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Effects of clonidine and xylazine on body temperature in the rat

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1 Clonidine and xylazine injected intraperitoneally both produced a dose-dependent hypothermia in unanaesthetized, freely moving rats.

2 The α_2-antagonist yohimbine produced an antagonism of the xylazine-induced hypothermia but a potentiation of the clonidine-induced hypothermia.

3 The opioid antagonist naloxone had little effect on the xylazine hypothermia, but potentiated the clonidine hypothermia.

4 It is concluded that whilst the two α_2-agonists produce a similar hypothermic effect in the rat, they are acting by different mechanisms.

Introduction

There have been many studies on the role of noradrenaline in the control of body temperature in the rat. Since the studies of Keller & Hare in 1932, the majority of these investigations have centred on the hypothalamus, but a wide range of responses has been recorded. Using intracerebroventricular injections, Feldberg & Lotti (1967) saw either hyper- or hypothermia depending on the dose of noradrenaline used. Avery (1972) observed a variety of responses following intrahypothalamic injections depending on the ambient temperature, and Poole & Stephenson (1977) showed a biphasic response to intrahypothalamic injections of noradrenaline.

There have also been many studies on the role of 5-hydroxytryptamine (5-HT) in the control of body temperature (Cox & Lee, 1979; Cox, Kerwin, Lee & Pycock, 1980) and evidence exists for a relationship between 5-HT neurones and both noradrenergic and dopaminergic neurones in other areas of the CNS (Baraban, Wang & Aghajanian, 1978; Cox *et al.*, 1980).

Drugs which act as agonists at the noradrenergic binding site were originally considered to exert their effect by inhibiting noradrenaline release presynaptically (Langer 1974); however, evidence has accumulated that other neurotransmitters, such as acetylcholine, may have their release inhibited by α_2-agonists (Drew 1978) and that postsynaptic α_2-receptors may play an important part in the effects of α_2-active drugs (Langer & Shepperson, 1982; McGrath, 1983).

Since the role of noradrenaline in the central control of body temperature has been widely documented, the present study was done to examine the effects of α_2-agonists on body temperature in unrestrained rats in the hope that it might provide further information on this complex issue, as the presynaptic effects should be to reduce noradrenaline release. Two drugs were examined, clonidine, which although it is not regarded as a particularly selective agonist, has been extensively investigated in a variety of systems and is active at very low doses (Hoefke, 1980; Ishii, Yamamoto & Kato, 1982); and xylazine, which although not as potent nor as widely documented, is thought to be more selective in its actions on the α_2-binding site (Hsu, 1981). In addition the effects were measured of two α_2-antagonists, yohimbine and RS21361 (Michel & Whiting, 1981) on the responses to the α_2-agonists. Since it has been reported that morphine hypothermia can be antagonized by the α_2-antagonist yohimbine, (Lawrence & Livingston, 1981), the effect of the opiate antagonist naloxone on the responses to clonidine and xylazine was also measured to see if any antagonism could be demonstrated.

Methods

Adult male Wistar rats, body weight 300–400 g were used, and body temperature was measured using a rectal thermistor probe inserted 5 cm.

Ambient temperature was between 21 and 24 °C and since restraint and ambient temperature have been shown to produce significant effects on body temperature (Thornhill, Cooper & Veale, 1980), the animals were unrestrained and left in the test environment for 1 h to adapt before any readings were taken. Drugs were made up in sterile 0.15 M saline and injected intraperitoneally. The following drugs were used: clonidine hydrochloride, kindly supplied by Boehringer Ingelheim Ltd; xylazine hydrochloride, kindly supplied by Bayer Ltd; naloxone hydrochloride kindly supplied by Endo Labs Ltd; RS21361 2-(1-ethyl-2-imidazolyl methyl)-1,4-benzodioxan kindly supplied by Syntex Pharmaceutics Ltd and yohimbine hydrochloride (Sigma). Control animals were injected with 0.2 ml sterile 0.15 M saline. For the effects of drugs on their own, two control readings were made for each animal before injection and then readings at 20 min intervals for 2 h were taken. Where antagonists were used these were injected 10 min before the drug under investigation and readings taken at 40 min intervals thereafter.

Clonidine was injected at doses ranging from 0.2 to 2.0 mg kg^{-1}, xylazine at doses ranging from 5 to 30 mg kg^{-1}, naloxone at 5 mg kg^{-1}, RS21361 at 10 mg kg^{-1} and yohimbine at 2 mg kg^{-1}.

Results are expressed in terms of the change in body temperature from the preinjection control value and the mean values from groups of six animals obtained, and compared using Student's *t* test.

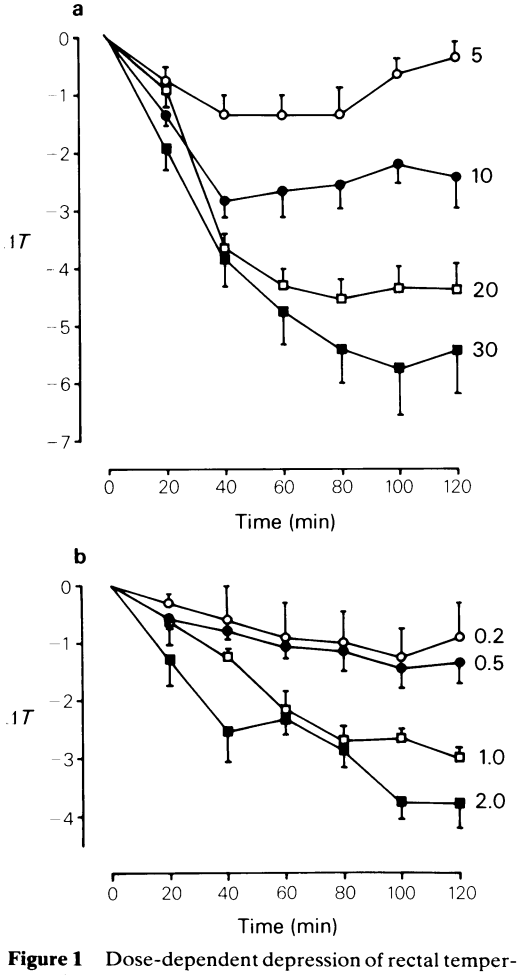

Figure 1 Dose-dependent depression of rectal temperature in groups of six rats treated with xylazine (a) and clonidine (b). Mean values are shown with vertical lines indicating s.e. mean. Numbers against each curve show dose in mg kg^{-1}.

Results

The control experiments measuring change in body temperature following injections of saline, yohimbine 2 mg kg^{-1}, naloxone 5 mg kg^{-1} or RS21361 10 mg kg^{-1} showed no significant effects over the 2 h examination period.

Injections of 0.2, 0.5, 0.7, 1.0, 1.5 and 2.0 mg kg^{-1} of clonidine and 5, 10, 15, 20, 25 and 30 mg kg^{-1} of xylazine were followed by a dose-dependent fall in body temperature as shown in Figure 1. Following the lower doses, the effects were not only smaller in amplitude, but also of shorter duration. The magnitude of effect however was greater with xylazine than clonidine over the dose-range measured. There were clear side effects including sedation, diuresis, exophthalmos and piloerection at all dose levels and these were more marked at the higher levels for both drugs.

The effect of the α_2-antagonist yohimbine was as expected on the α_2-agonist xylazine, showing some inhibition of the hypothermia produced at all dose levels at 40, 80 and 120 min. Figure 2 shows the effect at 80 min when a significant inhibition ($P<0.05$) could be seen at all dose levels except at 5 and 15 mg kg^{-1}. The antagonist RS21361 was only measured at three dose levels of xylazine, but it can be seen that the effects were very similar to those of yohimbine with the effect at the 30 mg kg^{-1} dose being significant ($P<0.05$). The effects of yohimbine on the clonidine hypothermia, however, were not the same as those seen on xylazine (Figure 2). In this case there was no antagonism of the hypothermia, on the contrary, in all cases there was a potentiation of the effect. Figure 2 shows the values obtained at 80 min and the values for 0.5, 0.7, 1.5 and 2.0 mg kg^{-1} clonidine are significantly potentiated ($P<0.05$). Similarly the antagonist RS213661 produced no antagonism of clonidine hypothermia.

The opioid antagonist naloxone produced equally

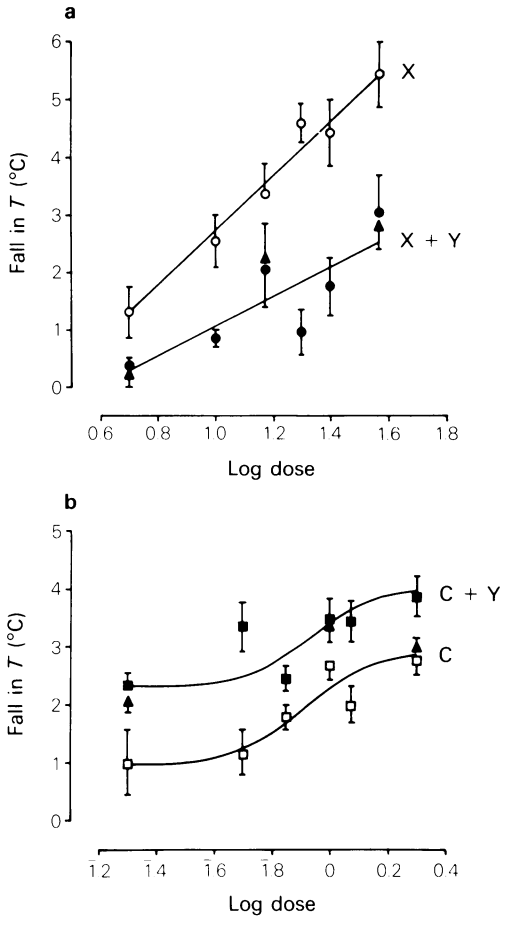

Figure 2 Log dose-response curves to show effect of pretreatment with yohimibine (y) 2 mg kg^{-1} (●,■), on the depression of rectal temperature produced by xylazine (X) in (a) and clonidine (C) (○,□) in (b), 80 min after injection in groups of six rats. Also shown are effects produced by pretreatment with RS21361 10 mg kg^{-1} (▲); values are means with vertical lines indicating s.e.mean.

unexpected effects. At all levels of xylazine, naloxone produced a slightly but mostly insignificant fall in the hypothermic effect and the values for 80 min are shown in Figure 3. Significant reversal of hypothermia was seen at 40 min with a dose of 15 mg kg^{-1} ($P < 0.05$) but this was the only significant example recorded. The effect of 5 mg kg^{-1} naloxone on the hypothermia produced by clonidine however was very different from that seen with xylazine. Figure 3 shows that at 80 min there was a significant ($P < 0.05$) potentiation of hypothermia at all but the lowest doses of clonidine. Similar effects were recorded at 120 min, but at 40 min significant potentiation was seen only at 0.5 and 1.0 mg kg^{-1} clonidine.

Discussion

Although clonidine and xylazine both produced a clear dose-dependent hypothermia in the rat, it would appear from the effects of α_2-antagonists and naloxone that the mechanism by which this hypothermia was produced is different for the two drugs.

Although clonidine and xylazine are fairly similar in structure and are both classed as α_2-agonists, there are several clear differences in their effects *in vivo*. Xylazine is generally a less potent drug in terms of both hypotension and sedation and its clinical use is

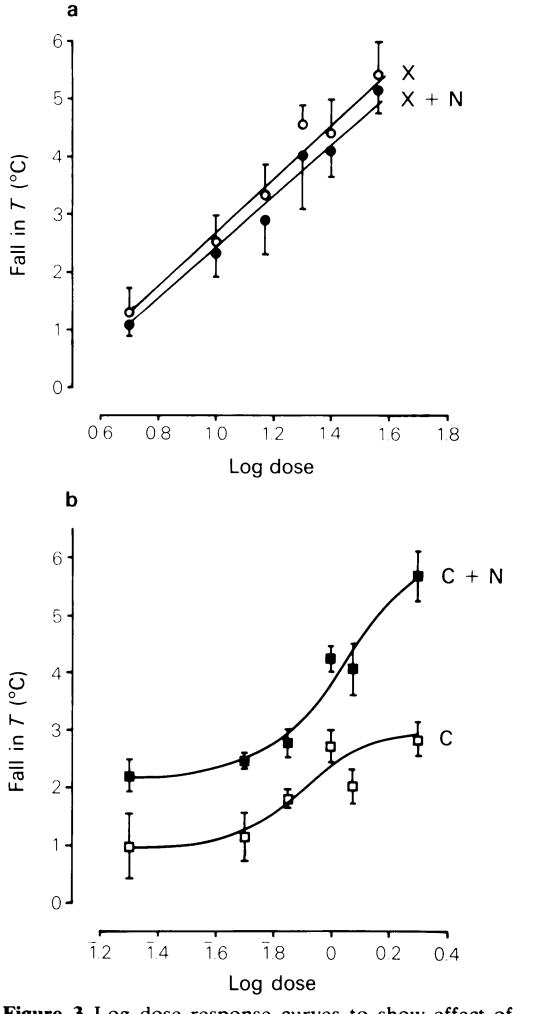

Figure 3 Log dose-response curves to show effect of pretreatment with naloxone (N) 5 mg kg^{-1} (●,■) on the depression of rectal temperature produced by xylazine (X) in (a) and clonidine (C) (○,□) in (b), 80 min after injection in groups of six rats; values are means with vertical lines indicating s.e. mean.

as a veterinary sedative and hypnotic with the hypotension being classed as a side effect. In contrast, clonidine is used clinically as a hypotensive with sedation as a side effect. It is interesting to note, however, that the hypotension which might be expected to arise from the peripheral sympatholytic activity of clonidine, is in fact attributed to a central effect (Kobinger & Walland, 1967; Constantine & McShane, 1968). Thus both xylazine and clonidine can act directly on the central nervous system.

As stated earlier, the central control of body temperature is located in the hypothalamus and several amines have been implicated in the various mechanisms involved. Both clonidine and xylazine may interfere with noradrenergic mechanisms presynaptically and there is also much evidence to suggest a postsynaptic mode of action for clonidine in the CNS (Haensler & Finch, 1972; Greenberg, U'Prichard & Snyder, 1976; Warnke and Hoefke, 1977). In addition it has been suggested that clonidine has inhibitory actions both on 5-HT-mediated responses in the spinal cord (Franz, Hare & Neumayr, 1978) and on 5-HT turnover (Anden, Corrodi, Fuxe, Hokfelt, Hokfelt Rydin & Svensson, 1970). Moreover, there are reports of 5-HT endings having both 5-HT and α_2-receptors (Timmermans & van Zweiten, 1982); consequently it is possible that clonidine may be exerting its effects on body temperature via 5-HT neurones rather than through noradrenaline. Unfortunately, there is little information on the central actions of xylazine, but the greater sensitivity to the actions of yohimbine suggests that its influence may be solely via noradrenaline. The potentiating effects of naloxone on clonidine hypothermia also suggests the involvement of 5-HT since it has been suggested (Feldberg & Lotti, 1967) that the hypothermic effects of morphine may be mediated via a 5-HT mechanism and the hypothermia seen with morphine was sensitive to antagonism by naloxone-like drugs (Lotti, Lomax & George, 1965). Again, this effect was not seen with xylazine which would also suggest that xylazine was acting via noradrenergic mechanisms.

The hypothermic effects of naloxone in conjunction with clonidine may be related to the findings of Geller, Hawk, Keinath, Tallarida & Adler (1983) who examined the effect of naloxone on various types of opioids affecting body temperature. They found that one group of opioids including meperidine, normorphine and (+)-pentazocine, which had no effect on body temperature themselves, produced hypothermia when used in conjunction with naloxone.

Thus it would appear that clonidine and xylazine, whilst both producing hypothermia, do so via different mechanisms.

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