)	(7·6–8·0)
Caffeine 10 mg kg^{-1} i.p.,	8	1·2±0·10*	1·1±0·12*	1·3±0·14*	1·4±0·15*	0·06±0·03	1·7±0·25*
n = 6		(36·7–37·9)	(35·2–36·3)	(21·9–23·2)	(9·9–11·3)	(0·20–0·26)	(7·9–9·6)
Control vehicle, l.c.v.	8	0·1±0·06	0·2±0·09	0·5±0·22	−0·5±·023	0·05±0·02	−0·5±0·09
n = 4		36·8–36·9)	(35·5–35·7)	(22·3–22·8)	(9·5–9·0)	(0·17–0·22)	(8·0–7·5)
Caffeine 100 μg, l.c.v.	8	1·1±0·09*	1·0±0·08*	1·2±0·15*	1·2±0·16*	0·05±0·03	1·6±0·27*
n = 6		(36·6–37·7)	(35·4–36·4)	(22·1–23·3)	(9·7–10·9)	(0·19–0·24)	(7·7–9·3)
Control vehicle, i.p.,	22	−0·2±0·09	−0·2±0·08	−0·5±0·19	0·5±0·22	0·04±0·02	0·5±0·09
n = 4		(37·6–37·4)	(35·5–35·3)	(28·7–28·2)	(25·4–25·9)	(0·39–0·43)	(4·7–5·2)
Caffeine 10 mg kg^{-1} i.p.	22	1·4±0·11*	1·2±0·09*	1·3±0·13*	1·5±0·12*	0·06±0·04	2·2±0·31*
n = 6		(37·7–39·1)	(35·4–36·6)	(28·9–30·2)	(25·6–27·1)	(0·40–0·46)	(4·6–6·8)
Control vehicle, l.c.v.	22	−0·1±0·06	−0·2±0·07	0·4±0·19	−0·5±0·21	−0·05±0·03	−0·6±0·15
n = 4		(37·5–37·4)	(35·6–35·4)	(29·0–29·4)	(26·1–25·6)	(0·39–0·34)	(4·8–4·2)
Caffeine 100 μg, l.c.v.	22	1·2±0·09*	1·0±0·08*	1·1±0·14*	1·4±0·13*	0·06±0·03	1·6±0·24*
n = 6		(37·6–38·8)	(35·4–36·4)	(28·9–30·0)	(25·9–27·3)	(0·38–0·44)	(4·9–6·5)
Control vehicle, i.p.	30	−0·3±0·11	−0·2±0·08	−0·6±0·19	−0·5±0·21	0·05±0·03	0·5±0·14
n = 4		(38·5–38·2)	(36·5–36·3)	(34·5–33·9)	(33·1–32·8)	(0·76–0·81)	(4·0–4·5)
Caffeine 10 mg kg^{-1} i.p.	30	1·1±0·09*	1·0±0·08*	1·1±0·14*	1·4±0·13*	0·06±0·03	1·6±0·24*
n = 6		(38·7–39·8)	(36·7–37·7)	(34·7–35.8)	(33·4–34·8)	(0·78–0·84)	(4·1–5·7)
Control vehicle, l.c.v.,	30	0·2±0·09	−0·2±0·07	0·4±0·19	0·6±0·25	0·06±0·02	0·5±0·13
n = 4		(38·9–39·1)	(36·4–36·2)	(34·8–35·2)	(33·7–34·3)	(0·80–·086)	(3·8–4·3)
Caffeine 100 μg, l.c.v.	30	1·0±0·08*	0·9±0·09*	1·2±0·11*	1·3±0·14*	0·05±0·02	1·4±0·27*
n = 6		(38·6–39·6)	(36·5–37·4)	(34·6–35·8)	(33·8–35·1)	(0·81–0·86)	(3·6–5·0)

* Significantly different from corresponding control value before the drug injection, $P < 0.05$ (Student's t-test). The values are expressed as the mean ± s.e.m. (range). n, numbers of animals tested.

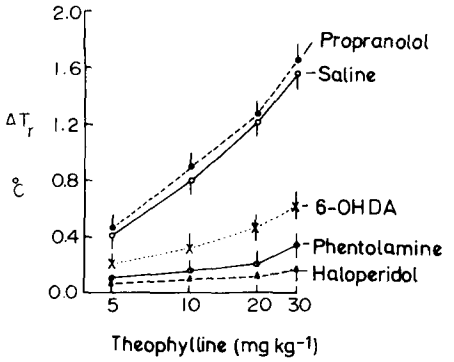

FIG. 4. Effects of 6-OHDA (100 μg. lateral cerebral ventricle, 7 days before), phentolamine (1 mg kg^{-1} i.p., 30 min before), haloperidol (1 mg kg^{-1} i.p., 60 min before), and propranolol (5 mg kg^{-1} i.p., 30 min before) on the hyperthermia induced by caffeine in rats at an ambient temperature of 22°C. Each point contains five animals. The points represent the mean change in rectal temperature (T_r in degree Celsius) and the vertical bars denote ± s.e.m.

induced by theophylline (Fig. 3) and caffeine (Fig. 4) was greatly attenuated by pretreatment of animals with 6-OHDA, phentolamine and haloperidol, but not by propranolol.

DISCUSSION

The present results show that either systemic or central administration of theophylline and caffeine produced a dose-related rise in rectal temperature of rats. The hyperthermia in response to the xanthines was due to an increased metabolic heat production. In addition, administration of the xanthines produced behavioral excitation, cutaneous vasodilation and diuresis. Furthermore, either destruction of central catecholaminergic nerve fibres (with 6-OHDA mine) or blockade of α-adrenergic and dopaminergic (with phentolamine and haloperidol) receptors was shown to antagonize the hyperthermia induced by the xanthines. The data suggest that the xanthines elicit a central activation of both adrenergic and dopaminergic receptors via release of endogenous noradrenaline and dopamine and lead to behavioral excitation and hyperthermia.

The role played by the noradrenergic pathways within brain in the regulation of body temperature in rats has not been determined. For example, intracerebral injection of noradrenaline produced dose-related increases in rectal temperature of the rats at room temperature (Myers & Yaksh 1968; Beckman 1970). In contrast, intracerebral injections of noradrenaline, or a noradrenaline depletor 6-OHDA, produced a fall in rectal temperature of the rats at the same ambient temperatures. (Simmonds & Uretsky 1970; Nakamura & Thoenen 1971; Avery 1972; Breese et al 1972). Moreover, Feldberg & Lotti (1967) found that the action of intracerebral noradrenaline may be hyperthermic at a small dose or hypothermic at a larger doses. Furthermore, administration of either dopamine or apomorphine produces a fall in rectal temperature of rats at room temperature (22°C) and below it (Bruinvels 1970; Kruk 1972; Lin et al 1979a). In the cold, the hypothermic effects in response to apomorphine (a dopaminergic agonist) was due to a decrease in heat production, while at room temperature the hypothermia was due to an increase in cutaneous circulation and a decrease in heat production (Lin et al 1979a). Thus, it appears that central catecholaminergic pathways, concerned with temperature regulation, may not be responsible for the development of hyperthermia induced by the xanthines.

There is a growing evidence for the involvement of central catecholamines in affective disorders. According to a catecholamine hypothesis of affective disorders, behavioral depression may be related to a deficiency of catecholamines at central catecholaminergic receptor sites, while mania or behavioral excitation results from excess function of central catecholamines (Snyder 1972; Schildkraut 1973; Cooper et al 1978). In the present results, administration of the xanthines produced hyperthermia, behavioral excitation and cutaneous vasodilation and hyper-metabolism in rats. Probably, activation of central catecholaminergic receptors with the xanthines may have resulted in behavioral excitation and thus caused increases in metabolic heat production (or increased motor activity) which leads to hyperthermia. With the destruction of central catecholaminergic pathways or the blockade of central catecholaminergic receptors, treated rats showed few alterations in both behavioral and thermoregulatory responses in response to the administration of the xanthines.

It thus seems reasonable to view the hyperthermia induced by the xanthines as resulting from this drug-induced increase in activity rather than from a direct action on the central thermoregulatory structures.

Two types of effects of the methylxanthines have received major attention in studies of their mechanism of action: those mediated by cyclic nucleotides and those associated with intracellular translocations of calcium (Sattin & Rall 1970; Blinks et al 1972). According to Laburn et al (1974), dibutyryl cAMP,

when injected into the anterior hypothalamus, produced a dose-dependent rise in rectal temperature of rabbits. Also, addition of theophylline to prostaglandin E_1 (a pyrogenic substance) injectate into one side of the anterior hypothalamus resulted in a greater fever than that induced by the same dose of prostaglandin E_1 alone into the control side. In addition, when the ventricular system of animals was perfused with a low dose of calcium solution there was a rise in rectal temperature (Feldberg et al 1970; Myers & Brophy 1972). In the same experiments, a high dose of calcium solution produced a fall in rectal temperature. These observations tend to indicate that the xanthines may act on the AMP metabolism and intracellular translocations of calcium to induce hyperthermia.

Acknowledgements

This work was supported by the National Science Council (Republic of China) and J. Aron Charitable Foundation (N. Y., U.S.A.). The authors are grateful to Dr C. Y. Chai for his advice and support, to Dr C. J. Shih and Mr C. C. Wei for their generous support.

REFERENCES

Avery, D. D. (1972) J. Physiol. (London) 220: 257–266

Beckman, A. L. (1970) Am. J. Physiol. 218: 1596–1604

Blinks, J. R., Olson, C. B., Jewell, B. R., Braveny, P. (1972) Circulation Res. 30: 367–392

Breese, G. R., Moore, R. A., Howard, J. L. (1972) J. Pharmacol. Exp. Ther. 180: 591–602

Bruinvels, J. (1970) Neuropharmacology 9: 277–282

Cooper, R. J., Bloom, F. E., Roth, R. H. (1978) The Biochemical Basis of Neuropharmacology, 3rd ed. Oxford University Press, New York

De Groot, J. (1959) J. Comp. Neurol. 113: 389–406

Feldberg, W., Lotti, V. J. (1967) Br. J. Pharmacol. 31: 152–161

Feldberg, W., Myers, R. D., Veale, W. L. (1970). J. Physiol. (London) 207: 403–416

Kruk, Z. L. (1972) Life Sci. 11: 845–850

Laburn, H. P., Rosendorff, C., Willies, G., Woolf, C. (1974) J. Physiol. (London) 240: 49–50 p

Lin, M. T. (1978) J. Pharmacol. Exp. Ther. 204: 39–45

Lin, M. T. (1979) Can. J. Physiol. Pharmacol. 57: 16–23

Lin, M. T., Chern, Y. F., Chern, S. I. (1978) Experientia 34: 1595–1596

Lin, M. T., Chern, Z. F., Wang, Z., Wang, H. S. (1979a) Can. J. Physiol. Pharmacol. 57: 469–475

Lin, M. T., Chern, Y. F., Liu, G. G., Chang, T. C. (1979b) Proc. Nat. Sci. Counc., R.O.C. 3, 46–52

Myers, R. D., Brophy, P. D. (1972) Neuropharmacology 11: 351–361

Myers, R. D., Yaksh, T. L. (1968) Physiol. Behav. 3: 917–928

Nakamura, K., Thoenen, H. (1971) Eur. J. Pharmacol, 16: 46–54

Sattin, A., Rall, T. W. (1970) Molec. Pharmacol. 6: 13–23

Schildkraut, J. J. (1973) Annu. Rev. Pharmacol. 13: 427–489

Simmonds, M. A., Uretsky, N. J. (1970) Br. J. Pharmacol. 40: 630–638

Snyder, S. H. (1972) Arch. Gen. Psychiatry 27: 169–177

Wilson, D. F. (1973) Am. J. Physiol. 225: 862–865