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THERMOREGULATORY CHANGES INDUCED BY CHOLINOMIMETIC SUBSTANCES INTRODUCED INTO THE CEREBRAL VENTRICLES OF SHEEP

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1 Thermoregulatory responses have been recorded from Welsh Mountain sheep exposed to warm, neutral or cold environments while injections of cholinomimetic drugs and/or their antagonists have been given into a lateral cerebral ventricle.

2 Carbachol and physostigmine inhibited panting of animals at high ambient temperature (t_a), caused vasoconstriction and initiated shivering at neutral t_a, and accentuated shivering at low t_a. Rectal temperature (t_{re}) invariably increased. Oxotremorine had apparently identical effects.

3 Nicotine and another ganglionic stimulant, the quaternary methyl derivative of dopamine, had no effects on thermoregulation.

4 Atropine given 10 min before injections of carbachol, physostigmine or oxotremorine completely inhibited their hyperthermic effects, but pretreatment with the ganglion-blocking drug, pempidine, caused no inhibition. The cholinergic synapses that respond to cholinomimetic drugs injected into the lateral cerebral ventricles of sheep are therefore muscarinic and not nicotinic.

5 When atropine was given to sheep exposed to cold, no detectable reduction of shivering occurred and t_{re} decreased only slightly, even with doses of atropine far greater than needed to inhibit shivering induced by physostigmine. This may be because shivering is controlled by neural pathways unaffected by drugs administered intracerebroventricularly or because the cholinergic synapses activated by physostigmine do not carry the input from cold sensors.

Introduction

Administration of putative neurotransmitters to the cerebroventricular space is a technique used to study the influence of such substances on nerve pathways close to the ventricular wall. In an analysis of the central control of body temperature made with this technique, Bligh, Cottle & Maskrey (1971) found that intracerebroventricular (i.c.v.) injections of certain cholinomimetic drugs into sheep increased body temperature. Since the rise in body temperature was brought about in animals at low ambient temperature (t_a) by an increase in heat production, and in animals at high t_a by an inhibition of heat loss, it was proposed that the drugs were acting at cholinergic synapses in the neural path between cold sensors and heat production effectors at a point before the divergence of an inhibitory link to a path presumed to run between warm sensors and heat loss effectors. The reports by Maskrey (1971) that atropine injected into the cerebral ventricles of sheep exposed to cold inhibited shivering and that the responses elicited by i.c.v. carbachol or physostigmine could be blocked by prior i.c.v. injection of atropine supported the proposition that an atropine-sensitive cholinergic synapse exists in the neuronal chain linking the input from cold sensors with the output controlling heat production.

Cats respond similarly to sheep when given i.c.v. injections of those cholinomimetic drugs whose actions are blocked by atropine (Hall, 1972; Baird & Lang, 1973). In cats, i.c.v. administration of nicotine also elicits a thermoregulatory response, involving activation of heat loss mechanisms and a fall in body temperature. This has been taken to indicate that nicotinic receptors exist in the neural pathway controlling heat loss (Hall, 1972; Baird & Lang, 1973). The 'tremorgenic' drugs, tremorine and oxotremorine, have also been injected into the CNS of cats to induce a condition akin to Parkinsonism (Conner, Rossi & Baker, 1966a). These drugs elicit responses somewhat similar to those seen after injections of analogues of acetylcholine (ACh) when introduced

into the cerebral ventricles, the hypothalamus or the caudate nucleus (Connor *et al.*, 1966a & b; Hall, 1972; Rudy & Wolf, 1972). Body temperature usually rises although the rise depends on drug dose and can be variable, and an increase occurs in involuntary muscular activity but whether this is 'tremor' or 'shivering' is uncertain (Rudy & Wolf, 1972).

The present experiments were carried out to gain further information about the nature of the sensitivity to cholinomimetic substances injected into the cerebroventricular space of sheep; the drugs nicotine and oxotremorine have been included to determine any related influence they may have on thermoregulatory control. A preliminary report of the experiments has been made (Johnson & Smith, 1974).

Methods

Experiments were done on nine castrated male Welsh Mountain sheep weighing between 31.6 and 52.5 kg. The animals were shorn every 3 weeks and kept indoors at 25 ± 3° C t_a to standardize the thermal stress. When experiments were done at neutral temperatures, animals were placed in a temperature controlled chamber maintained at 25° C t_a. For experiments in hot conditions the temperature in the chamber was raised to 40° C and for experiments in cold conditions it was lowered to 10° C, except when extreme cold was required when t_a was lowered to between −2° and +4° C.

Experiments usually lasted for 3-4 h and were begun in the morning 17 h after the animals were fed. Measurements were made of rectal (t_{re}), trunk skin and ear temperature with fine uncovered thermocouples, the output of each of which was registered on a potentiometric recorder every 36 seconds. Respiratory movements were sensed with a pneumatic belt and recorded for 30 s every 5 min; respiratory frequency was counted for periods of 10-20 seconds. Electromyograms (EMG) were detected from needle electrodes through the skin of the thigh and recorded for 30 s in every 5 minutes. The occurrence of shivering was also checked visually and, on the basis of such checks and the EMG records, shivering was scored numerically between zero (no shivering) and five (sustained vigorous shivering).

Injections or infusions were made into a lateral cerebral ventricle through a stainless steel cannula (Barton, Bligh & Sharman, 1968) implanted under general anaesthesia at least 1 week before the animal was first used in an experiment. The accurate placement of each cannula was confirmed by x-radiography. Only one drug was injected in each 3-4 h experiment unless the interaction of two drugs was being studied; then the injections were given 10 min apart. Recordings were made for 1 h before the first injection and for 2-3 h afterwards. Drug doses were related to body weight and administered in a volume of 0.005 ml/kg; the volume of drug solution injected was therefore approximately 0.2 ml and it was flushed into the ventricle with 0.2 ml isotonic saline. Drugs were dissolved in sterile physiological saline (0.9% w/v NaCl solution) which was then passed through a 0.22 μm Millipore filter before injection.

The drugs were atropine sulphate (B.D.H.), carbachol (carbamylcholine chloride, B.D.H.), physostigmine sulphate (B.D.H., eserine), nicotine hydrogen (+)-tartrate (B.D.H.), oxotremorine hydrochloride (May & Baker Research Laboratories), pempidine tartrate (May & Baker) and the quaternary methyl derivative of dopamine (3,4-dihydroxyphenylethyltrimethylammonium iodide, prepared by Dr J.D.M. Pearson, A.R.C. Babraham).

Control experiments were done for all drug treatments. To test whether the injection procedure itself elicited a response, 0.4 ml saline was given i.c.v. 1 h before injection of the drug in 96 experiments. To check for any response to injection of a volume of 0.8 ml, an initial injection of 0.4 ml saline was followed after 1 h by a second injection of 0.4 ml saline in 6 experiments. In a further 6 experiments injections of sodium sulphate 6 nmol/kg were given as controls for the injections of physostigmine sulphate 12 nmol/kg. In no control experiment was any significant change observed in the thermoregulation of the animal.

Results

The effects of each drug (except oxotremorine) were examined at four or more dose levels. Each dose was given to 2 animals but all four doses were not necessarily given to the same two animals. Values given in the text or figures are mean values from experiments on two animals.

Physostigmine

Physostigmine was injected in doses of 1.5, 3, 6 and 12 nmol/kg into the cerebral ventricles of 6 sheep held at 10°, 25° or 40° C t_a. The non-thermoregulatory behaviour of all animals was normal except when 12 nmol/kg physostigmine caused some animals to become restless and noisy for about 5 minutes. The responses to all doses of physostigmine were an inhibition of panting in

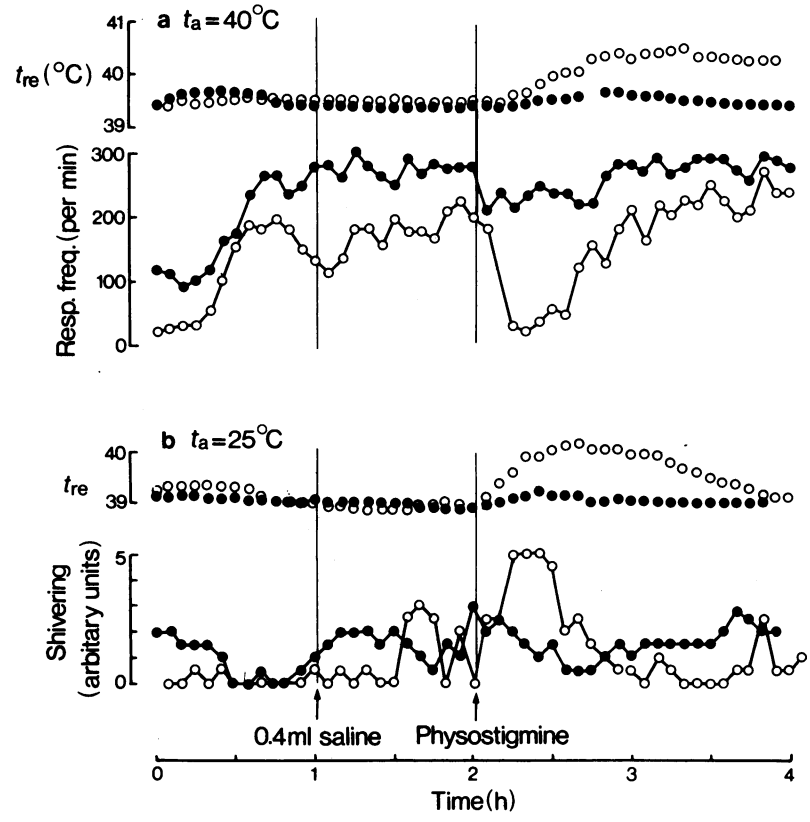

Figure 1 Effects of physostigmine 1.5 nmol/kg (•) or 12 nmol/kg (○) on (a) rectal temperature (t_{re}) and respiratory frequency at 40° C ambient temperature (t_a) and (b) t_{re} and shivering at 25° C t_a.

animals at 40° C t_a, an initiation of shivering in animals at 25° C and an increase in the intensity of shivering which already existed in animals at 10° C; in all animals t_{re} rose. Ear temperature was near t_a in all experiments and did not alter following injections of physostigmine. The shivering or respiratory responses and the changes in t_{re} which followed injections of the largest (12 nmol/kg) and smallest (1.5 nmol/kg) doses of physostigmine at 25° and 40° C t_a are shown in Figure 1. The rise in t_{re} which followed 3 nmol/kg physostigmine was intermediate between the rises recorded after 1.5 and 12 nmol/kg, but the rise after 6 nmol/kg was smaller than after 1.5 nmol/kg; why this response to 6 nmol/kg did not follow the dose-response relation for 1.5, 3 and 12 nmol/kg is not clear. At 10° C, the pattern of the response to physostigmine was similar to that at 25° C.

Carbachol

Carbachol was injected in doses of 0.7, 1.4, 2.7 or 5.5 nmol/kg into 6 sheep held at 10°, 25° and 40° C t_a. The respiratory and shivering responses observed were qualitatively similar to those seen after injections of physostigmine, although not as consistent. At 25° C t_a, the ear temperatures of some animals were near to t_{re}; following injections of carbachol the ear temperatures of these sheep decreased to near t_a indicating a peripheral vasoconstriction. The t_{re} rose by an average of 0.63° C within 1 h of injection in 19 of 24 experiments, but t_{re} was unchanged in 2 experiments and fell in 3 experiments. One of the animals used was inconsistent in its responses and the 3 experiments in which t_{re} decreased after carbachol injection were all done on this animal. However, even when the observations on this animal are disregarded, the rises in t_{re} measured following carbachol injections were not obviously correlated with the doses of carbachol administered.

Nicotine

Nicotine was injected i.c.v. into 4 sheep in single

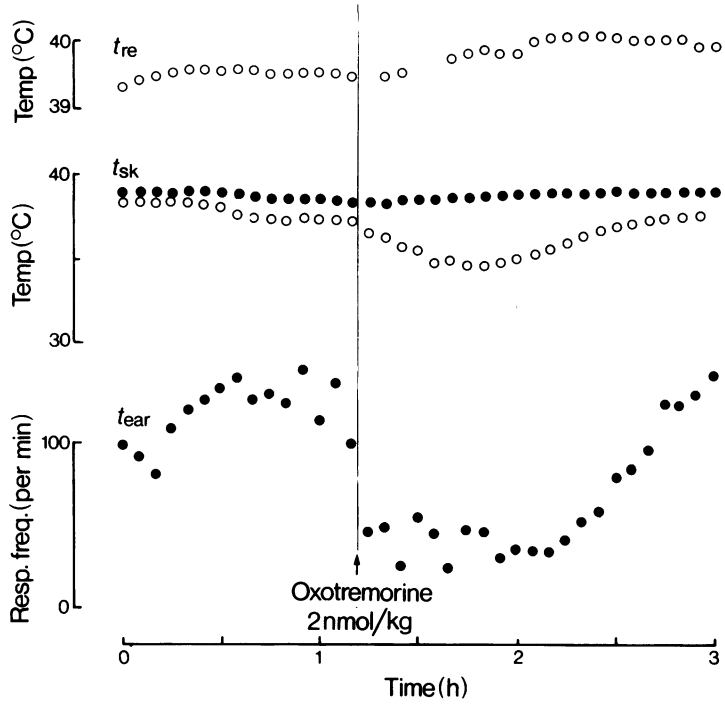

Figure 2 Effect of oxotremorine 2 nmol/kg on rectal (t_{re}), trunk skin (t_{sk}) and ear (t_{ear}) temperatures and respiratory frequency of sheep at 32° C ambient temperature.

doses ranging from 6 to 48 nmol/kg at 10°, 25° and 40° C t_a. No changes in t_{re} were observed following injections of doses of 24 nmol/kg or less, but after administration of 48 nmol/kg, animals became very restless for 15-20 min and during that period t_{re} rose. Continuous infusions of nicotine were made into the ventricles of 4 animals at dose rates between 2 and 6 nmol kg^{-1} min^{-1} for periods of 70-80 minutes. No thermoregulatory changes were observed and t_{re} increased by an average of only 0.3° C in that time.

The quaternary methyl derivative of dopamine is 3-12 times more effective than nicotine as a ganglionic stimulant in a number of tissue preparations (Cuthbert, 1964). In 8 experiments on 3 sheep, i.c.v. injections of this substance in doses ranging from 0.5 to 50 nmol/kg were completely ineffective in eliciting any thermoregulatory response or alteration in t_{re}.

Oxotremorine

Injections of 2 nmol/kg oxotremorine given into the cerebral ventricles of 2 sheep held at 25° C t_a caused an immediate fall in ear temperature, initiation of muscular activity indistinguishable from shivering and rises in t_{re} of 0.5° and 1.2° C in the two animals during the following 75 minutes. When the same dose of oxotremorine was given to 2 animals with moderately elevated respiratory frequencies held at 32° C t_a, ear temperature and respiratory frequencies both began to decrease immediately and t_{re} increased by 0.5° and 0.7° C in the 2 animals during the following 70 min (Figure 2).

Atropine

Atropine injected alone into the cerebroventricular system of 4 sheep in doses of between 1.25 and 10 nmol/kg at 10°, 25° and 40° C t_a caused neither consistent nor substantial changes in the thermoregulatory pattern. This lack of response was unexpected because Maskrey (1971) had reported that atropine inhibited shivering and decreased body temperature of sheep exposed to cold; further experiments were therefore done with atropine administered at higher doses to sheep at lower t_a, in which conditions Maskrey (1971) suggested the response was best observed. In 6 experiments, animals were injected i.c.v. with 10 or 20 nmol/kg atropine at t_a between −2° and +4° C. Throughout these experiments the animals continued to shiver strenuously, and even after the

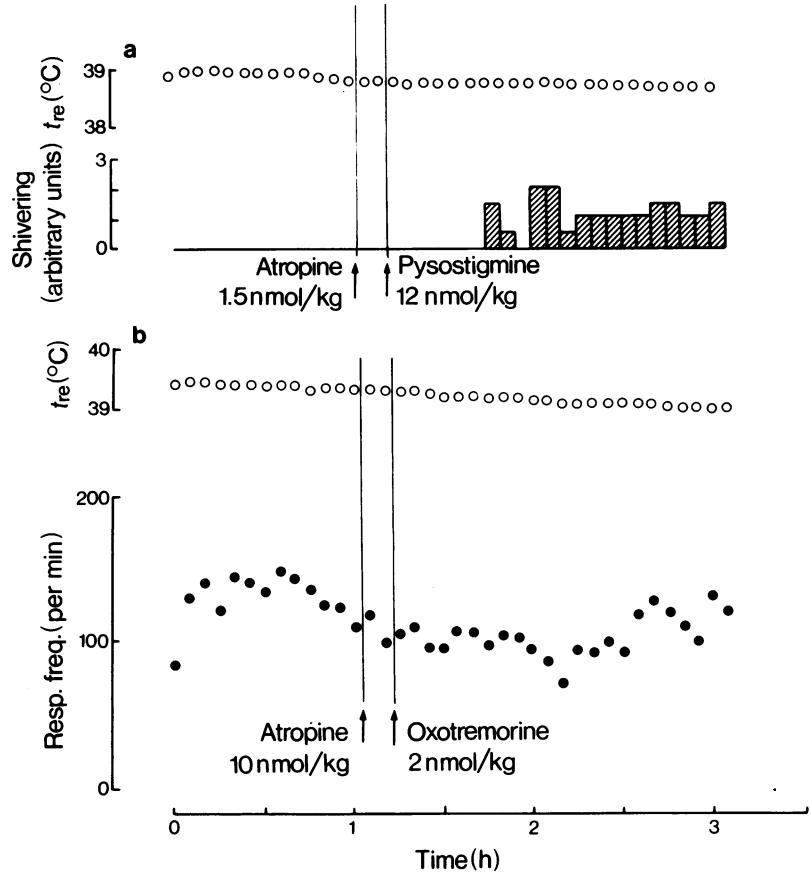

Figure 3 Inhibition by prior administration of atropine of the hyperthermic effects (a) of physostigmine given to sheep at 25° C ambient temperature (t_a) and (b) of oxotremorine given to sheep at 28° C t_a.

injection of 20 nmol/kg atropine i.c.v., t_{re} decreased by an average of only 0.26° C in 2 hours.

When atropine 12 nmol/kg was injected i.c.v. 10 min before physostigmine 12 nmol/kg, the initiation of shivering and rise in t_{re} usually seen after physostigmine injection no longer occurred. In successive experiments, the atropine dose preceding the physostigmine was reduced from 12 to 6, 3, 1.5 and 0.15 nmol/kg. An atropine dose of 1.5 nmol/kg completely blocked the hyperthermic effect of 12 nmol/kg physostigmine (Figure 3a) but an atropine dose of 0.15 nmol/kg did so incompletely. Atropine also provided a complete block of the thermoregulatory effects induced by oxotremorine. If atropine (10 nmol/kg, i.c.v.) was given 10 min before oxotremorine 2 nmol/kg, animals at 25° C t_a no longer began to shiver and at higher t_a the reduction in panting usually induced by i.c.v. oxotremorine no longer occurred (Figure 3b).

The effect of carbachol was only partially inhibited by doses of atropine which were completely effective against physostigmine and oxotremorine. Some shivering and a slight rise in t_{re} were recorded when carbachol 6 nmol/kg was injected 10 min after atropine 10 nmol/kg and these responses were even greater when only 5 nmol/kg atropine was given prior to the carbachol.

Pempidine

Pempidine is a long-acting ganglion-blocking agent which inhibits the peripheral nicotinic actions of cholinomimetic drugs (Corne & Edge, 1958). When injected in doses of between 3 and 24 nmol/kg into 4 sheep held at 10°, 25° or 40° C t_a, pempidine caused no change in thermoregulation and led to no change in t_{re}. When pempidine (24 nmol/kg i.c.v.) was injected 10 min

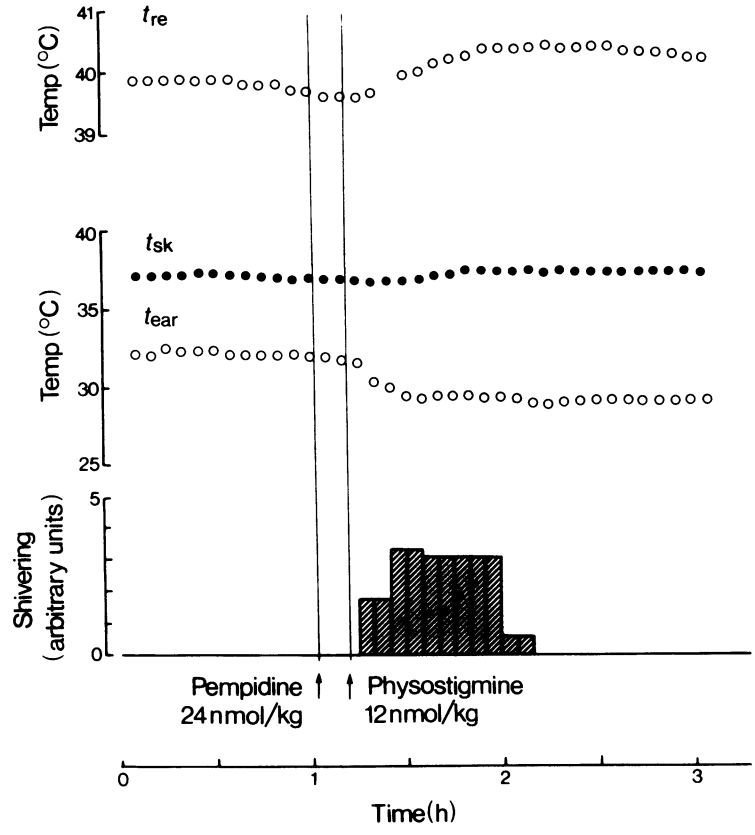

Figure 4 Failure of the ganglion-blocking drug, pempidine, to inhibit the rises in rectal temperature (t_{re}) and skin temperature (t_{sk}), fall in ear temperature and initiation of shivering following i.c.v. physostigmine given to sheep at 25° C ambient temperature.

before injections of physostigmine (12 nmol/kg) (Figure 4), carbachol (6 nmol/kg) or oxotremorine (2 nmol/kg) the inhibition of heat loss, accentuation of heat production and rise in t_{re} usually observed when these drugs were injected alone occurred apparently unaltered by the pretreatment with pempidine.

Discussion

The report of sensitivity to cholinomimetic drugs in the cerebroventricular space of sheep made by Bligh *et al.* (1971) is substantiated by the observations made here. Both the acetylcholine analogue, carbachol, and the anticholinesterase, physostigmine, inhibited panting of animals in the heat, caused vasoconstriction in the ears when this did not already exist and, in cool conditions, initiated or accentuated shivering. Since the physostigmine presumably had its effect by raising the levels of endogenous ACh, the response to both drugs can reasonably be attributed to stimulation of cholinergic synapses adjacent to the ventricular wall. The carbachol and physostigmine increased heat production at low t_a and decreased heat loss at high t_a; therefore, the cholinergic synapses might be considered to lie in the neural path by which heat production is controlled, at a point before the origin of a path inhibiting heat loss, as Bligh *et al.* (1971) suggested. Whether the same cholinergic synapses transmit information from cold sensors will be discussed later.

When nicotine was given in progressively increasing doses, no initiation or inhibition of thermoregulatory responses was ever observed. The highest single dose used (48 nmol/kg) caused animals to become restless for 15-20 min and t_{re} rose but this was the result of greater activity and not of a coordinated thermoregulatory effect. Continuous infusions of nicotine or injections of a powerful ganglionic stimulant, the quaternary

methyl derivative of dopamine, were similarly ineffective, causing only restlessness when given in large doses. No receptors sensitive to i.c.v. nicotine therefore appeared to exist in the thermoregulatory control system of these sheep.

These observations are supported by the findings of complete, or almost complete, inhibition by atropine of the effects of intracerebroventricularly administered carbachol, physostigmine and oxotremorine, and the lack of effect of the ganglion-blocking drug, pempidine. Because no natural or induced thermoregulatory response was ever blocked by pempidine, one cannot be certain that an effective dose of this drug was used; but since pempidine 24 nmol/kg did not block the effect of physostigmine 12 nmol/kg whereas atropine 1.5 nmol/kg inhibited the response completely (Figure 3a), atropine must be at least 16 times more effective in blocking these cholinergic synapses than pempidine. In pharmacological terms, the sensitivity to cholinomimetic substances given into the lateral cerebral ventricles of sheep was muscarinic and not nicotinic.

The action of oxotremorine has frequently been related to the activity of ACh (e.g. George, Haslett & Jenden, 1966), and oxotremorine has been described as the most potent muscarinic agent known (Holmstedt, 1967). The similarity of the responses to oxotremorine and to other cholinomimetic drugs was, therefore, not surprising. From the present observations, there was no reason to believe that the response to oxotremorine was due to stimulation at a different site from that affected by physostigmine, carbachol and atropine. The muscular activity induced by i.c.v. injection of oxotremorine was not apparently different from shivering. Although details of the EMG were not studied closely, the muscular response to oxotremorine was similar, both in the EMG and visually, to that of sheep exposed to cold and to that induced by i.c.v. physostigmine or carbachol. Injections of oxotremorine which elicited an increased muscular activity at neutral t_a did so to a lesser extent or not at all in animals in warmer conditions; the rise in t_{re} in such conditions resulted from an inhibition of panting. Therefore, although there was no unequivocal identification of shivering and differentiation from tremor, the results indicated that, like other cholinomimetic drugs given i.c.v. in sheep, oxotremorine was affecting primarily thermoregulatory control rather than muscular tremor.

Although the observations made here confirm the report of Bligh *et al.* (1971) of cholinomimetic sensitivity in the cerebroventricular system of sheep, they are not entirely consistent with their proposal that the sensitivity is due to the presence of a cholinergic synapse in the neural pathway between cold sensors and heat production effectors. Physostigmine injected i.c.v. led to shivering and elevation of t_{re} presumably by allowing the level of endogenous ACh in some sensitive region to increase. Atropine, a competitive antagonist of Ach, completely inhibited the shivering and hyperthermia, presumably by counteracting this effect of endogenous ACh. However, atropine given alone in doses more than 10 times greater than required to block the action of physostigmine did not appear to change the intensity of shivering of animals exposed to cold, and t_{re} decreased only very slightly. Similar observations have been made on cats. Although 0.5 mg/kg atropine given intraperitoneally completely inhibited the tremor induced by carbachol injected into the caudate nucleus (Connor *et al.*, 1966b), larger doses of atropine (1-3 mg/kg i.p.) had no inhibitory effect on cold-induced shivering and led to no significant change in t_{re} (Stuart, George, Freeman, Hemingway & Price, 1961). An immediate deduction that the site of action of physostigmine, and possibly also of carbachol and oxotremorine, might not be on the cold sensor-heat production pathway is, however, only one of several explanations which can be offered for the observation.

One reason why cold-induced shivering was not depressed by doses of atropine which totally inhibited the response to i.c.v. physostigmine may be that shivering can be activated by a relatively small proportion of the periventricular neural pathways which are apparently involved in the control of heat production. If this were so, then i.c.v. injections of physostigmine, carbachol and oxotremorine could initiate shivering by stimulating these pathways, and atropine would block the effect. During exposure to cold, similar pathways either more distant from the site of injection or more remote from the ventricular wall might carry sufficient facilitatory information from peripheral cold sensors to initiate and maintain shivering despite the presence within the ventricles of a dose of atropine many times that necessary to block a stimulatory dose of a cholinomimetic drug.

A related explanation depends on the proposition that the main control of shivering may be exercised by structures quite outside the region affected by drugs added to the cerebroventricular space, possibly in the spinal cord. Evidence can be adduced for some extra-hypothalamic control of shivering, with only limited facilitation or inhibition arising from the hypothalamic region (Bligh, 1973). If, in the cold conditions used in the present experiments, only a limited facilitation of shivering was exerted by neurones adjacent to the cerebral ventricles, block of this small influence by

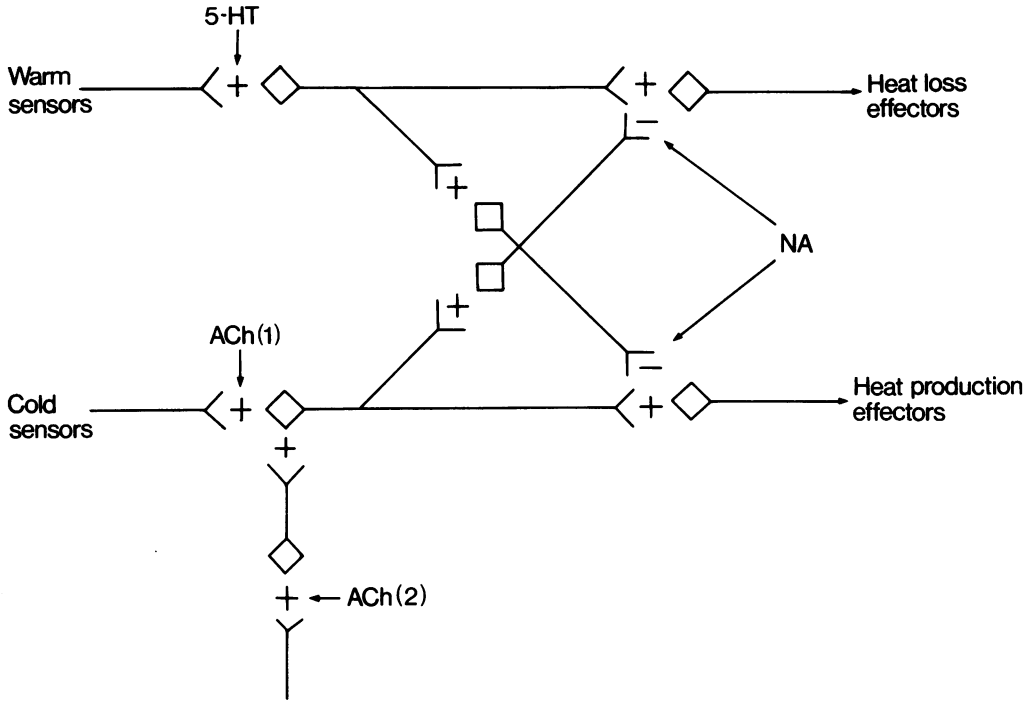

Figure 5 Neuronal model of the central control of body temperature in sheep, proposed by Bligh, Cottle & Maskrey (1971). Acetylcholine (ACh) (1) is the suggested site of a cholinergic synapse according to Bligh *et al.*; ACh (2) is a possible alternative site of a cholinergic synapse suggested by the present results. 5-HT = 5-hydroxytryptamine; NA = noradrenaline.

atropine may have passed undetected as a slight decrease in shivering and led to the observed small fall in core temperature. However, if such is the explanation, the thermoregulatory system proposed by Bligh *et al.* (1971) cannot be considered to be truly descriptive of the central control of body temperatures of sheep during exposure to cold.

A further possibility is that the cholinergic synapses activated by cholinomimetic drugs do not carry input signals from cold sensors. If the cholinergic synapses occurred, not in the input pathway from cold sensors as postulated by Bligh *et al.* (1971), but in an input converging onto the pathway between cold sensors and heat production effectors after the afferent input and before the divergence of the proposed crossed inhibition to the parallel pathway between warm sensors and heat loss effectors, the predicted behaviour of such a neuronal network would be consistent with all observations. In the format used by Bligh *et al.* (1971) the network would be represented as shown in Figure 5.

The report by Maskrey (1971) that i.c.v. atropine blocked shivering when given to cold-exposed sheep under conditions virtually identical to those used here remains unexplained by any of the proposals made above, and further experiments are needed to determine the repeatability of the response. A more detailed analysis is also required of the muscular activity elicited by i.c.v. injections of cholinomimetic drugs to determine whether it is tremor, shivering or a combination of the two. The fact that the muscular activity induced by i.c.v. cholinomimetic drugs in sheep is coordinated with changes in panting and peripheral vasomotor tone indicates strongly that the overall response is thermoregulatory and that the muscular activity is, at least partly, shivering. The general conclusion remains that, in sheep, atropine-sensitive cholinergic synapses apparently occur close to the ventricular wall in neural pathways involved in thermoregulatory control.

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References

BAIRD, J. & LANG, W.J. (1973). Temperature responses in the rat and cat to cholinomimetic drugs injected into the cerebral ventricles. *Eur. J. Pharmac.*, **21**, 203-211.

BARTON, A.J., BLIGH, J. & SHARMAN, D.F. (1968). Improved techniques for the chronic cannulation of the lateral cerebral ventricle and the cisterna magna of the Welsh Mountain sheep. *J. Physiol., Lond.*, **200**, 25-27P.

BLIGH, J., COTTLE, W.H. & MASKREY, M. (1971). Influence of ambient temperature on the thermoregulatory responses to 5-hydroxytryptamine, noradrenaline and acetylcholine injected into the lateral cerebral ventricles of sheep, goats, and rabbits. *J. Physiol., Lond.*, **212**, 377-392.

BLIGH, J. (1973). *Temperature Regulation in Mammals and Other Vertebrates*, p. 74. Amsterdam: North-Holland.

CONNOR, J.D., ROSSI, G.V. & BAKER, W.W. (1966a). Analysis of the tremor induced by injections of cholinergic agents into the caudate nucleus. *Int. J. Neuropharmac.*, **5**, 207-216.

CONNOR, J.D., ROSSI, G.V. & BAKER, W.W. (1966b). Characteristics of tremor in cats following injections of carbachol into the caudate nucleus. *Expl. Neurol.*, **14**, 371-382.

CORNE, S.J. & EDGE, N.D. (1958). Pharmacological properties of pempidine (1:2:2:6:6-pentamethylpeperidine), a new ganglion-blocking compound. *Br. J. Pharmac. Chemother.*, **13**, 339-349.

CUTHBERT, M.F. (1964). Relative actions of quaternary methyl derivatives of tyramine, dopamine and noradrenaline. *Br. J. Pharmac. Chemother.*, **23**, 55-65.

GEORGE, R., HASLETT, W.L. & JENDEN, D.J. (1966). The production of tremor by cholinergic drugs: central sites of action. *Int. J. Neuropharmac.*, **5**, 27-34.

HALL, G.H. (1972). Changes in body temperature produced by cholinomimetic substances injected into the cerebral ventricles of unanaesthetized cats. *Br. J. Pharmac.*, **44**, 634-641.

HOLMSTEDT, B. (1967). Mobilization of acetylcholine by cholinergic agents. *Ann. N.Y. Acad. Sci.*, **144**, 433-453.

JOHNSON, K.G. & SMITH, C.A. (1974). The nature of the thermoregulatory sensitivity to cholinomimetic drugs in the cerebral ventricles of sheep. *J. Physiol., Lond.*, **236**, 31-32P.

MASKREY, M. (1971). Transmitters and modulator substances in the control of body temperature. Ph.D. thesis, Univ. of Cambridge.

RUDY, T.A. & WOLF, H.H. (1972). Effect of intracerebral injections of carbamylcholine and acetylcholine on temperature regulation in the cat. *Brain Res.*, **38**, 117-130.

STUART, D.G., GEORGE, R., FREEMAN, W.J., HEMINGWAY, A. & PRICE, W.M. (1961). Effects of anti- and pseudo-Parkinson drugs on shivering. *Expl Neurol.*, **4**, 106-114.

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