ELSEVIER

Behavioural Brain Research 134 (2002) 75–82

BEHAVIOURAL BRAIN RESEARCH

www.elsevier.com/locate/bbr

Research report

Yawning/cortical activation induced by microinjection of histamine into the paraventricular nucleus of the rat

Yoshinari Seki, Ikuko Sato-Suzuki, Ichiro Kita, Mitsugu Oguri, Hideho Arita *

Department of Physiology, Toho University School of Medicine, 5-21-16 Omori-nishi, Ohta-ku, Tokyo 143-8540, Japan

Received 6 September 2001; received in revised form 15 November 2001; accepted 15 November 2001

Abstract

The effects of microinjection of histamine into the paraventricular nucleus (PVN) of the hypothalamus on yawning responses were investigated in anesthetized, spontaneously breathing rats. Yawning responses were evaluated by monitoring the intercostal electromyogram (EMG) as an index of inspiratory activity and digastric EMG as an indicator of mouth opening. We also recorded the electrocorticogram (ECoG) to determine the arousal response during yawning. Autonomic function was evaluated by measuring blood pressure and heart rate. Microinjection of histamine into the medial parvocellular subdivision (*mp*) of the PVN elicited a yawning response, i.e. a single large inspiration with mouth opening, and an arousal shift in ECoG to lower voltage and faster rhythms. Microinjection of HTMT dimaleate, an H1 receptor agonist, into the PVN also caused the yawning/arousal response. Pretreatment with pyrilamine, an H1 receptor antagonist, inhibited the histamine induced yawning behavior. These data demonstrate that a histamine receptive site for triggering yawning/arousal responses exists in the PVN, and suggest that these responses are mediated by activation of H1 receptor within the PVN.

Keywords: Yawning; Arousal; Paraventricular nucleus; Histamine

1. Introduction

We have reported that a stereotyped yawning response can be evoked by several chemical stimulations of the paraventricular nucleus (PVN) in anesthetized, spontaneously breathing rats [16,28,29]. A typical yawning response is induced by microinjection of L-glutamate in the medial parvocellular subdivision (*mp*) of the PVN [16], and is characterized by an initial depressor response and an arousal shift in electrocorticogram (ECoG) followed by a single large inspiration with mouth opening. Our concept that the PVN is essential for the occurrence of yawning is compatible with previous data of Argiolas and Melis [2,3,20] who demonstrated that lesions of the PVN prevents yawning induced by apomorphine and oxytocin in freely moving rats. On the other hand, two physiological roles of the PVN concerning yawning have been newly suggested by our recent findings. (1) An oxygen sensor may exist within the PVN and yawning may be an arousal behavior caused by higher brain ischemia, since microinjection of cyanide into the *mp* caused the stereotyped yawning response [16]. (2) The PVN may play an important role in triggering arousal mechanisms, since microinjection of orexin-A, a neuropeptide involved in sleep and arousal mechanisms, into the *mp* caused the stereotyped yawning response [28].

Yawning is one typical symptom of motion sickness [10,22]. The involvement of histamine in neural processes of motion sickness is suggested by several pieces of evidence [31,32]. In animal models of motion sickness, the histamine level in the hypothalamus increases after rotation [31]. Another piece of evidence comes from Takeda et al.'s observation that administration of α-fluoromethylhistidine, an inhibitor of histamine synthesizing enzyme, suppresses the signal of motion sick-

* Corresponding author. Tel.: +81-3-3762-4151; fax: +81-3-3762-8148

E-mail address: aritah@med.toho-u.ac.jp (H. Arita).

0166-4328/02/$ - see front matter
PII: S0166-4328(01)00454-5

ness in rats [31]. It is also clinically well known that antihistaminics are effective in preventing motion sickness in humans. These data on motion sickness led us to speculate that the yawning behavior might also be affected by histaminergic neural transmission.

The neurons responsible for yawning are proved to be the oxytocinergic parvocellular neurons in the PVN projecting to the lower brain stem [28,29]. On the other hand, oxytocinergic parvocellular neurons in the PVN are reported to be activated by histamine [17]. These two data together support our idea that yawning might be affected by histamine.

The present study focused on the yawning response induced by microinjection of histamine into the PVN. Attempts were further made to identify whether the H1 receptor in the PVN mediates this response.

2. Materials and methods

Experiments were performed on 14 male Wistar rats weighing 300–450 g. The rats were anesthetized with 50 mg/kg pentobarbital sodium intraperitoneally, and additional doses were given as needed. All experiments were approved by the Animal Experimentation Ethics Committee of Toho University School of Medicine. Every effort was made to minimize animal suffering and the number of animals used. Surgical procedures were essentially the same as described previously [16,28,29]. In brief, catheters were placed in the femoral artery to monitor arterial blood pressure (BP). Heart rate (HR) was measured from the BP pulse with a tachometer (AT-601G, Nihon Kohden, Japan). To monitor inspiratory activity, a pair of twisted wire electrodes, insulated except for 1 mm at the tips, were implanted into the lower intercostal space, by way of a 23-gauge hypodermic needle; the needle was then withdrawn, leaving the wires in the intercostal muscle. Similarly, pairs of wire electrodes were implanted in the digastric muscle to monitor mouth opening activity. For ECoG recordings, holes were drilled in the skull and two screw electrodes were implanted. ECoG signals were amplified and filtered (0.1–50.0 Hz bandpass). These polygraphic signals were all stored in a DAT data recorder (PC208Ax, Sony, Japan) for further analysis. Rectal temperature was maintained at 37 °C with a heating lamp.

The animal was fixed prone in a stereotaxic frame and a parietal craniotomy was performed. For drug microinjections into the PVN of the hypothalamus, we constructed a four-barrel glass micropipette, which was connected to a fine cannula. The free end of the cannula was attached to a picopump (PV830 Pneumatic, WPI) for injections. The volumes injected were monitored under a dissecting microscope and measured from the movement of the fluid meniscus. The barrels were filled with either 0.1 M L-glutamate, 0.1–1 M histamine, 5–50 mM HTMT dimaleate (Biomol Res Lab), 10 mM pyrilamine (Sigma), 0.1–1 M dimaprit 2HCl (Biomol Res Lab) or 5 mM ranitidine hydrochloride (Sigma). In all cases, the drug doses reported are for the base. Drugs were freshly dissolved in artificial cerebrospinal fluid (ACSF: 147 mM NaCl, 3 mM $NaHCO_3$, 3.5 mM KCl, 1.3 mM $CaCl_2$, 1.1 mM $MgCl_2$, 0.5 mM NaH_2PO_4 and 1.1 mM urea; pH 7.30–7.35), except for HTMT dimaleate, which was dissolved in a drop of ethanol and then diluted with ACSF. As a control for L-glutamate, histamine, pyrilamine, dimaprit 2HCl and ranitidine hydrochloride, the same amount of ACSF was injected in responsive sites. As a control for HTMT dimaleate, the same amount of ethanol in ACSF was injected in responsive sites. The chemicals were obtained from Wako, Japan.

The dura overlying the cortex was opened for advancement of the micropipette, which was inserted into an area 0.2–0.3 mm lateral to the midline, 1.1–2.4 mm posterior to the bregma and 5.8–7.2 mm vertical from the parietal dura, according to the Paxinos and Watson [26] atlas of the rat brain. We made a systematic search for the sites from which yawning responses were evoked by microinjection of L-glutamate (10–100 nl). After exploratory tracking with L-glutamate, we identified sites where maximal yawning responses were obtained. A small volume (10–100 nl) of histamine, pyrilamine, HTMT dimaleate, dimaprit 2HCl or ranitidine hydrochloride solution was then slowly injected into the same responsive site to determine whether a yawning response could be induced. We examined the effects of various kinds of injection in 5–11 sites (one site per rat). Response to each injection was examined repeatedly, at least three times in the same condition. On completion of the experiment, successful stimulation sites were marked with 0.5 M acetate-pontamine for histological examination. The rat was then deeply anesthetized with pentobarbital sodium (100 mg/kg) to remove the brain.

After the experiments, ECoG signals were displayed on an oscilloscope together with an intercostal electromyogram (EMG). For power spectrum analysis the ECoG signal was digitized at a sampling rate of 1024 Hz and subjected to fast Fourier transform. Analyses were performed on data sampled during pre-injection (control), early yawning and late yawning (recovery) periods of each response. The early yawning period corresponded to the response prior to the final yawning event (a single large inspiration). The late yawning (recovery) period was 5–125 s until ECoG activity returned to pre-injection activity levels.

3. Results

We made a systematic search for yawning-responsive sites in and around the PVN by microinjection of L-glutamate. Microinjection of L-glutamate in the *mp* of the PVN elicited a stereotyped yawning response. We confirmed that no other sites in and around the PVN could elicit this stereotyped response. A typical example of a stereotyped yawning response induced by L-glutamate was essentially the same as in our previous study [16,28,29]. Namely, the response was characterized by an initial depressor response followed by a final yawning event (a single large inspiratory effort). Power spectral analysis of ECoG waves revealed that the very slow waves (< 3 Hz) during the control period before chemical stimulation, were replaced by lower voltage and faster rhythms (4–8 Hz, data not shown).

We next examined the effect of microinjection of histamine in the specific region with the stereotyped yawning. Fig. 1 shows a typical example of the effect of histamine injection into the site where the stereotyped yawning response was elicited by L-glutamate injection. Histamine injection (10 nmol) qualitatively caused similar sequential events of a stereotyped yawning response. The initial response represented an increase in HR and an arousal shift in the ECoG to lower voltage, as precisely described in the next paragraph. EMG_{JAW} showed quite rhythmical activity. These early yawning responses were followed by the development of a single large inspiration. Compared with L-glutamate, a decrease in respiratory rate was seen after the final yawning behavior. Regarding BP change, we noticed that a pressor response which lasted for ~ 90 s appeared after the yawning behavior.

Power spectral analysis of ECoG waves revealed that an early change in ECoG occurred prior to a yawn, as shown in Fig. 2. The high voltage and very slow waves during the control period before chemical stimulation (< 3 Hz, Fig. 2, control), were replaced by faster rhythms concurrent with the cardiovascular and EMG_{JAW} responses (4–5 Hz, Fig. 2, early). The component of faster rhythms remained after the final yawning behavior (Fig. 2, late, 1; late, 2), and returned gradually to control levels (Fig. 2, late 3; late 4). We examined the effects of histamine injection in eleven sites (one site per rat) and found that histamine injection caused the yawning/arousal response in all sites examined.

We further evaluated the effect of H1 receptor agonist HTMT dimaleate injection in the specific regions where the stereotyped yawning responses were elicited by both L-glutamate and histamine injections, as shown in a typical example in Fig. 3. HTMT dimaleate injection caused a single large inspiration with a shorter delay of ~ 15 s. It was also noticed that the fall in BP was weak or negligible. An arousal shift in the ECoG to lower voltage and faster rhythms compared with the control was observed after HTMT dimaleate injection (~ 6 Hz, Fig. 4, early). The component of faster rhythms remained after the final yawning behavior (Fig. 4, late, 1; late, 2), and returned gradually to

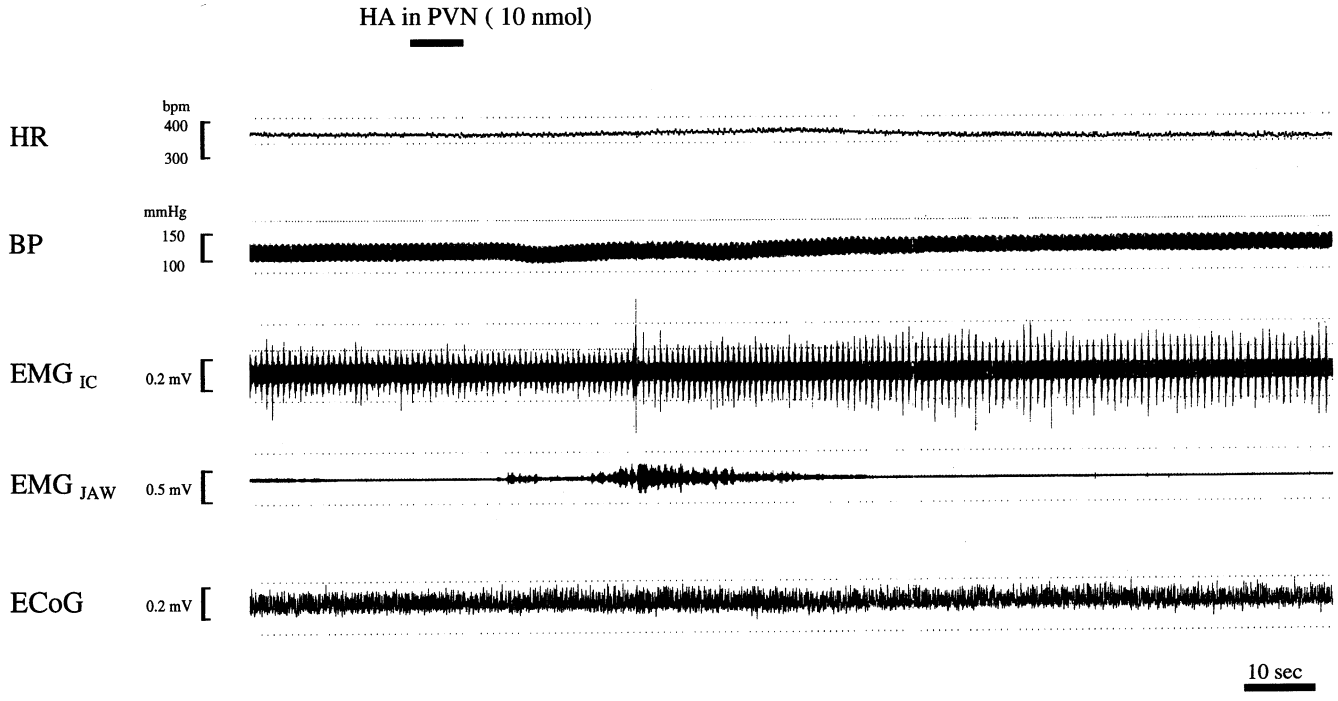

Fig. 1. Representative yawning responses evoked by microinjection of histamine (HA, 10 nmol) into the PVN. BP, blood pressure; HR, heart rate; EMC_{IC}, intercostal EMG; EMC_{JAW}, digastric EMG; ECoG, cortical electroencephalograph.

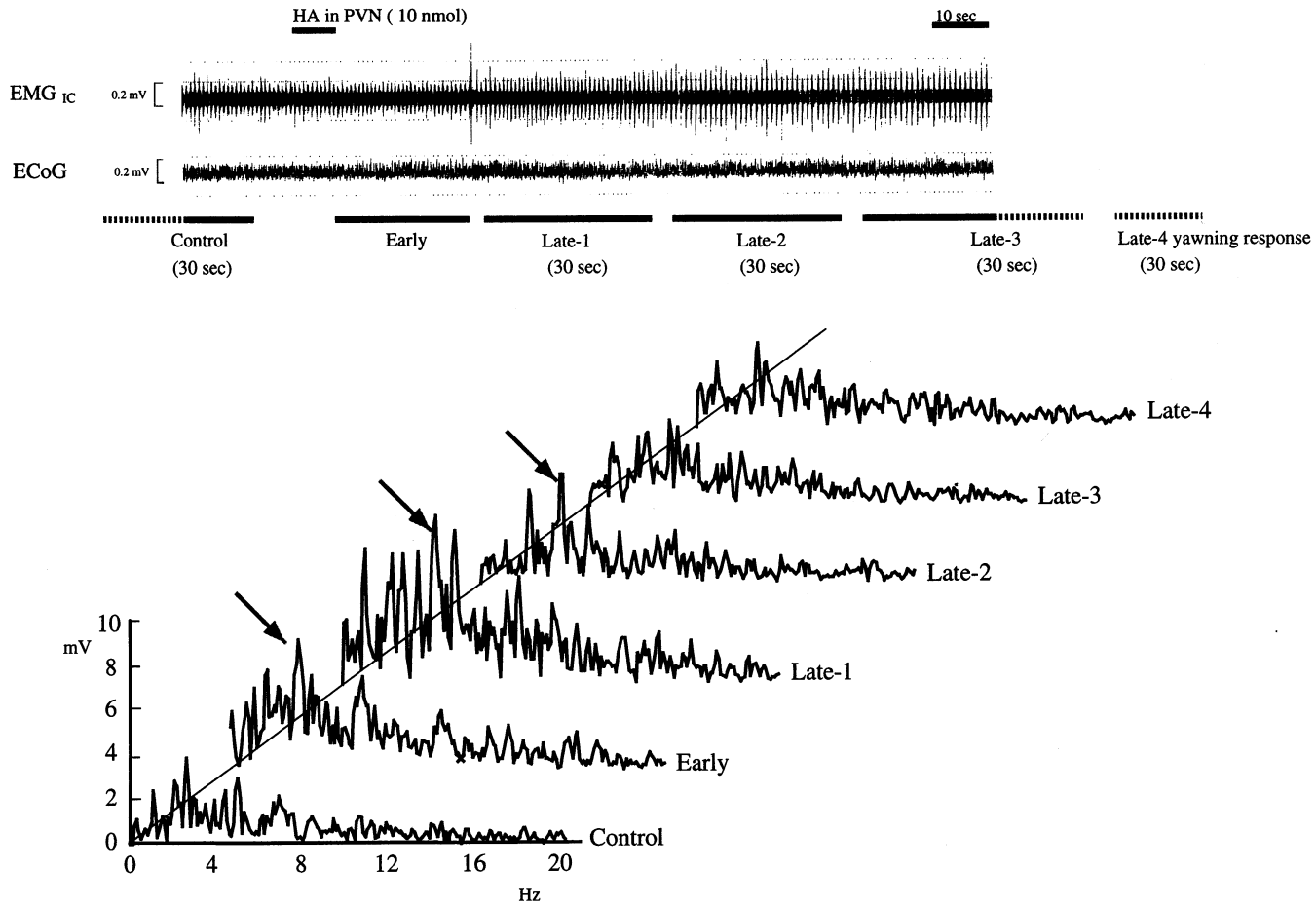

Fig. 2. Typical changes in the cortical electroencephalograph recording during yawning induced by microinjection of histamine (10 nmol) into the PVN. The very slow waves (< 3 Hz) during the control period before chemical stimulation, were replaced by faster rhythms (4–5 Hz, indicated by arrows) after injection of histamine into the PVN.

control levels (Fig. 4, late, 3). Similar responses to HTMT dimaleate injection were obtained from all sites examined (five sites from five rats).

Afterthat, we examined the effect of H1 receptor antagonist pyrilamine on yawning/arousal responses evoked by microinjection of histamine into the PVN. Microinjection of pyrilamine into the PVN did not induce either the yawning or the arousal response. Furthermore, pretreatment with pyrilamine injected into the PVN completely blocked the yawning/arousal response induced by histamine.

As for H2 receptor agonist dimaprit 2HCl, an increase in HR was prominent after injection into the PVN, however, the yawning response was not reproducibly induced compared with that by either histamine or H1 agonist. Similar responses to dimaprit 2HCl injection were obtained from all sites examined (five sites from five rats). We also observed that pretreatment with H2 receptor antagonist ranitidine hydrochloride into the PVN did not block the yawning/arousal response induced by histamine.

Light microscopic examination revealed that both histamine and HTMT dimaleate responsive sites were located in the medial part of the rostral PVN (Fig. 5). No responsive sites for the yawning/arousal response were identified in the lateral part of the rostral PVN, in the caudal PVN, or regions around the PVN.

4. Discussion

We found that the stereotyped yawning response was activated by local application of histamine into the PVN, which indicates that the PVN is a region receiving histaminergic inputs from the tuberomammillary nucleus (TM) of the posterior hypothalamus, a region where histaminergic neurons are densely located [7]. Our view that the PVN is a histamine sensitive site is supported by three pieces of evidence. Panula et al. demonstrated that histaminergic neurons originating in the TM project to other brain areas including the PVN [25]. Another piece of evidence comes from an autoradiographic study showing the

presence of histamine receptor in the PVN [24]. Finally, Bealer and Abell [5] caused effective cardiovascular responses by applying histamine through microdialysis probes into the PVN.

Our data showing that histamine administered into the PVN induced yawning, together with an arousal shift in the ECoG, confirms the results of previous studies which showed that histamine is an important neurotransmitter in arousal regulation [7]. Experimental lesioning of the posterior hypothalamus has been reported to induce a state of somnolence in rats [21]. In addition, the decrease in histamine content of the brain suppresses wakefulness in rats [12]. Conversely, histaminergic drugs cause wakefulness in rats [13]. These arousal effects of histaminergic systems are thought to be mediated by H1 receptors [19]. It is also clinically well known that antihistaminics induce drowsiness. Moreover, particularly high levels of histamine receptors are present in area involved in arousal, i.e. the cerebral cortex, cholinergic cell groups in the mesopontine tegmentum (MPT) and in the basal forebrain, as well as the locus coeruleus (LC) [7,25].

We further indicate that not only histamine but also PVN is involved in the arousal signaling pathway, since microinjection of histamine into the PVN induced the arousal shift in the ECoG together with the yawning response. It should be emphasized that no other area in or around the PVN elicited the arousal/yawning response. These data may strengthen the hypothesis proposed in our previous study, that the PVN is a site involved in the arousal mechanism [28].

According to the arousal pathway referred to as histaminergic transmission, several possible pathways are nominated. Arousal may be due to histaminergic neurons originating in the TM directly projecting to the cerebral cortex [27]. On the other hand, it may be due to histaminergic neurons projecting to the cholinergic cell groups in the MPT [18] or in the basal forebrain [8,15] as well as the LC [25]. Here we suggest another signaling pathway of the arousal mechanism via the PVN. Namely, histaminergic neurons from the TM may directly activate parvocellular neurons in the PVN, which in turn project to the basal forebrain or the LC (Fig. 6).

We previously reported that the neurons responsible for yawning might be the oxytocinergic parvocellular neurons in the PVN projecting to the lower brain stem

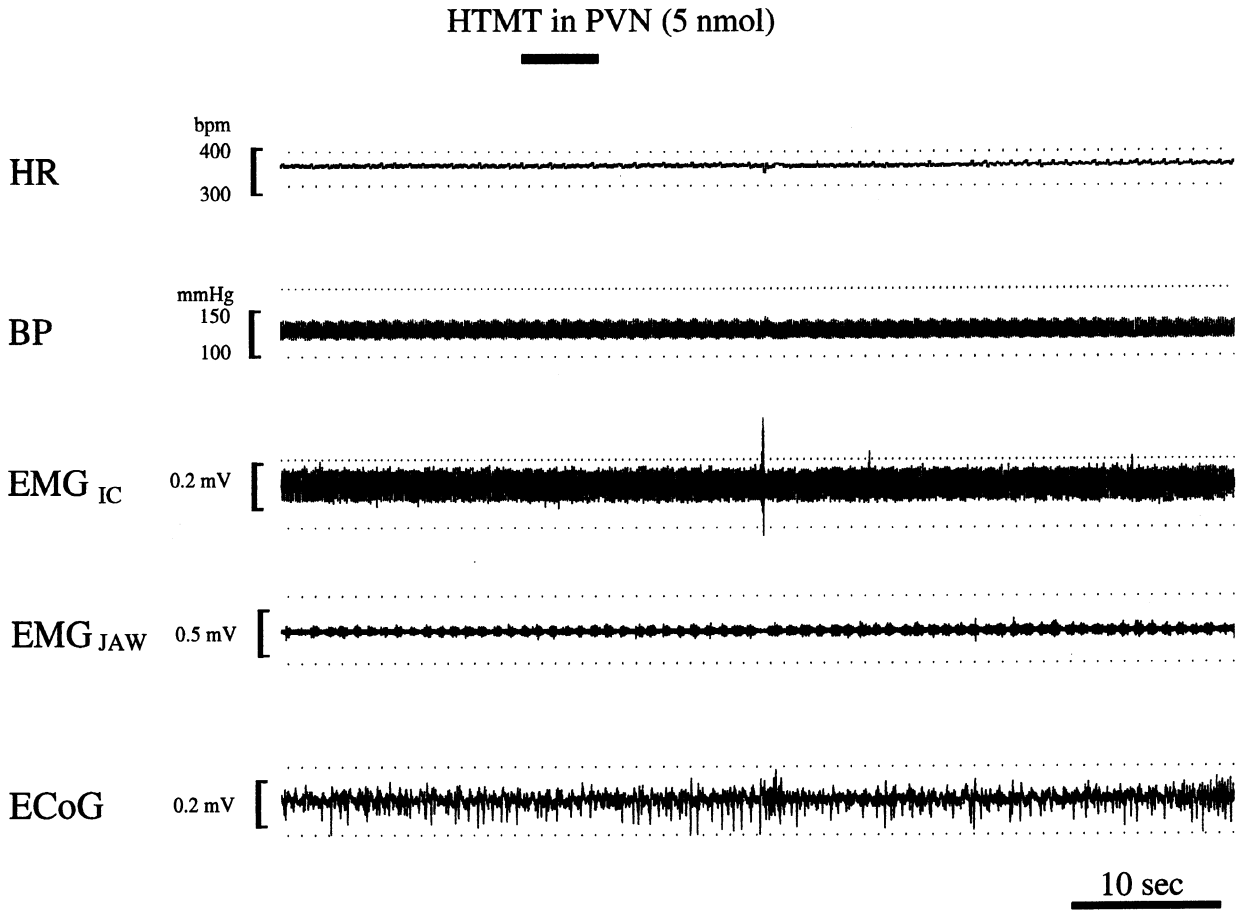

Fig. 3. Representative yawning responses evoked by microinjection of HTMT dimaleate (HTMT, 5 nmol) into the PVN. Abbreviations as in Fig. 1.

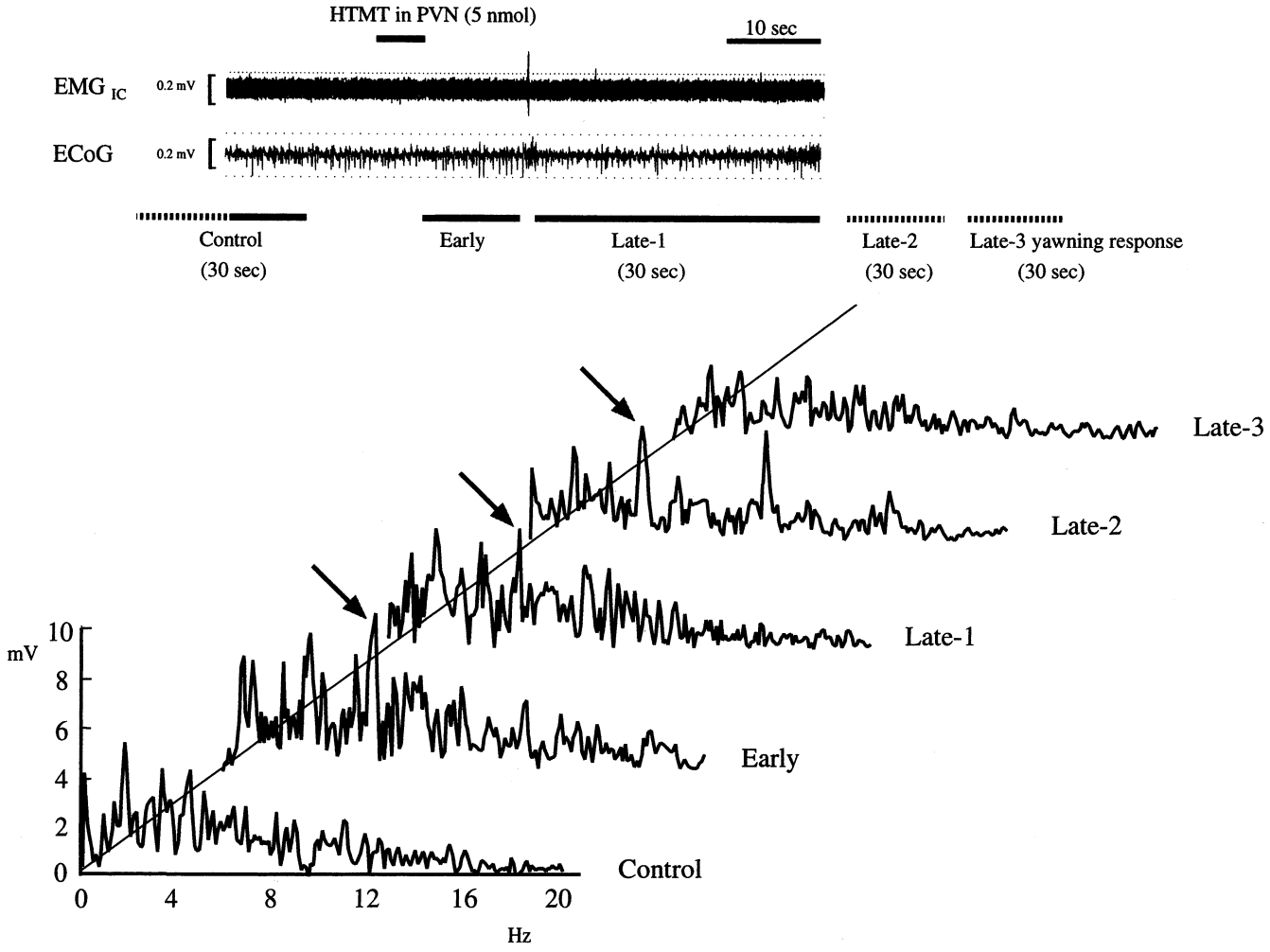

Fig. 4. Typical changes in the cortical electroencephalograph recording during yawning induced by microinjection of HTMT dimaleate (5 nmol) into the PVN. The very slow waves (< 2 Hz) during the control period before chemical stimulation, were replaced by faster rhythms (4–6 Hz, indicated by arrows) after injection of HTMT dimaleate into the PVN.

[28,29]. Our suggestion was mainly based on the report of Sawchenko and Swanson [30] who demonstrated that oxytocinergic parvocellular neurons in the PVN send descending axons to the lower brain stem which region is involved in arousal, respiratory, cardiovascular, and other autonomic functions. According to this notion, the present data can be explained by the possibility that histamine activates the oxytocinergic parvocellular neurons in the PVN projecting to the lower brain stem. This possibility is strongly supported by the data of Kjær et al. [17] who reported that c-fos expression in oxytocinergic parvocellular neurons within the PVN is induced by central administration of histamine. They further suggested that histamine might be indirectly involved in the regulation of neurons in the lower brain stem and the spinal cord via the PVN.

Although we suggest that the oxytocinergic parvocellular neurons in the PVN are involved in yawning, we can not exclude the possibility that the PVN may contain more than one pathway mediating this response. For instance, ACTH injected into the PVN and surrounding periventricular region induces yawning that is not prevented by oxytocin antagonist [1]. Corti-

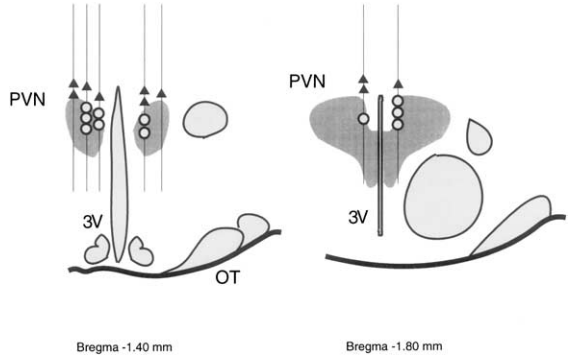

Fig. 5. Schematic drawings of transverse sections of the PVN, showing effective (○) or ineffective (▲) injection sites where yawning / arousal responses were obtained by histamine. OT, optic tract; 3V, third ventricle.

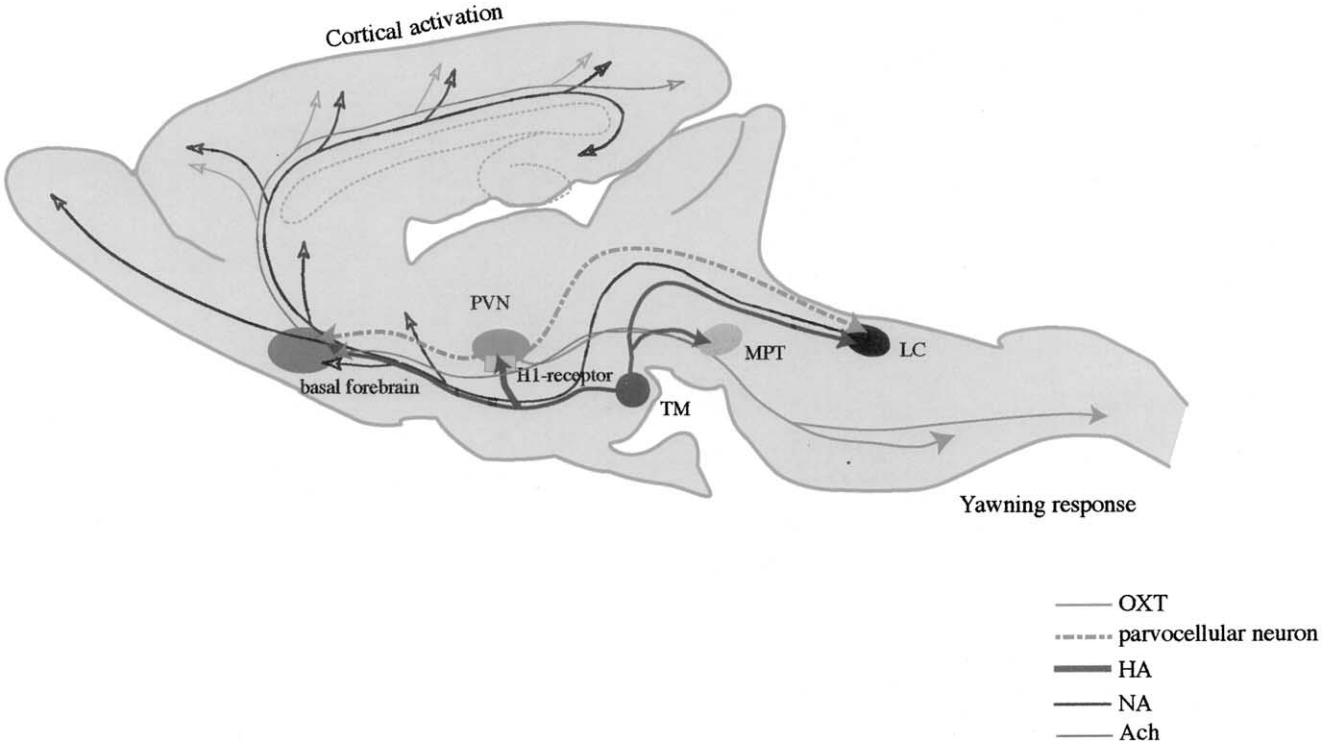

Fig. 6. Schematic representation of our model on yawning/arousal responses mediated through the PVN. Histamine neurons from the TM nucleus may directly activate neurons in the PVN, which in turn project to the LC or the basal forebrain. Oxytocinergic neurons from the PVN project to the lower brainstem, including the facial nucleus and respiratory related neurons which are implicated in the yawning response. OXT, oxytocin; HA, histamine; NA, noradrenaline; Ach, acetylcholine; MPT, mesopontine tegmentum.

cotropin releasing factor (CRF) neurons within the PVN could be responsible for the ACTH-induced yawning, however, there is little evidence that CRF neurons project to the lower brain stem which region is involved in arousal, respiratory, cardiovascular, and other autonomic functions regarded to yawning.

We indicate that histamine activates the H1 receptor in the PVN, since the yawning response induced by histamine was inhibited by pretreatment with pyrilamine, an H1 receptor antagonist. Moreover, microinjection of HTMT dimaleate, an H1 receptor agonist, into the PVN, evoked the yawning response. Similarly to our data, Gower et al. [11] reported that H1 receptor antagonist injected subcutaneously into conscious rats inhibited the apomorphine-induced yawning response. With regard to this view, an autoradiographic study revealed the presence of H1 receptor in the PVN [24]. The existence of H1 receptor in the PVN has also been examined in functional studies. For example, Bealer demonstrated that histamine evokes norepinephrine [6] as well as oxytocin [4] release by activation of H1 receptors in the PVN. H1 receptors in the PVN are also reported to be involved in histaminergic suppression of food intake [23].

Although H1 receptor may be important in the modification of the yawning/arousal response [33], the role of other types of histamine receptors cannot be fully eliminated. In this connection, Ferrari and Baggio [9] reported that H2 receptor antagonist dose-dependently antagonized the yawning and penile erection induced by apomorphine. In line with their data, yawning response was sometimes induced by H2 receptor agonist into the PVN in the present study. However, H2 receptor agonist induced a yawning response with a much longer onset latency compared with that of histamine or H1 agonist. In addition, pretreatment with H2 receptor antagonist did not block the yawning response induced by histamine in our study. Further study is required before the precise mechanism of histamine actions on yawning/arousal responses can be identified.

We noticed that a pressor response appeared after the yawning behavior induced by microinjection of histamine into the PVN, which lasted over ~ 90 s. In this connection, Bealer and Abell [5] demonstrated that administration of histamine through microdialysis probes in the PVN increased BP in conscious rats. It is suggested that the PVN is a brain site where histaminergic and noradrenergic systems interact to regulate BP, and the pressor response evoked by histamine may be due to adrenoreceptor stimulation of vasopressin release [5]. Although the increase in HR was prominent after injection of H2 agonist into the PVN (data not shown), we could not observe a clear increase in HR by histamine injection into the PVN, which result differs

from that in conscious animals [5]. The lack of changes in HR by histamine in this study might be attributed to the possibility of anesthetics used, since anesthesia is well known to affect the cardiovascular and autonomic responses [14].

Since yawning is one typical symptom of motion sickness [10,22], a disorder which is modified by histaminergic transmission, the present results can be explained by the notion that yawning is triggered by motion sickness which signaling pathway is mediated by histaminergic neurons [31,32]. According to our data showing that yawning was inhibited by H1 receptor antagonist, together with previous data showing that motion sickness is suppressed by H1 antagonist [31], it is likely that histamine induces yawning as well as motion sickness through H1 receptor within the PVN.

References

[1] Argiolas A, Melis MR, Murgia S, Schiöth HB. ACTH- and α-MSH-induced grooming, stretching, yawning and penile erection in male rats: site of action in the brain and role of melanocortin receptors. Brain Res Bull 2000;51:425–31.

[2] Argiolas A, Melis MR. The neuropharmacology of yawning. Eur J Pharmacol 1998;343:1–16.

[3] Argiolas A, Melis MR, Mauri A, Gessa GL. Paraventricular nucleus lesion prevents yawning and penile erection induced by apomorphine and oxytocin but not by ACTH in rats. Brain Res 1987;421:349–52.

[4] Bealer SL, Crowley WR. Stimulation of central and systemic oxytocin release by histamine in the paraventricular hypothalamic nucleus: evidence for an interaction with norepinephrine. Endocrinology 1999;140:1158–64.

[5] Bealer SL, Abell SO. Paraventricular nucleus histamine increases blood pressure by adrenoreceptor stimulation of vasopressin release. Am J Physiol 1995;269:H80–5.

[6] Bealer SL. Histamine releases norepinephrine in the paraventricular nucleus/anterior hypothalamus of the conscious rat. J Pharmacol Exp Ther 1992;264:734–8.

[7] Brown RE, Stevens DR, Haas HL. The physiology of brain histamine. Prog Neurobiol 2001;63:637–72.

[8] Cecchi M, Passani M, Bacciottini L, Mannaioni PF, Blandina P. Cortical acetylcholine release elicited by stimulation of histamine H1 receptors in the nucleus basalis magnocellularis: a dual-probe microdialysis study in the freely moving rat. Eur J Neurosci 2001;13:68–78.

[9] Ferrari F, Baggio G. Influence of cimetidine, ranitidine and imidazole on the behavioral effects of (±) *N*-*n*-propylnorapomorphine in male rats. Psychopharmacology 1985;85:197–200.

[10] Graybiel A, Knepton J. Sopite syndrome: a sometimes sole manifestation of motion sickness. Aviat Space Environ Med 1976;47:873–82.

[11] Gower AJ, Berendsen HHG, Broekkamp CL. Antagonism of drug-induced yawning and penile erections in rats. Eur J Pharmacol 1986;122:239–44.

[12] Itowi N, Yamatodani A, Kiyono S, Hiraiwa ML, Wada H. Effect of histamine depletion on the circadian amplitude of the sleep–wakefulness cycle. Physiol Behav 1991;49:643–6.

[13] Kalivas PW. Histamine-induced arousal in the conscious and pentobarbital-pretreated rat. J Pharmacol Exp Ther 1982;222:37–42.

[14] Kannan H, Hayashida Y, Yamashita H. Increase in sympathetic outflow by paraventricular nucleus stimulation in awake rats. Am J Physiol 1989;256:R1325–30.

[15] Khateb A, Fort P, Pegna A, Jones BE, Mühlethaler M. Cholinergic nucleus basalis neurons are excited by histamine in vitro. Neuroscience 1995;69:495–506.

[16] Kita I, Sato-Suzuki I, Oguri M, Arita H. Yawning responses induced by local hypoxia in the paraventricular nucleus of the rat. Behav Brain Res 2001;117:119–26.

[17] Kjær A, Larsen PJ, Knigge U, Møller M, Warberg J. Histamine stimulates c-fos expression in hypothalamic vasopressin-, oxytocin-, and corticotropin-releasing hormone-containing neurons. Endocrinology 1994;134:482–91.

[18] Lin JS, Hou Y, Sakai K, Jouvet M. Histaminergic descending inputs to the mesopontine tegmentum and their role in the control of cortical activation and wakefulness in the cat. J Neurosci 1996;16:1523–37.

[19] Lin JS, Sakai K, Jouvet M. Evidence for histaminergic arousal mechanisms in the hypothalamus of cat. Neuropharmacology 1988;27:111–22.

[20] Melis MR, Argiolas A, Gessa GL. Apomorphine-induced penile erection and yawning: site of action in brain. Brain Res 1987;415:98–104.

[21] Nauta WJH. Hypothalamic regulation of sleep in rats. An experimental study. J Neurophysiol 1946;9:285–316.

[22] Noel P, Norris C. Motion sickness. J La State Med Soc 1996;148:7–11.

[23] Ookuma K, Yoshimatsu H, Sakata T, Fujimoto K, Fukagawa K. Hypothalamic sites of neuronal histamine action on food intake by rats. Brain Res 1989;490:268–75.

[24] Palacios LM, Wamsley JK, Kuhar MJ. The distribution of histamine H1-receptors in the rat brain: an autoradiographic study. Neuroscience 1981;6:15–37.

[25] Panula P, Pirvola U, Auvinen S, Airaksinen S. Histamine-immunoreactive nerve fibers in the rat brain. Neuroscience 1989;28:585–610.

[26] Paxinos G, Watson C. The Rat Brain in Stereotaxic Coordinates, second ed. San Diego, CA: Academic Press, 1986.

[27] Reiner PB, Kamondi A. Mechanisms of antihistamine-induced sedation in the human brain: H1 receptor activation reduces a back-ground leakage potassium current. Neuroscience 1994;59:579–88.

[28] Sato-Suzuki I, Kita I, Seki Y, Oguri M, Arita H. Cortical arousal induced by microinjection of orexins into the paraventricular nucleus of the rat, Behav Brain Res, in press.

[29] Sato-Suzuki I, Kita I, Oguri M, Arita H. Stereotyped yawning responses induced by electrical and chemical stimulation of paraventricular nucleus of the rat. J Neurophysiol 1998;80:2765–75.

[30] Sawchenko PE, Swanson LW. Immunohistochemical identification of neurons in the paraventricular nucleus of the hypothalamus that project to the medulla or to the spinal cord in the rat. J Comp Neurol 1982;205:260–72.

[31] Takeda N, Morita M, Kubo T, Yamatodani A, Watanabe T, Wada H, Matsunaga T. Histaminergic mechanism of motion sickness: neurochemical and neuropharmacological studies in rats. Acta Otolaryngol 1986;101:416–21.

[32] Takeda N, Morita M, Hasegawa S, Kubo T, Matsunaga T. Neurochemical mechanisms of motion sickness. Am J Otolaryngol 1989;10:351–9.

[33] Yanai K, Okamura N, Tagawa M, Itoh M, Watanabe T. New findings in pharmacological effects induced by antihistamines: from PET studies to knock-out mice. Clin Exp Allergy 1999;29:29–36.