released by the action of amphetamine. However, topically applied noradrenaline had no significant effect on local blood flow in the present experimental conditions. Only isoproterenol showed a small effect on CoBF which is in line with previous observations[7,8]. An increasing amount of evidence points to the idea that cerebral blood flow might be under the control of neurogenic mechanisms originating in the brain stem[9-14]. The fact that chlorpromazine[15,16] which is known to depress brain stem mechanisms, completely blocked the cerebrovascular effect of amphetamine suggests that the reported indirect effect of amphetamine on cortical blood vessels might be related to the activation of a dilatatory system originating in the brain stem and projecting to the cerebral cortex. The blockade by atropine of the cerebrovascular effect of amphetamine is suggestive that the proposed brain stem mechanism might include a cholinergic step.

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Are there 2 cholinergic thermoregulatory centres in rats?

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Summary. An attempt was made to replicate the conflicting previous reports of hypo- and hyperthermic effects of intrahypothalamically administered carbachol. Despite using the same coordinates, injection parameters, and strain of rats reported by others, only hypothermia was conclusively demonstrated. It was concluded that the cholinergic system mediates heat loss mechanisms in rats.

There exists a considerable controversy over the role of the cholinergic system in the central control of thermoregulation in rodents. Despite a wealth of studies indicating that acetylcholine (ACh) may mediate heat loss mechanisms[1-6], some investigators still maintain that ACh is involved in heat gain mechanisms and have even developed elegant models of thermoregulation on the basis of scanty evidence[7-10]. Since there were some variations in technique and other procedures between these conflicting studies, it was decided to attempt to replicate two of the key experiments in this area[11,12]. The results of the present studies are consistent with the view, expressed by the majority of research workers in this field, that a central cholinergic system mediates heat loss mechanisms in rats.

In one experiment female Sprague-Dawley rats, approximately 130–190 g and 70 days old, were unilaterally

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Maximum deviation of temperature and time to reach maximum effect after intrahypothalamic injections of carbachol

Treatment group	N	Sex	Coordinates	Maximum deviation (°C) (mean ± SEM)	Time for response (min) (mean ± SEM)
2 µg carbachol / 1 µl saline	6	F	AP 1.8 ML 1.5 DV − 8.5	− 2.7 ± 0.3	27 ± 1
2 µg carbachol / 1 µl saline	3	M	AP 1.7 ML 0.8 DV − 8.5	− 1.1 ± 0.4	20 ± 4
5 µg carbachol / 0.5 µl saline	6	M	AP 1.7 ML 0.8 DV − 8.5	− 2.1 ± 0.5	34 ± 8

cannulated (23 gauge stainless steel cannula guides) under sodium pentobarbital (35 mg/kg) anesthesia. The tip of the cannula was aimed for the anterior preoptic area of the hypothalamus[12]. 1–2 weeks after surgery, the rats were given a central injection of 2 μg of carbachol in a volume of 1 μl of isotonic saline solution. Body temperatures were recorded by means of a thermistor probe that was inserted 6–8 cm into the rectum and taped loosely to the tail. The animals were virtually unrestrained while temperatures were being recorded.

In the second experiment male Hooded-Wistar rats, approximately 260–320 g and 90 days old, were used. They were unilaterally cannulated in the anterior preoptic area at one of 2 sites: AP 1.7, ML 0.8–1.0, DV −8.5[11] or AP 1.8, ML 1.5, DV −8.5[12] in order to produce eithera hyper- or hypothermic response, respectively. Body temperatures were recorded after either 2 μg carbachol in 1 μl isotonic saline[12] or 5 μg carbachol in 0.5 μl isotonic saline[11].

The results of these experiments are summarized in the table. There was a mean decrease of 2.7 °C in 6 of the 11 female rats which exhibited any obvious change in temperature. Histological examination revealed that those 6 which exhibited hypothermia had their cannula tips in the lower boundary of the anterior preoptic area, while those which did not respond were found to have their cannula tips below the lower border of the anterior preoptic area.

Many of the male rats dislodged their cannula guides before they could be tested. Both animals which were given 2 μg of carbachol in 1 μl of isotonic saline at the coordinates used by Overstreet et al.[12] exhibited hypothermia. So did those animals which had cannulae aimed at the coordinates of Avery[11], as can be seen in the table. Hypothermia was observed regardless of the dose or volume of injections. Control injections of isotonic saline produced no obvious temperature variation.

These results have confirmed the reports of many previous workers[1–6,12] which have shown that intrahypothalamic administration of cholinergic agonists produces a hypothermic response, but have failed to replicate the findings of Avery[7–11] of hyperthermia following intrahypothalamic injections. Since the present experiments used the same anatomical coordinates, parameters of injection and strain and sex of rats used in previous conflicting reports, these variables do not appear to be the critical ones in accounting for differences in findings. However, because of loosening of the cannulae, the actual sites of injections could not be conclusively pin-pointed. Therefore, the possibility that there are 2 cholinergic thermoregulatory centres, one for heat loss and one for heat gain, must remain open.

Nevertheless, it should be emphasized that the evidence for a cholinergic system mediating heat loss mechanisms in rats in much more compelling. One example is that atropine has been shown to block the hypothermic effects of centrally administered carbachol[13], but there is no evidence on whether it blocks the hyperthermic effects of this cholinomimetic. Until such evidence is forthcoming, we suggest that the cholinergic system in the anterior preoptica area is predominantly involved in heat loss mechanisms in rats and that the models of thermoregulation previously reported[9,10] should be regarded with caution.

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Demonstration of microtubule independent protein secretion from rat liver

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Summary. The induced secretion of rat liver microsomal β-glucuronidase into serum is, unlike other proteins secreted from liver, not dependent upon an intact microtubule apparatus.

Secretion in many cell systems has now been documented to be a microtubule dependent, colchicine inhibitable, cellular process. Studies on rodent liver, both in vitro and in vivo, have indicated that the secretion of newly synthesized albumin into serum or incubation media is markedly depressed by colchicine or other agents which disrupt the microtubule network[1,2]. These studies have shown that total hepatic protein synthesis, as measured by amino acid incorporation, is unaffected by the experimental conditions imposed. A build-up of labelled albumin has been detected within hepatocytes in the presence of the anti-microtubule agents coincident with the depression of secretion. Similar findings have been reported by groups studying fibrinogen[3] and lipoprotein[4] secretion from liver into serum. Although morphologic evidence for complete microtubule disruption has not always been obtainable[1], the lack of any effect of lumicolchicine[1,3] suggests a microtubule site of action for the colchicine effect.

The administration of any one of several organo- phosphates to rats has been shown to elicit a specific rise in serum β-glucuronidase levels[5]. This rise is independent of new protein synthesis and the source of the secreted enzyme is the microsomal pool of β-glucuronidase contained in hepatocytes[6]. Microsomal markers including glucose-6-phosphatase and microsomal albumin (immunologically detectable) are unaffected by organophosphate injection. In light of the aforementioned studies, the dependence of diisopropylfluorophosphate (DFP) induced secretion of β-glucuronidase on an intact microtubule system was evaluated.

Fasted, anesthetized 200-g female Wistar rats were used for all studies. At zero time, 25 μCi of ^{14}C D-leucine was injected into the femoral vein with or without colchicine

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