Br. J. Pharmac. (1974), 51, 497-501

EFFECTS OF CATECHOLAMINES ON THERMOREGULATION IN PIGEONS

NARESH CHAWLA, M.B.L. JOHRI, P.N. SAXENA & K.C. SINGHAL
Department of Pharmacology, J.N. Medical College, A.M.U., Aligarh, India

1 In unanaesthetized pigeons, kept at room temperature (20-23°C) the effects on cloacal temperature were examined of catecholamines, phenoxybenzamine and propranolol, injected into the cerebral ventricles.

2 Noradrenaline, adrenaline, dopamine and isoprenaline caused a fall in cloacal temperature.

3 Phenoxybenzamine produced a long-lasting small rise in cloacal temperature. This rise is attributed to removal of the hypothermic effect of noradrenaline released continuously from adrenergic neurones ending in the anterior hypothalamus. Propranolol produced a slight fall in cloacal temperature.

4 The hypothermic effects of noradrenaline, adrenaline and dopamine were prevented by phenoxybenzamine but not by propranolol. They are therefore attributed to activation of α-adrenoceptors.

5 The hypothermic effect of isoprenaline was not prevented by either phenoxybenzamine or propranolol. The effect can therefore not be attributed to activation of either α or β-adrenoceptors. Propranolol actually accentuated the isoprenaline-induced hypothermia.

Introduction

The nature of chemical mediation of thermoregulation in homoeothermic animals is species dependent. It is now well known (Feldberg, 1968) that the catecholamines, noradrenaline and adrenaline, acting on the thermostat located in the anterior hypothalamus, produce hypothermia in the cat, dog and monkey, hyperthermia in the rabbit and sheep, a biphasic change in body temperature in the rat and mouse and have no effect in the goat and the ox. It has been further suggested that the hypothermic effect in the cat as well as the hyperthermic effect in the rabbit and the rat is mediated through α-adrenoceptor activation (Banerjee, Feldberg & Lotti, 1968; Feldberg & Saxena, 1971; Dhawan & Dua, 1971; Saxena, 1973).

Thermoregulatory responses to the catecholamines in birds, a class of homoeothermic animals different from the mammals, have not been as extensively studied. As suggested by Feldberg (1968), difficulties in implantation of intraventricular or intrahypothalamic cannulae in their small sized brains have mainly been responsible for this lack of study. He suggested that in birds, too, the catecholamines lower body temperature by acting on the hypothalamus. This conclusion is based on the work of Allen & Marley (1967) in which the catecholamines were injected intravenously in 1-28 days old chicks before the blood brain barrier to the amines developed. The investigators suggested that the hypothermic effect of the catecholamines was mediated through α-adrenoceptor activation since the effect was prevented by pretreatment with phenoxybenzamine.

Later, Marley & Stephenson (1970) confirmed the hypothermic effect of catecholamines by microinfusing them into the hypothalamic region of the young chicks. They attributed the hypothermia to β-adrenoceptor activation. The latter finding does not fall in line with the foregoing suggestion that change in either direction in body temperature brought about by the catecholamines in different species of the mammals is mediated through α-adrenoceptor activation. It was, therefore, decided to investigate the mechanism of adrenergic mediation of thermoregulatory response in the domestic pigeon, using the intraventricular route for administration of the amines.

Methods

Healthy pigeons of either sex and weighing between 200-300 g were used. Food and water

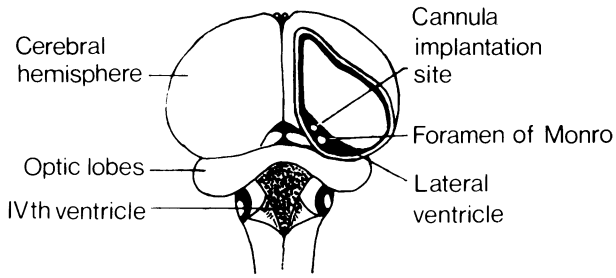

Fig. 1 Diagrammatic illustration of the dorsal side of the pigeon's brain. The layer of the right cerebral hemisphere overlying the lateral ventricle was removed to expose its cavity. The site of the tip of the intraventricular cannula inside the cavity is indicated.

were withheld during the period of experiment. The room temperature was maintained between 20-23°C.

Pentobarbitone (30 mg/kg) was given intravenously to produce surgical anaesthesia. The operations were performed aseptically. A stereotaxic instrument designed for rats was used, and the head, cleaned of its feathers, was fixed by means of the ear bars and beak bar with the distance between the ear and beak bars adjusted to 16 mm. Stereotaxic coordinates for placement of the cannula in the lateral cerebral ventricle had first been determined in six birds.

The skin over the head was incised in the midline and the underlying skull bone cleaned. A 1.0 mm hole was made in the skull at a point 1.5 mm right to the midline and 4 mm posterior to the interaural plane with a dental burr by slow manual rotation. The dura was punctured by a cutting needle.

The intraventricular cannula consisted of a 9.0 mm long shaft of a No. 20 hypodermic needle with a 2 mm diameter thin aluminium disc soldered 3 mm from one end. The cannula, held by its longer end in an electrode holder, was directed anteriorly 5° in the vertical plane and lowered 3 mm through the hole. The aluminium disc which rested on the surface of the skull was fixed to the bone with acrylic cement. When the cement had hardened, the electrode holder was removed. The cannula was flushed by injecting 0.02 ml of 0.9% w/v NaCl solution through it. A stainless steel wire of appropriate length and thickness was left inside the cannula to serve as a stilette. Penicillin powder was dusted over the wound and the skin sutured. The pigeon was returned to its cage and allowed two days to recover. A cannulated pigeon was used on alternate days for temperature studies.

For recording the temperature, a thermistor probe was inserted about 2 cm deep into the cloaca. The protruding end of the probe was fixed in the centre of a 30 cm long adhesive tape and the two ends of the tape crossed over the back, brought forward over the chest and stuck over each other. This held the probe firmly in place. The cloacal temperature was taken from a telethermometer usually at 6 or 15 min intervals. The temperature records of Figs. 2-5 were plotted from these readings.

For the intraventricular injections 12 mm of the shaft of a No. 24 hypodermic needle was used, connected by polythene tubing, 20 cm long, to a tuberculin syringe, with a rubber disc mounted over the terminal 2 mm of the shaft, at the junction with the tubing. By gentle pressure of the disc against the cannula during an injection, backflow of the fluid was prevented. Before the injection was made syringe, tubing and needle were filled with the solution to be injected, the pigeon was taken out of its cage, the stilette removed from the cannula, and the needle inserted through its entire length. The drugs, in whatever dose tested, were always injected in a volume of 0.02 ml and the injections were made slowly whilst the rubber disc was pressed gently against the cannula. After injection, the needle was withdrawn, the stilette replaced, and the pigeon returned to its cage.

The placement of the cannula was verified macroscopically in every bird at the end of a series of experiments by injection of 0.02 ml of a 1% solution of bromophenol blue, dissolved in distilled water, through the cannula; staining of the ventricular walls was observed with the naked eye after the bird had been killed by an overdose of pentobarbitone sodium and the ventricles opened. The correct site of placement of the cannula is shown diagrammatically in Figure 1.

The following drugs were used: (−)-noradrenaline bitartrate, (−)-adrenaline bitartrate, dopamine hydrochloride, (±)-isoprenaline

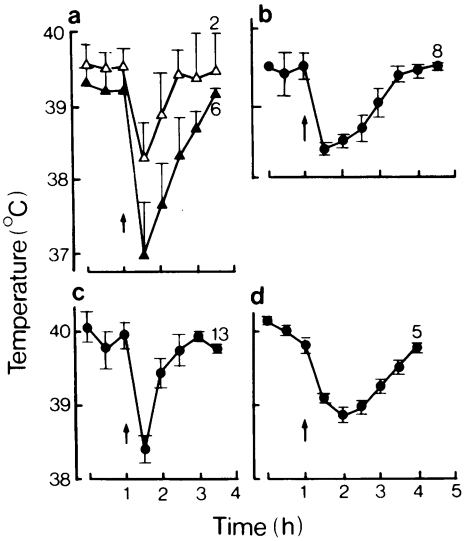

Fig. 2 Records of mean cloacal temperature of unanaesthetized pigeons. The figures indicate the number of observations and the vertical bars are the standard errors of the means (in record (a) shown only one one side). The arrows indicate time of intraventricular injection of the doses given of (a) noradrenaline (△, 20 μg; ▲, 30 μg); (b) adrenaline (20 μg); (c) dopamine (60 μg); or (d) isoprenaline (60 μg).

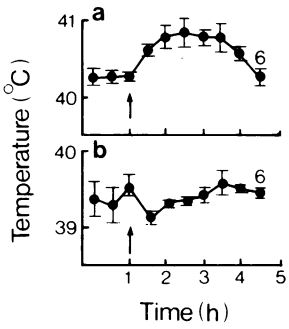

Fig. 3 Records of mean cloacal temperatures of unanaesthetized pigeons. The figures in brackets indicate the number of observations and the vertical bars are the standard errors of the means. The arrows indicate time of injection of 40 μg of (a) phenoxybenzamine or (b) propranolol.

sulphate, phenoxybenzamine hydrochloride and (±)-propranolol hydrochloride. All doses refer to the salts. The drugs were dissolved in pyrogen-free distilled water.

Results

Apart from a transient rise in cloacal temperature which occurred in some pigeons as a result of excitement due to handling, cloacal temperature was not affected by control injections of 0.02 ml of distilled water into the cerebral ventricles nor by intravenous injections of the drugs in the doses used for intraventricular injections. But all four catecholamines, adrenaline, noradrenaline, dopamine and isoprenaline, lowered temperature on intraventricular injection. They also produced sedation. Often the birds went to sleep standing with their wings applied closely to the trunk.

The hypothermic effects of the four catecholamines are illustrated in Figure 2. Noradrenaline and adrenaline were more potent, being effective in smaller doses, than dopamine and isoprenaline. With noradrenaline, adrenaline and dopamine temperature began to fall about 6 min after the injection and the maximal fall was reached about 30 min later, whereas with isoprenaline temperature began to fall about 12 min after the injection and the maximal fall was reached about 60 min later. The hypothermic effect of adrenaline and isoprenaline lasted longer than that of noradrenaline and dopamine.

Phenoxybenzamine did not produce any changes in the behaviour of the pigeons but it produced a long-lasting small rise in cloacal temperature, as illustrated in Figure 3a. In addition, the phenoxybenzamine prevented the pronounced hypothermia produced by noradrenaline, adrenaline and dopamine. The effect on cloacal temperature of the three catecholamines, when injected after phenoxybenzamine, consisted solely in abolition of the small prolonged rise produced by the phenoxybenzamine. On the other hand, the pronounced hypothermic effect of isoprenaline was not prevented by phenoxybenzamine. These results are illustrated in Figure 4.

Propranolol did not affect the behaviour of the pigeons but it produced a slight fall in cloacal temperature. This is illustrated in Fig. 3b and the records of Fig. 5 illustrate that it did not prevent the hypothermias produced by noradrenaline, adrenaline and dopamine. The hypothermia produced by isoprenaline was in fact accentuated by propranolol. This is evident when the effects of isoprenaline (60 μg) shown in record d of Figs. 2 and 5, are compared.

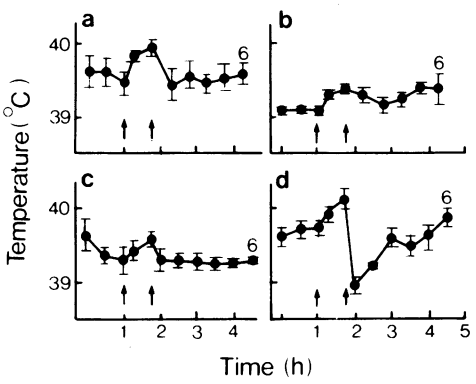

Fig. 4 Records of the mean cloacal temperatures of unanaesthetized pigeons. The figures indicate the number of observations and the vertical bars the standard errors of the means. In each record phenoxybenzamine (40 μg) was injected intraventricularly at the time shown by the first arrow. Forty-five minutes later, either 20 μg of (a) noradrenaline or (b) adrenaline; or 60 μg of (c) dopamine, or (d) isoprenaline was injected intraventricularly as shown by the second arrow.

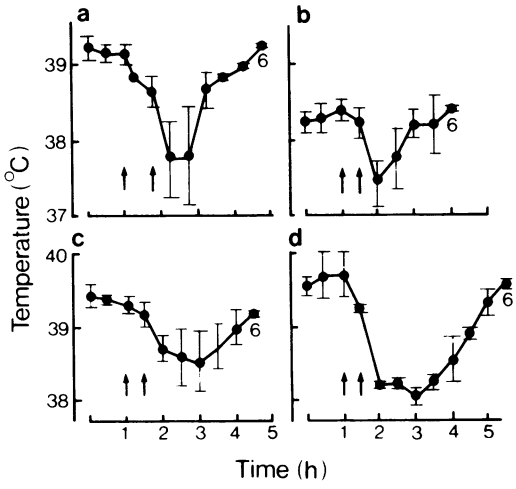

Fig. 5 Records of the mean cloacal temperatures of unanaesthetized pigeons. The figures in brackets indicate the number of observations and the vertical bars standard errors of the means. In each record the first arrow shows the time when propranolol (40 μg) was injected intraventricularly. Forty five minutes later (a) 20 μg of noradrenaline or 30 min later (b) 20 μg of adrenaline, or (c) 60 μg of dopamine, or (d) 60 μg of isoprenaline was injected intraventricularly as shown by the second arrow.

Discussion

The hypothermias produced by injection of noradrenaline, adrenaline, dopamine and isoprenaline into the cerebral ventricles of pigeons confirm the observations made by Allen & Marley (1967) that in chicken the catecholamines lower temperature by acting on the central nervous system. The site of action is the hypothalamus because Marley & Stephenson (1970) obtained hypothermia on microinfusion of the catecholamines into the hypothalamus of chicken. It would thus appear that the catecholamines have the same effect on temperature in birds as in cats, dogs and monkeys, when acting on the hypothalamus.

Dopamine was found to be less potent than noradrenaline or adrenaline in lowering temperature in the pigeon. This finding is consistent with previous observations in cats and rabbits. In cats dopamine was found to be weaker than noradrenaline or adrenaline in abolishing tremor or shivering (Domar & Feldberg, 1960) and in lowering rectal temperature (Saxena, 1973); in rabbits dopamine was found to be weaker in raising rectal temperature (Dhawan & Dua, 1971). The comparatively weak hypothermic action of dopamine in pigeons and cats can be explained on the assumption that it acts as a precursor of noradrenaline and consequently causes an increase in the concentration of the neurohumour. This idea is supported by an observation of Marley & Stephenson (1970) who found that the hypothermic effect of dopamine in the chicken could be brought to light by prior administration of a monoamine oxidase inhibitor. Since the onset of the effect was as rapid as with noradrenaline or adrenaline the hypothermic effect could be attributed, at least in part, to dopamine itself.

The small but long-lasting rise in temperature which was obtained with phenoxybenzamine can be attributed to removal of the hypothermic effect of continuously released noradrenaline from adrenergic fibres ending in the anterior hypothalamus. A rise in temperature was also observed in cats on injection of phenoxybenzamine into the cerebral ventricles, but the effect was not obtained consistently (Feldberg & Saxena, 1971). This suggests that a tonic influence of continuously released noradrenaline plays a greater part in maintaining body temperature in birds than in cats. In rabbits, the role of continuously released noradrenaline on maintenance of body temperature would be more comparable to that in birds because phenoxybenzamine regularly affected temperature on intraventricular injection. But in this species, in which noradrenaline raises temperature when acting on the hypothalamus, phenoxybenzamine lowered temperature.

The hypothermic effects of noradrenaline, adrenaline and dopamine appear to be mediated through activation of α-adrenoceptors in the hypothalamus because they were blocked by phenoxybenzamine but not by propranolol. The same conclusion was reached by Allen & Marley (1967) for the hypothermic effect of these catecholamines in chickens. Activation of α-adrenoceptors has also been considered responsible for the hypothermic effects of noradrenaline and adrenaline in cats and for their hyperthermic effects in rabbits and rats (Banerjee *et al.*, 1968; Feldberg & Saxena, 1971; Dhawan & Dua, 1971; Saxena, 1973).

Isoprenaline is generally regarded as a potent β-adrenoceptor activating agent. Its hypothermic effect in the pigeon was not abolished by phenoxybenzamine. If it were abolished by propranolol it would suggest that β-adrenoceptor activation may contribute to the hypothermic effects of the catecholamines. Indeed, Marley & Stephenson (1970) are of the opinion that the hypothermia caused by the catecholamines in the chickens is mediated through activation of β-receptor or a catecholamine receptor with α and β characteristics as it was blocked by both phenoxybenzamine and propranolol microinfused into the hypothalamus. Intravenous injection of phenoxybenzamine but not of propranolol also blocked the hypothermia, suggesting the existence of a blood-brain barrier for propranolol. They inferred that the blocking effect of propranolol is specific while that of phenoxybenzamine non-specific as the latter possesses a wide spectrum of activity against many types of receptors.

However, the hypothermic effect of isoprenaline in the pigeon was not abolished by the intraventricular administration of propranolol but was actually enhanced. A similar enhancement has been observed by Marley & Stephenson (1970) when propranolol was given intravenously. In the present experiments, diffusion of propranolol into the nervous structures concerned with thermoregulation must have occurred because of its hypothermic effect. It is therefore difficult to explain the disparity of the present results with the observation made by Marley & Stephenson. It seems relevant to mention in this connection the finding that isoprenaline did not affect rectal temperature in cats on microinfusion into the anterior hypothalamus while noradrenaline and adrenaline similarly applied lowered the temperature (Saxena, 1973).

Phenoxybenzamine hydrochloride was kindly supplied by Smith, Kline & French Laboratories, Bangalore, India, and isoprenaline sulphate by Burroughs Wellcome & Co., India.

References

ALLEN, D.J. & MARLEY, E. (1967). Effect of sympathomimetic and allied amines on temperature and oxygen consumption in chickens. *Br. J. Pharmac. Chemother.*, **31**, 290-312.

BANERJEE, U., FELDBERG, W. & LOTTI, V.J. (1968). Effect on body temperature of morphine and ergotamine injected into the cerebral ventricles of cats. *Br. J. Pharmac. Chemother.*, **32**, 523-538.

DHAWAN, B.N. & DUA, P.R. (1971). Evidence for the presence of α-adrenoceptors in the central thermoregulatory mechanisms of rabbits. *Br. J. Pharmac.*, **43**, 497-503.

DOMER, F.R. & FELDBERG, W. (1960). Tremor in cats: the effect of administration of drugs into the cerebral ventricles. *Br. J. Pharmac. Chemother.*, **15**, 578-587.

FELDBERG, W. (1968). The monoamines of the hypothalamus as mediators of temperature responses. In: *Recent Advances in Pharmacology*, ed. Robson, J.M. and Stacey, R.S. 4th edition. London: J.A. Churchill Ltd.

FELDBERG, W. & SAXENA, P.N. (1971). Effects of adrenoceptor blocking agents on body temperature. *Br. J. Pharmac.*, **43**, 543-554.

MARLEY, E. & STEPHENSON, J.D. (1970). Effects of catecholamines infused into the brains of young chickens. *Br. J. Pharmac.*, **40**, 639-658.

SAXENA, P.N. (1973). Mechanism of hypothermic action of catecholamines in the cat. *Ind. J. Pharmac.*, **5**, 374-377.

(Received October 27, 1973)