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Higher Environmental Temperature-Induced Increase of Body Temperature: Involvement of Central Opioidergic-GABAergic Interaction

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GHOSH (NEÉ BISWAS), S. AND M. K. PODDAR. *Higher environmental temperature-induced increase of body temperature: Involvement of central opioidergic-GABAergic interaction.* PHARMACOL BIOCHEM BEHAV **52**(1) 73–76, 1995.—Exposure (2 h) of male albino rats to higher environmental temperature (HET, 40°C) significantly increased the body temperature (BT). Administration of bicuculline (1 mg/kg, IP), physostigmine (0.2 mg/kg, IP), or their combination significantly raised the BT of normal rats (kept at 28°C) or of HET-exposed rats. Atropine (5 mg/kg, IP) abolished the hyperthermic effect of bicuculline in normal and HET-exposed rats. The BT of normal and HET-exposed rat was increased with morphine (1 mg/kg, IP) and was reduced with naloxone (1 mg/kg, IP). Bicuculline or physostigmine-induced rise in BT of HET-exposed rats was potentiated following cotreatment of physostigmine with morphine. Atropine-induced hypothermia was abolished due to the cotreatment of atropine with morphine in HET-exposed rats. Further, (morphine + bicuculline)-induced hyperthermia in HET-exposed rats was potentiated with physostigmine but was attenuated with atropine. In normal rats (kept at 28°C), only atropine attenuated (morphine + bicuculline)-induced hyperthermia. L-Dopa + carbidopa or haloperidol did not significantly affect the BT of rats under similar conditions. These results suggest that short-term (2 h) exposure to HET activates the opioidergic neuron, which activates cholinergic activity through the inhibition of GABAergic system and, thus, enhances the BT.

Environmental temperature Body temperature GABA Choline Opioid

IN HOMEOTHERMS, a constant body temperature (BT) is maintained by the balance between the heat loss and heat gain mechanism. The primary site of thermoregulatory control in mammals is the hypothalamic nuclei of the brain, with its array of thermosensitive neurons that received afferent neuronal input from cold and warm sensors in both the periphery and other parts of the central nervous system (CNS) (5,6). Because the balance between heat production and heat loss may be disturbed either by changing the ambient temperature or by other means, the output of thermoeffectors are subject to constant control. The control is provided by neurons distributed in CNS. The central transduction of afferent signals from thermoreceptor into appropiate thermoregulatory output entails complex interneuronal communications, which involves neurotransmitters (15). Considerable evidences now exist to show the involvement of central cholinergic, dopaminergic, and GABAergic systems in thermoregulation (12,16–20). Our previous study (3) shows the possible interaction between central GABAergic and cholinergic system in the regulation of BT of animals exposed to higher environmental temperature (HET).

Ambient temperature profoundly influences the thermic responses to opioids (1,8,21) and other neurotransmitters. In recent years, the role of the opioid system in thermoregulation has been established (1,25). BT may be subjected to modification by endogenous opioids, depending upon the specific peptide released and the type of receptor activated (24). It has been reported that HET-induced hyperthermia is associated with the release of β-endorphin (24). The thermoregulatory effects of β-endorphin are quite similar to morphine (1). Morphine has been reported to affect the BT, depending upon several factors such as species, dose, route of administration, and degree of restraint imposed on the animals as well as on ambient temperature (7,9,10). Because neurotransmitter and

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peptide may coexist within the same neuron and act synergistically in different functions of the brain (2), it is not unlikely that there may be an interaction between opioids and other neurotransmitters. Several evidences have already suggested that the opioid system interacts with the dopaminergic, cholinergic, GABAergic, and other transmitter systems (1,10), although interrelationship among these systems in relation to thermoregulation remains to be explored. In the present investigation, our object is to study the interaction of opioid with central GABA, if any, in the regulation of a HET-induced change in BT.

METHOD

Animals

Male albino rats (120–130 g) of the Charles Foster strain were kept in a 12 L : 12 D cycle at room temperature (28 ± 1°C) with constant relative humidity (80 ± 5%), and were maintained on standard laboratory diet and water ad lib. Experiments were carried out between 1030–1230 h.

Drugs

Bicuculline, atropine sulfate, physostigmine, L-dopa, and naloxone were purchased from Sigma Chemical Company, St. Louis, MO. Carbidopa (peripheral decarboxylase inhibitor) was purchased from Merck, Sharp and Dohm, USA. Haloperidol was purchased from Searle Ltd., Bombay, India.

Physostigmine, morphine, naloxone, haloperidol, and atropine sulfate were solubilized in distilled water. Bicuculline was dissolved in 0.01 (N) HCl. L-Dopa and carbidopa were suspended in distilled water and given orally. All other drugs were administered intraperitoneally (IP).

Drug Treatment and/or Exposure to HET

Animals were divided into different groups of six animals each. The animals of groups 1–16 were treated with vehicle (saline), morphine, naloxone, physostigmine, atropine, L-dopa + carbidopa, haloperidol, and bicuculline individually or in different combinations and were exposed to HET (40 ± 1°C) in a thermostatically controlled ventilated chamber at a fixed relative humidity (80 ± 5%) for exactly 2 h. The animals of groups 17–32, kept in a chamber at room temperature (28 ± 1C) with constant relative humidity (80 ± 5%), were considered as control and were treated with vehicle or the above-mentioned individual drug or the different combinations under similar conditions described in the Experimental Animals section. Doses and durations of treatments of different drugs with and without HET exposure are described in detail in Table 1.

Measurement of Rectal Temperature

Rectal temperature, considered as an index of body temperature, was recorded with a themistor probe inserted 2 cm into the rectum of normal and HET-exposed rats. The rectal temperature of HET-exposed rats (both control and drug treated) was measured at the end of the 2nd hour of HET exposure.

TABLE 1

EFFECT OF ENVIRONMENTAL TEMPERATURE ON RECTAL TEMPERATURE OF RATS TREATED WITH AGONIST AND ANTAGONIST OF OPIOIDERGIC GABAERGIC AND CHOLINERGIC SYSTEMS

Treatment (s)	Rectal Temperature (°C) at 28°C (Room Temperature)	Rectal Temperature (°C) at 40°C
Control	36.2 ± 0.14	39.0 ± 0.20*
Morphine (1 mg/kg, IP)	38.4 ± 0.25*	40.1 ± 0.23*
Naloxone (1 mg/kg, IP)	35.4 ± 0.18*	38.2 ± 0.20*
Bicuculline (1 mg/kg, IP)	37.3 ± 0.20*	40.0 ± 0.24*
Physostigmine (0.2 mg/kg, IP)	37.1 ± 0.20*	39.9 ± 0.24*
Atropine (5 mg/kg, IP)	35.5 ± 0.15*	38.1 ± 0.10*
Bicuculline + physostigmine	37.9 ± 0.20*	40.8 ± 0.26*
Bicuculline + atropine	36.5 ± 0.25*	39.0 ± 0.23*
Morphine + bicuculline	38.5 ± 0.23*	41.0 ± 0.24*
Morphine + physostigmine	38.8 ± 0.18*	40.6 ± 0.20*
Morphine + atropine	37.2 ± 0.18*	39.2 ± 0.23*
Naloxone + atropine	35.2 ± 0.15*	37.9 ± 0.15*
Morphine + bicuculline + physostigmine	38.2 ± 0.23*	41.8 ± 0.25*
Morphine + bicuculline + atropine	37.6 ± 0.20*	40.3 ± 0.23*

Results are expressed as mean ± SEM, n = 4–6. No significant change was observed in rectal temperature when control rats were treated with vehicle (0.89% saline) intraperitoneally or orally. Saline or the individual drug or their different combinations administered separately in different groups of animals were either exposed to HET (40°C) for 2 h or kept at 28°C. Doses of drugs used in combinations were the same as those used individually. In HET-exposed rats, rectal temperature was measured at the 2nd hour of HET exposure. Bicuculline and naloxone were injected 30 min and morphine 1 h before measurement of rectal temperature in rats exposed to either 28°C or 40°C.

Significance was calculated by two-way analysis of variance (ANOVA), and it was found that the interaction effect was nonsignificant ($p > 0.05$), while between the treatments the effect was significant (*$p < 0.01$).

Statistical Analysis

The statistical significance was assessed by two-way analysis of variance (ANOVA).

RESULTS

It is evident from Table 1 that exposure of the rats to 40°C for 2 h significantly increased the BT. The BT of normal (kept at 28°C) and HET-exposed rats was significantly increased with morphine (the opioid agonist; 1 mg/kg, IP) and was reduced with naloxone (the opioid antagonist; 1 mg/kg, IP). Treatment with bicuculline (GABA antagonist; 1 mg/kg, IP) or physostigmine (acetylcholinesterase inhibitor; 0.2 mg/kg, IP) or their combination significantly enhanced the BT of normal or heat-exposed rats. Atropine (acetylcholine antagonist, 5 mg/kg, IP) alone reduced the BT of normal and HET-exposed rats. Although coadministration of bicuculline with atropine did not significantly alter the BT of normal or HET-exposed rats, it significantly increased the BT of atropine- or bicuculline-treated rats. Cotreatment with morphine and atropine significantly raised the BT as compared to that of control rats and also that of atropine-treated rats either kept at 28°C or exposed to 40°C, but decreased the BT with respect to that of morphine-treated normal or HET-exposed rats. Further, it is observed from Table 1 that the BT of normal and HET-exposed rats was significantly reduced under cotreatment with naloxone and atropine. Again, administration of morphine with bicuculline significantly enhanced the BT of normal rats and of bicuculline-treated rats kept at 28°C or 40°C and also with respect to the BT of morphine-treated HET-exposed rats. Coadministration of morphine and physostigmine significantly enhanced the BT of control rats and of physostigmine-treated rats either kept at 28°C or exposed to 40°C. Table 1 also shows that physostigmine potentiated the morphine + bicuculline-induced rise in BT of HET exposed rats without affecting the BT of morphine + bicuculline-treated rats kept at 28°C. Atropine, on the other hand, significantly reduced the morphine + bicuculline-induced increase in BT of normal rats (at 28°C) or of HET-exposed rats (at 40°C).

Table 2 shows that treatment with L-dopa + carbidopa (dopamine agonist, 100 mg/kg + 10 mg/kg, PO) slightly but significantly reduced the BT of normal rats but failed to show any appreciable change in the BT of HET-exposed rats. Haloperidol (dopamine antagonist, 1 mg/kg, IP) did not significantly alter the BT of normal and HET-exposed rats.

TABLE 2

EFFECT OF ENVIRONMENTAL TEMPERATURE ON RECTAL TEMPERATURE OF RATS TREATED WITH AGONIST AND ANTAGONIST OF DOPAMINERGIC SYSTEM

Treatment (s)	Rectal Temperature (°C) at 28°C (Room Temperature)	Rectal Temperature (°C) at 40°C
Control	36.2 ± 0.14	39.0 ± 0.20
L-dopa (100 mg/kg, PO) + Carbidopa (10 mg/kg, PO)	35.7 ± 0.17	38.9 ± 0.20
Haloperidol (1 mg/kg, IP)	36.6 ± 0.19	39.2 ± 0.23

Results are expressed as mean ± SEM, n = 4-6.

L-Dopa + Carbidopa was injected 1 h and haloperidol 30 min before measurement of rectal temperature in rats kept at 28°C or exposed to 40°C.

DISCUSSION

Our previous study shows that central GABA regulates the HET-induced change in BT through the interaction with the cholinergic system (3). Because opioids play an important role in thermoregulation (1,8,26) and there is an interaction of opioids with other neurotransmitters including GABA, dopamine, and choline, the authors have studied in the present investigation the mechanism of the regulation of BT under HET-exposed conditions in relation to opioidergic-GABAergic interaction.

The results of the present study have shown that morphine (1 mg/kg, IP) an opioid receptor agonist, produces hyperthermia in normal rats kept at 28°C. This is in accord with the previous reports of Ushijima et al. (27). Morphine also increases HET-induced hyperthermia (Table 1). The opioid receptor antagonist, naloxone (1 mg/kg, IP), on the other hand, reduces the BT of normal as well as of heat-exposed rats (Table 1). Naloxone-induced hypothermia at room temperature was also reported by previous workers (13). Thus, the involvement of opioidergic neurons in thermoregulation under normal (7,8,28) and heat-exposed conditions may be suggested. Several groups of authors have reported the biphasic response of morphine on BT (11,27). Although the exact mechanism involved in the change of BT due to the exposure to high ambient temperature is still not known with certainty, several findings have suggested that the hyperthermic response of morphine is due to an upward setting of the hypothalamic set point and a consequent vesoconstriction that decreases heat loss (9). Recently, restraint stress has been reported to alter the thermoregulatory response to morphine, possibly because of the endogenous endorphin release (13). It has already been reported that HET-induced hyperthermia is associated with the increased release of β-endorphin (14,24).

The treatment with bicuculline (1 mg/kg, IP), the GABA antagonist, or with physostigmine (0.2 mg/kg, IP), the acetylcholinesterase inhibitor, significantly increases the BT of normal rats kept at room temperature. Thus, the bicuculline or physostigmine-induced hyperthermia has been shown by others (20,21). The treatment with bicuculline or with physostigmine alone or in combination (Table 1) potentiate the HET-induced hyperthermia, suggesting that HET exposure may activate the cholinergic system through the inhibition of the GABAergic activity and increases the BT. Recently, we have shown that the exposure to HET reduces the hypothalamic GABAergic activity (4). Further, a bicuculline-induced increased in BT of both normal and HET-exposed rats (Table 1) is attenuated with atropine (Table 1), suggesting that GABA mediates its action through the cholinergic system to regulate BT. The potentiation of a bicuculline-induced rise of BT in normal and HET-exposed rats following the cotreatment of morphine with bicuculline (Table 1) also suggests that morphine has some inhibitory effect on GABAergic activity. The inhibitory effect of β-endorphin on GABAergic activity has already been reported by others (21). Again, physostigmine-induced hyperthermia is further increased due to the cotreatment of physostigmine (Table 1), with morphine showing that it may activate the cholinergic system in normal and HET-exposed conditions. The present results show that the BT of normal rats and the HET-induced increase in BT are reduced

with atropine (Table 1). The atropine-induced hypothermia at room temperature has also been reported by Lomax et al. (18). Further, it is observed that cotreatment of atropine with morphine prevent the atropine-induced hypothermia and increase the BT of normal and HET-exposed rats. Cotreatment of atropine with naloxone does not appreciably affect the atropine-induced hypothermia (Table 1) under similar conditions. These may further support the above assumption that activation of the opioidergic system may activate the cholinergic system. The potentiating effect of morphine on the bicuculline + physostigmine-induced hyperthermia of rats under HET-exposed conditions (Table 1) further support the suggestion that acute exposure to HET may activate the opioidergic system, which leads to activation of the central cholinergic system through the inhibition of central GABAergic activity and enhances the BT. The correlation between the rise in BT and activation of the central cholinergic system has been previously established (3). The present study suggests that the hyperthermic response of morphine is mediated through the interaction of central GABAergic and cholinergic neuronal activity.

In order to understand the involvement of the central dopaminergic system in thermoregulation, if any, we have studied the BT of normal and HET-exposed rats treated with dopamine agonists and antagonists. It is observed that L-dopa + carbidopa slightly reduce the BT of HET-exposed rats; however, these drugs produce a slight hypothermia in normal rats (Table 2). Ferguson et al. (1984) have also shown that administration of the dopamine agonist causes hypothermia (12). But haloperidol fails to show any appreciable change in the BT of normal and heat-exposed rats. Recently, Biswas and Poddar (3) have shown that the central dopaminergic system is not directly related to the GABA-mediated cholinergic regulation of HET-induced enhancement of BT. Thus, although dopamine content in some brain areas may be affected due to HET exposure of the animal (23), the involvement of dopamine receptors in brain mechanisms regulating the opioid-induced change in BT (28) is unlikely.

Finally, it may be concluded that HET-induced activation of the opioidergic system disinhibits the inhibitory effect of GABA on the cholinergic neuron and enhances the cholinergic activity and, hence, raises the BT under HET exposure.

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