博 士 論 文

Effects of area postrema lesions and bilateral subdiaphragmatic afferent vagotomy on emetine-induced conditioned taste avoidance in rats

（ラットにおけるエメチン誘発条件付け味覚忌避に対する最後野破壊および横隔膜下両側迷走神経求心路切除の効果）

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Effects of area postrema lesions and bilateral subdiaphragmatic afferent vagotomy on emetine-induced conditioned taste avoidance in rats.

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ABSTRACT

We investigated the effect of area postrema lesions and selective vagotomy of afferent fibers on emetine-induced nausea in rats. We evaluated the acquisition of the conditioned taste avoidance (CTA) to 0.1% saccharin solution after conditioning with emetine dihydrochloride (5.54 mg/kg, i.p., 1% BW). The CTA was measured in three groups of rats: a bilateral subdiaphragmatic afferent vagotomy group, an area postrema lesion group, and a sham lesion group. The bilateral vagotomy and sham groups of rats showed acquisition of CTA within 2 days of the test date. Taste avoidance was never conditioned in the area postrema lesion group. These results indicate that the area postrema plays a crucial role in the induction of emetine-induced nausea.

1. Introduction

Emetine is an alkaloid contained in the root of Cephaelis ipecacuanha that has been used as an emetic agent. Ipecac syrup was previously produced as a household emetic agent for emergency treatment of infant accidental ingestion of poisonous or hazardous materials [1]. Previous studies on the emetic effects of emetine have suggested both central and peripheral mechanisms for emetine action. In a study using intraventricular infusion of emetine hydrochloride (0.2 mg/kg) into the cerebral ventricles of dogs, investigators concluded that vomiting resulted from area postrema activation [2]. By contrast, oral administration of emetine in ferrets increased the activity of visceral afferent vagal nerve and 5-HT concentration in the intestinal tract, but not in the area postrema, and it was concluded that the mechanism was peripheral, mediated by the 5-HT_4 receptors [3]. Our previous study showed that intraperitoneal administration of emetine increased the activity of neurons in the area postrema and the nucleus tractus solitarius (NTS) by using immunohistochemical detection of Fos protein-expressing cells [4].

Reynolds et al. (1991) have reported that administration of cisplatin, an anticancer drug, induced cells expressing Fos protein in the area postrema and the NTS in ferrets [5]. The number of cisplatin-induced Fos protein-expressing cells in the NTS were significantly reduced by unilateral cervical vagotomy, but not in the area postrema. They have also reported that the 5-HT antagonist granisetron blocked cisplatin-induced vomiting, but it did not block the expression of c-Fos protein in the area postrema, suggesting that c-Fos-like immunoreactivity in the area postrema may depend on neither vagal nerve activity nor the activation of receptors that can be blocked by granisetron.

Fos protein-expressing cells are induced by LiCl, which is widely used as an unconditioned stimulus to induce malaise in the conditioned taste aversion experiment, in the central amygdaloid nucleus, external lateral subnucleus of the parabrachial nucleus, posteromedial and commissural parts of the NTS and the area postrema of rats [6]. Recently, we reported that inhaled methyl methacrylate, a major component of some popular dental resin, induced Fos-expressing neurons in the area postrema [7].

The area postrema, which is present at the caudal end of the fourth ventricle, has been implicated as an important central structure involved in the autonomic regulation of food intake [8–10], body fluid balance [11, 12], and cardiovascular system [13]. The area postrema is also well-known as the Chemoreceptor Trigger Zone for emesis in the animals that can vomit, such as cats, dogs [14] and ferrets [15]. Such features could allow area postrema neurons to respond to circulating emetogenic chemical substances [16, 17].

The NTS is the primary relay nucleus of the afferent vagal nerve, which is the important pathway for detecting emetic signals from the gastrointestinal tract [18]. Electrical stimulation of abdominal vagal afferents can induce emesis [18]. Abdominal vagotomy suppresses cisplatin-induced emesis [19–21] and copper sulfate-induced emesis [22]. Previous studies have shown reciprocal neuron projections

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between the area postrema and the NTS [23, 24].

The area postrema has the role of an interoceptor for circulating chemical substances and a role of relaying the vagal afferent inputs, and it is possible that these neural pathways are involved in inducing nausea, however, the details are still unclear. Therefore, to clarify the mechanism of emetine-induced nausea, we conducted an experiment using rats treated with area postrema lesions or subdiaphragmatic vagotomy. The aim of this study is to clarify the more detailed mechanism of emetine-induced nausea.

2. Methods

All experiments in the present study were conducted in compliance with the Hokkaido University Animal Experiment Guidelines. Twenty-five male Sprague–Dawley rats (20 rats with surgery and 5 intact rats, body weight 200–230 g at the start of an experiment) were used as experimental animals. Rats were kept in individual cages and fed freely on a 12 h light/dark cycle. Water deprivation period was set according to the following protocol of the experiment. The rats with surgery were free to feed and water for one week after the operation, and then experiment of conditioned taste avoidance (CTA) was started.

2.1. Measurement of CTA for evaluation of emetine-induced nausea

CTA was performed as described in our previous studies [4, 25, 26]. Fig. 1 shows the protocol for our CTA analysis. In this experiment, rats were given access to a sweet solution (0.1% saccharin sodium hydrate dissolved in distilled water, a conditioned stimulus) for 20 min and then rats were injected with emetine dihydrochloride (5.54 mg/kg, i.p., 1% BW, an unconditioned stimulus) that causes visceral discomfort. Previously, we showed that saline injection (1% body weight, i.p.) used in place of an emetine injection did not lead to CTA [25].

The dose of emetine dihydrochloride (MW 553.56, Sigma-Aldrich) was chosen based on our previous study that defined the IC_{50} for emetine as 3.345 mg/kg [4]. To analyze CTA acquisition, the intake of the saccharin solution was measured for 6 days after conditioning using the 1-bottle method.

The experiment spanned 13 days. Rats were handled for a few minutes a day until the conditioning day (day 6). During all experiment days, rats were acclimatized to water deprivation for a total of 20 h 40 min a day. The drinking time for a conditioned stimulus was set to 20 min from 9:00 am. An additional 3 h drinking period from 12:00 – 15:00 was set to provide sufficient water to the animal. On the day 6 of the experiment, the saccharin solution was given for 20 min, and immediately after that, 1 mM emetine dihydrochloride dissolved in saline was intraperitoneally administered at 1% of body weight (equivalent to 5.54 mg/kg emetine dihydrochloride hydrate, containing 4.8 mg/kg emetine). On day 7 of the experiment (first day after conditioning), rats ingested distilled water instead of the saccharin solution during either drinking time. On the day 8 – 13 of the experiment, the amount of saccharin solution ingested for 20 min was measured.

2.2. Bilateral subdiaphragmatic vagal afferent denervation (VX group)

Vagotomy was performed according to the surgical procedures described in previous papers [27–32]. Briefly, rats were subjected to general anesthesia using pentobarbital sodium (40 mg/kg, i.p., somnopentyl®, Kyoritsu Seiyaku co. Tokyo, Japan), supplemented hourly with a mixture given intramuscularly of medetomidine hydrochloride (0.03 mg/kg, Medetomin®, Meiji Seika, Tokyo, Japan) and midazolam (1.5 ml/kg, Midazolam® Fuji, Toyama, Japan). After surgically opening the abdomen, we isolated for the left and right vagal nerve trunks that accompany the esophagus with a stereoscopic microscope. The vagal nerve trunks were freed for 5 – 7 mm as close to the diaphragm as possible, and the epineurium around the vagal nerve trunk was opened and removed. Both nerve vagal tranks were hooked on a self-made bipolar metal clasp. A sponge (sponzel®) impregnated with 5% capsaicin (containing edible oil and 5% ethanol) was applied to the nerve part lifted by the clasp and held for 20 min. After that, the incised abdominal cavity was sutured and closed. In addition, as a sham operation group, the same operations without application of capsaicin were performed. These rats were subjected to CTA experiments one week after the operation.

2.3. Lesions of the area postrema (APX group)

Surgical destruction of the area postrema was performed referring to the surgical procedures of several previous papers [33–40]. General anesthesia was performed in the same manner as described above, and an incision was made approximately 10 mm from the midline of the posterior margin of the skull. Under stereomicroscopic observation, we made an incision from the midline of the rectus capitis posterior muscle to the deep part, and a midline incision in the posterior atlanto-occipital membrane. The position of the area postrema was identified from the

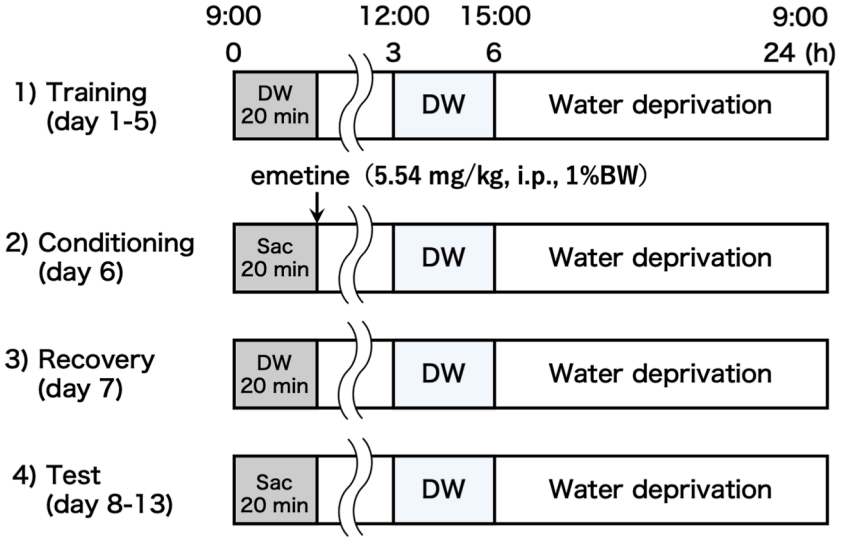

Fig. 1. Protocol for conditioned taste avoidance. Conditioned stimulus, 0.1% saccharin solution; Unconditioned stimulus, emetine-induced nausea. Sac: saccharin, DW: distilled water. Rats were injected with emetine (emetine dihydrochloride, 5.54 mg/kg, i.p., 1% BW).

opening site of the posterior atlanto-occipital membrane at the lower edge of the occipital bone, and the area postrema was lesioned using an electrosurgical knife (J. Morita Tokyo MFG. Corp.). After the incision, a sterile absorbent gelatin sponge (sponzel®) was put on the opening site of the posterior atlanto-occipital membrane to prevent leakage of cerebrospinal fluid, and the surrounding muscle and skin were sutured. In addition, we made sham operated group in which rats had the same surgical procedure of area postrema lesions without turning on the electric scalpel. Rats with surgery were subjected to experiments of CTA one week after the operation. After the experiment was completed, all rats in APX group were perfused with a 4% paraformaldehyde solution under general anesthesia. Tissue slices with 50 μm-thick were prepared from the brainstem and Nissl-staining was done. The lesioned area was confirmed with a microscope. Then, only those in which more than 90% of the area postrema was excised were used for analysis.

2.4. Statistical analysis

We used a one-way analysis of valiance with a Dunnett's multiple comparison test to compare the mean values of saccharine solution intake on each test day with that of conditioning day. The statistical analysis was performed using R (version 3.4.2) and StatPlus (version 7.6.11) with p value less than 0.05 considered statistically significant. All data are represented as mean ± SEM.

3. Results

The changes in body weight and food intake during the period from experimental days 1 – 13 are shown in Figs. 2 and 3, respectively. No persistent weight loss was observed throughout the experiment in any of the groups (Fig. 2). The APX group, however, had a decrease in feeding amount and weight after surgery, which resulted in a lower average weight for this group at the start of the experiment. The feeding amount decreased in all groups on day 6 (conditioning), but after that, feeding levels returned to the same level as before conditioning (Fig. 3). From these results, we concluded that the overall health of rats was well maintained, even after surgical treatment and water deprivation.

Fig. 4 shows the results of the CTA acquisition of rats that were conditioned with emetine dihydrochloride (5.54 mg/kg, i.p., 1% BW) and saccharin solution as an unconditioned and a conditioned stimulus, respectively. It shows the total daily fluid intake and saccharin solution intake on days 1 – 13. The total water consumption remained stable throughout the experiment, but saccharin solution intake varied.

Fig. 5 shows the details of saccharin solution intake on days 6 – 13 except for recovery day 7. In the intact rats, saccharin intake was significantly reduced on the day 8 (2.03 ± 0.41 g, $n = 5$) and day 9 (5.12 ± 0.99 g, $n = 5$) as compared with the conditioning day (10.85 ± 1.20 g, $n = 5$) ($F(6, 28) = 15.204$, $p = 0.00001$ and 0.0029, respectively) (Fig. 5A). In the sham VX group, saccharin solution intake was significantly reduced on day 8 (2.98 ± 0.62 g, $n = 5$) compared with the conditioning day (10.67 ± 1.56 g, $n = 5$) ($F(6, 28) = 6.765$, $p = 0.047$) (Fig. 5B1). In the sham APX surgery group, saccharin intake was significantly reduced on the day 8 (1.49 ± 0.48 g, $n = 5$) as compared with the conditioning day (8.77 ± 0.48 g, $n = 5$) ($F(6, 28) = 5.554$, $p = 0.037$) (Fig. 5B2). We conclude that emetine was effective as an unconditioned stimulus for CTA. In the VX group, saccharin intake was significantly reduced on the day 8 (2.86 ± 0.96 g, $n = 5$) and day 9 (5.34 ± 2.24 g, $n = 5$) as compared with the conditioning day (13.08 ± 0.91 g, $n = 5$) ($F(6, 28) = 11.304$, $p = 0.0001$ and 0.0021, respectively) (Fig. 5C1). Since CTA was significant both in the sham VX and the VX group, we concluded that vagotomy did not affect CTA acquisition.

On the other hand, in the APX group, saccharin solution intake on day 8 (7.21 ± 1.71 g, $n = 5$) and day 9 (9.26 ± 0.17 g, $n = 5$) did not differ from intake on the conditioning day (9.14 ± 0.64 g, $n = 5$) ($F(6, 28) = 3.615$, $p = 0.89$ and 1.0, respectively), indicating there was no CTA in the APX group (Fig. 5C2).

Fig. 6 shows example photomicrographs of coronal sections of the brainstem at the level of the area postrema in the APX group and the intake of distilled water and saccharin solution in this lesioned animal. From the histological image, it was confirmed that the area postrema was destroyed along the boundary with the NTS. This rat failed to acquire emetine-induced CTA. For other rats, we histologically confirmed

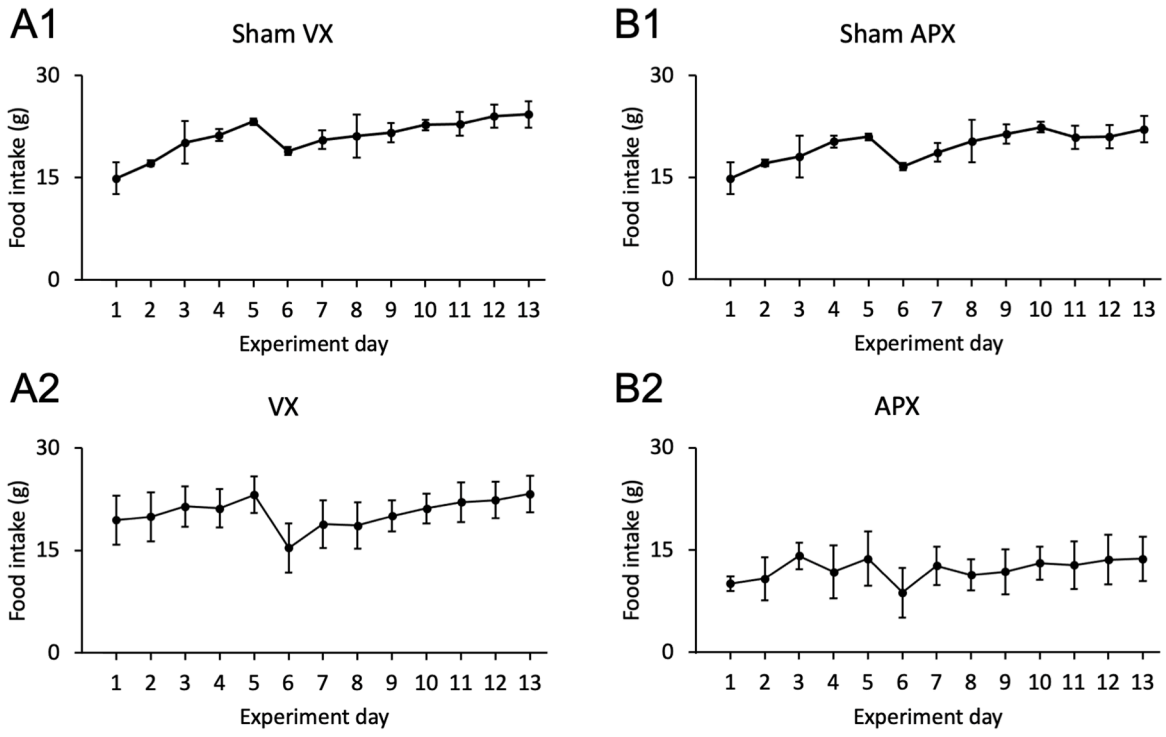

Fig. 2. Temporal change of food intake. Food intake in rats with sham vagotomy (sham VX), (A1), vagotomy (VX), (A2), sham area postrema lesions (sham APX), (B1), and area postrema lesions (APX), (B2). ($n = 5$ in each group).

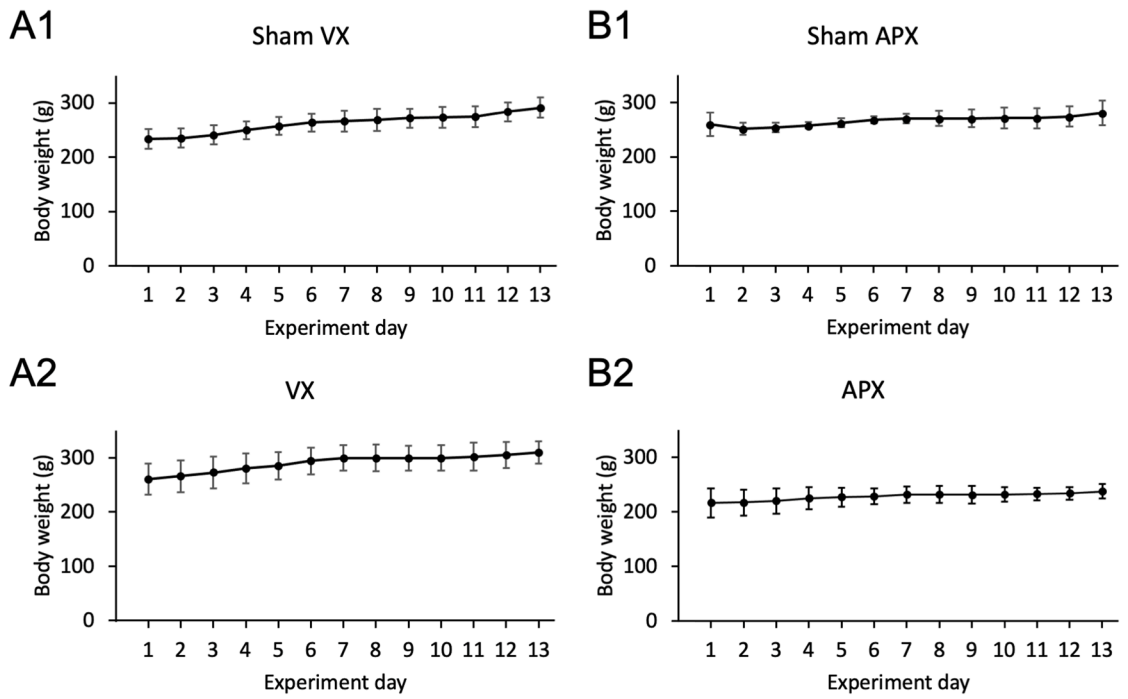

Fig. 3. Temporal change of body weight. Body weight in rats with sham vagotomy (sham VX), (A1), vagotomy (VX), (A2), sham area postrema lesions (sham APX), (B1), and area postrema lesions (APX), (B2). ($n = 5$ in each group).

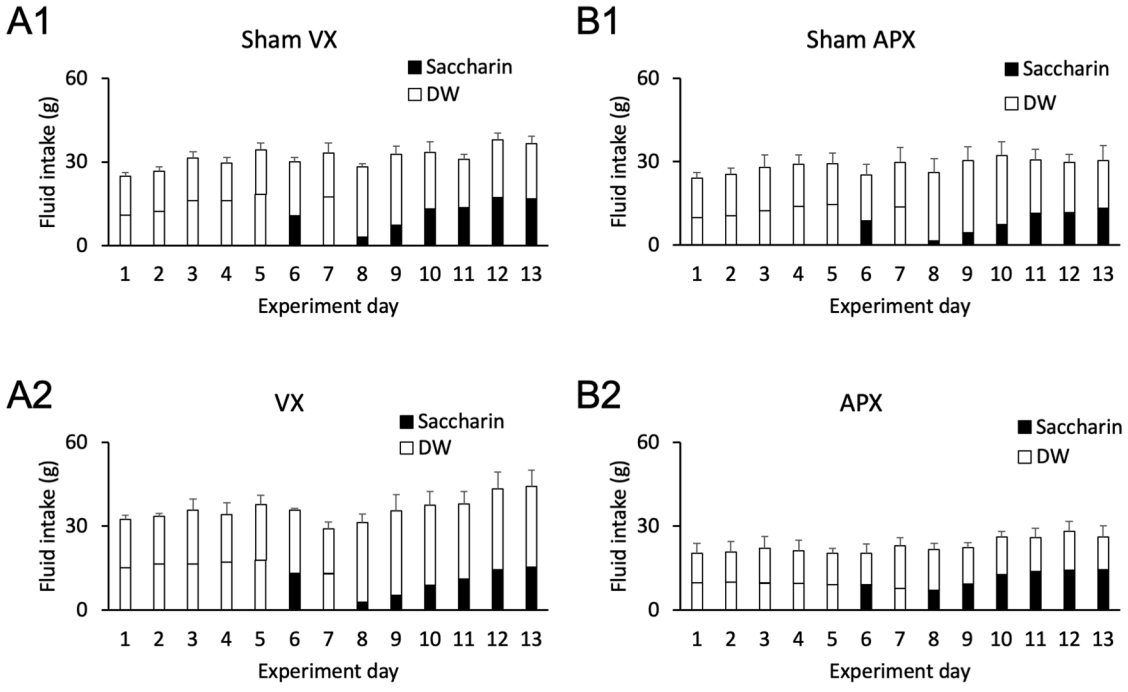

Fig. 4. Temporal change of fluid intake. Fluid intake in rats with sham vagotomy (sham VX, A1), vagotomy (VX, A2), sham area postrema lesions (sham APX, B1), and area postrema lesions (APX, B2). ($n = 5$ in each group).

that all rats in the APX group had damage to area postrema that was similar to that shown in the example illustrated in Fig. 6.

4. Discussion

We found that area postrema lesions, but not lesions of vagal afferent fibers eliminated conditioned taste avoidance in rats. We conclude that area postrema is a critical brain structure for inducing nausea by emetine. In addition, since vagotomized rats still displayed emetine-induced CTA, it is possible that vagal afferent information is not essential for emetine-induced nausea, and that the chemical substance released into the circulation by the action of emetine directly excites chemosensitive neurons in the area postrema. The results of the present study favor a central mechanism for CTA where emetine is the unconditioned stimulus.

Berger et al., 1973 suggested that bait shyness induced by different

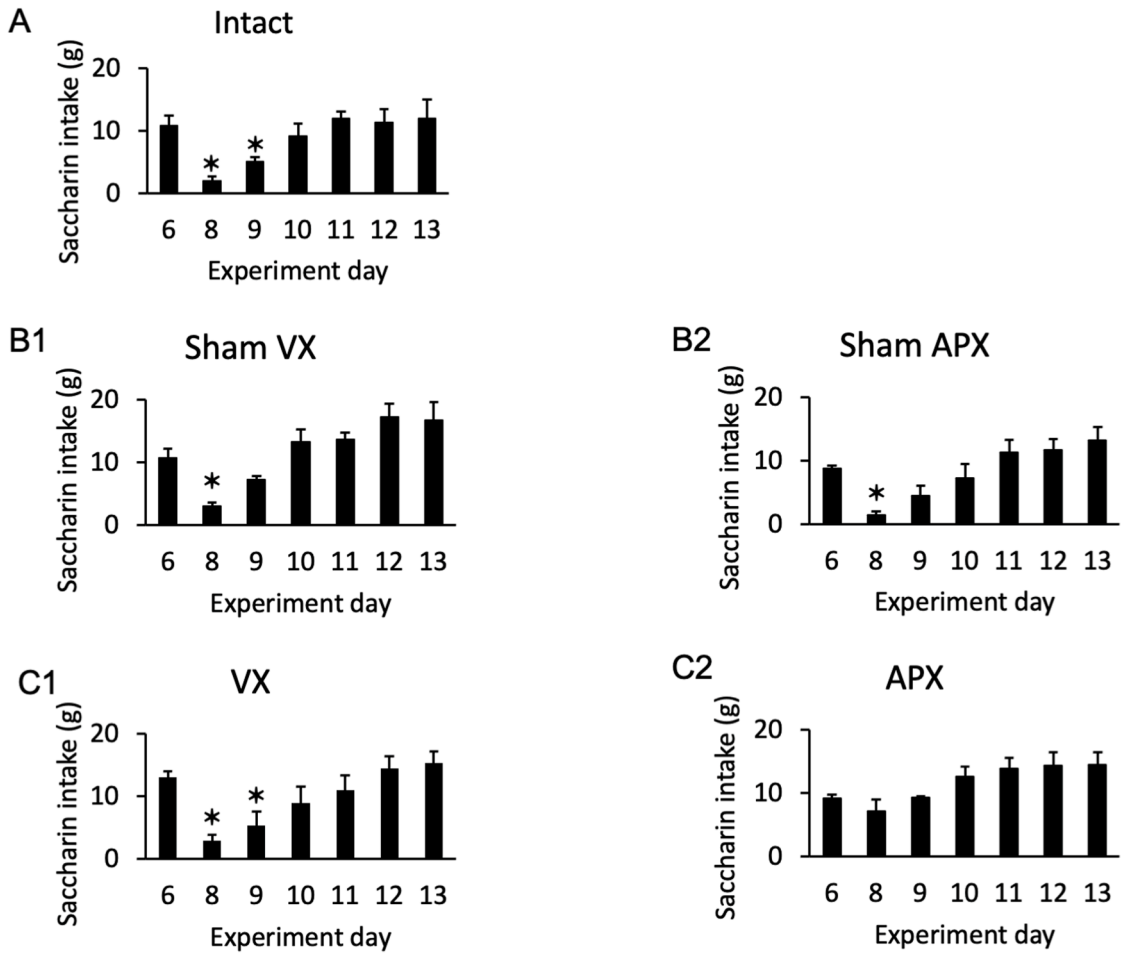

Fig. 5. Temporal change of saccharin intake. Saccharin intake in rats with no surgery (Intact) (A), sham vagotomy (sham VX) (B1) and sham area postrema lesions (sham APX) (B2), vagotomy (VX) (C1), and area postrema lesions (APX) (C2). A, saccharin intake in the intact rats significantly decreased on days of the 1st and 2nd CTA test (day 8 and 9), compared with that on the conditioning day. B1, 2, saccharin intake in both sham groups significantly decreased on days of the 1st CTA test (day 8), compared with that on the conditioning day. C1, saccharin intake in the VX group significantly decreased on days of the 1st and 2nd CTA test (day 8 and 9), compared with that on the conditioning day. C2, no significant decreases of saccharin intake in the APX group as compared with that on the conditioning day. Intact rats ($n = 5$), sham VX and sham APX groups ($n = 5$, in each group), VX: vagotomy group ($n = 5$), APX: area postrema lesion group ($n = 5$). * $p < 0.05$.

drugs are mediated by different mechanisms because the area postrema lesions prevented methylscopolamine-induced food aversion learning, but failed to affect the amphetamine-induced one [34]. The amphetamine-induced CTA is not accompanied by conditioned gaping [41, 42]. Considering that emetine-induced CTA was blocked by area postrema lesions in the present study, conditioned nausea may be produced by the administration of emetine. However, it is necessary to determine this point in further studies.

Our previous study reported that a central mechanism for emetine-induced nausea may involve neuronal activity of the area postrema and the nucleus tractus solitarius (NTS) [4]. The present study confirms that the area postrema is crucial for inducing nausea by emetine. Vagal afferent information may contribute to nausea, but does not do so independent of area postrema.

It is well known that the NTS is the primary relay nucleus of the vagal afferent pathway that conveys the visceral sensation. Synaptic inputs from vagal afferents end in the area postrema, monosynaptically or polysynaptically via the NTS, and can increase neuronal activity of the area postrema neurons [24, 27, 43–45]. In our previous study using c-Fos protein immunoreactivity as an indicator of neural activity, we detected the c-Fos immunoreactive cells in both neurons of the area postrema and the NTS when emetine was intraperitoneally administrated [4]. We also showed that the dendrites of the area postrema neurons extend to the NTS, and it is thought that the vagal afferent fibers are synaptically connected to the dendrites of area postrema neurons within the NTS [45], suggesting that area postrema serves as a higher relay nucleus to convey visceral sensation to other brain regions such as the vomiting center in the brainstem and other higher brain regions.

Since the blood-brain barrier is missing in the area postrema and area postrema neurons can detect chemical substances in the blood circulation, it may be that circulating 5-HT released from enterochromaffin cells in the intestine after emetine administration excites the area postrema neurons.

Although a previous study has reported that vagotomy suppressed cisplatin-induced emesis in ferrets more than 80% [46], emetine-induced CTA was elicited in vagotomized rats in the present study. These results suggest that the level of vagal involvement may differ for different substances that can induce nausea.

Previous studies have investigated vomiting induced by radiation, apomorphine, copper sulfate, emetine, cisplatin, lithium chloride etc. in animals that can vomit. Borison and Wang have demonstrated the neural mechanism of vomiting by the experiment in dogs and cats, that can vomit, and they called the area postrema the emetic chemoreceptor trigger zone [14]. Since their description, it has been demonstrated the effects of the area postrema lesions and antiemetic drugs on the radiation-induced vomiting in dogs [47]. The chemosensitivity of the dog area postrema neurons to various chemical substances, such as transmitters, peptides, cyclic nucleotides, has been elucidated by

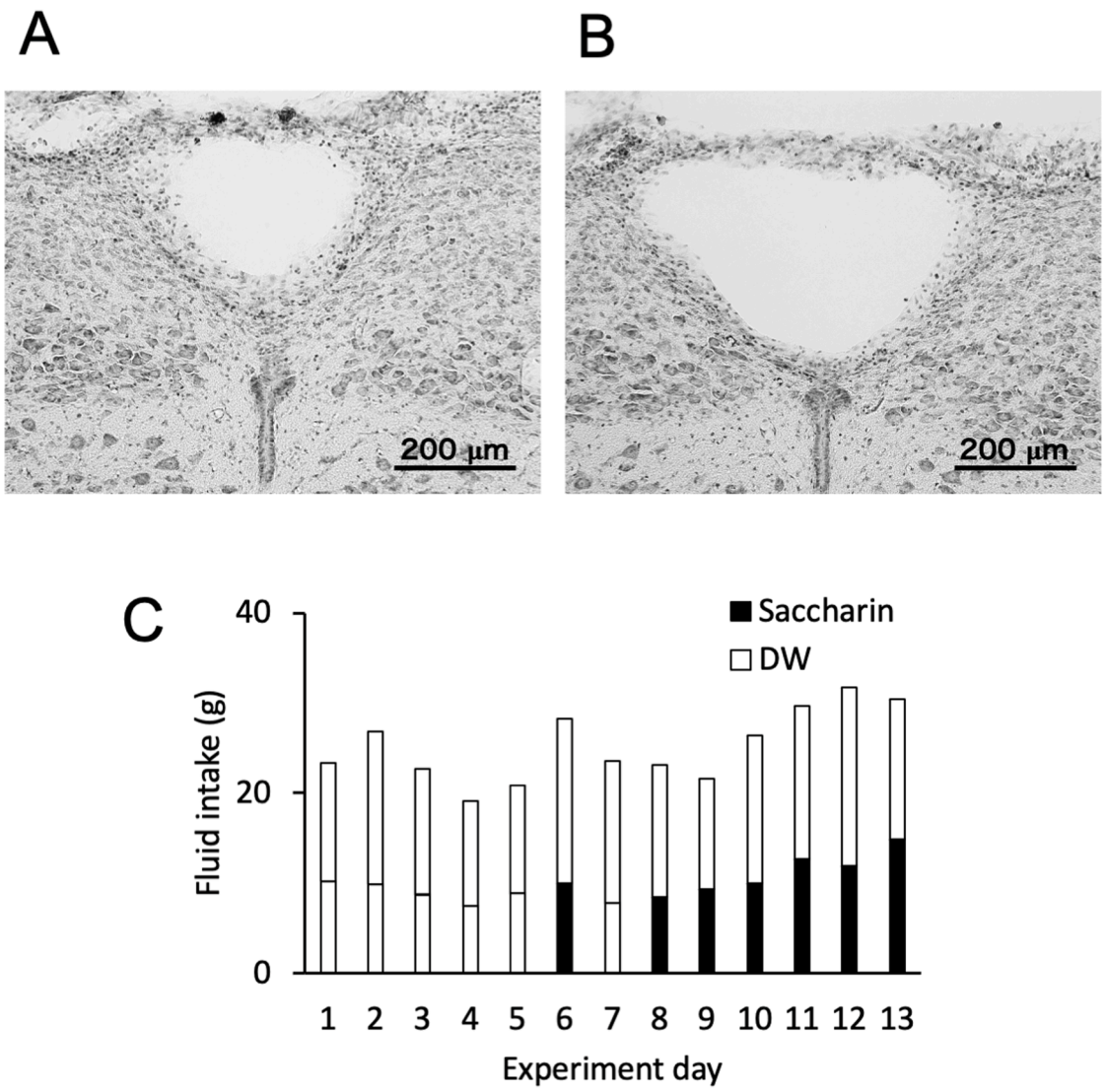

Fig. 6. Example data of a rat in the APX group. A, B, example photomicrographs of Nissl-stained coronal sections of the brainstem at the level of the caudal (A) and rostral (B) part of the area postrema. C, temporal change of fluid intake in the same rat.

extracellular recordings [48].

Although rats are animals that do not vomit, many investigators have conducted experiments with rats to investigate the effects of emetic substances, since it has been demonstrated conditioned taste aversion occurred when the emetic substance was given in pairs with a taste solution [34–36, 38, 49–53]. Several studies have demonstrated that area postrema lesions prevented conditioned taste aversion induced by methylscopolamine, LiCl and copper sulfate in rats [34–36]. The present study suggests that emetine-induced CTA implies a mechanism similar to that induced by methylscopolamine, LiCl and copper sulfate, as emetine-induced CTA was blocked by area postrema lesions.

While the avoidance of a taste paired with an emetic drug has been considered to be the conditioned taste aversion, Parker 2014 suggested that conditioned taste avoidance and conditioned gaping responses are useful as measures of conditioned nausea in rats [54]. This is based on the conditioned gaping reactions for disgusting taste in rats [53]. Conditioned taste avoidance learning is not a selective measurement of the emetic properties of drugs, because non-emetic treatments, such as low doses of amphetamine that reward rats, produce conditioned avoidance, but not conditioned gaping [34, 41, 42, 55]. Anti-emetic treatments are generally ineffective in suppression of conditioned avoidance produced by an emetic drug, but prevent the establishment of conditioned gaping in rats [54, 56].

As emetine is an emetogenic substance, it presumably induces conditioned taste aversion in rats, however, emetine-induced conditioned gaping remained to be determined. Therefore, the term conditioned taste avoidance was used to describe the behavior in rats of lower consumption of saccharin in the present study.

Fukui et al., 1994 suggested that vagal and splanchnic afferent nerve activities via 5-HT_4 receptors are involved in copper sulfate-induced emesis in dogs [57]. Horn et al., 2004 suggested that gastrointestinal vagal afferent nerve activities via 5-HT_3 receptors are involved in the toxic effects of cisplatin in rats [58]. Horn et al., 2014 have shown that the vagal emetic pathways from the gastrointesutinal tract to the NTS, the area postrema, and the lateral parabrachial nucleus are involved in copper sulfate-induced emesis in musk shrews (Sunkus murinas) using a viral tract tracing [59]. They have also reported that vagotomy failed to inhibit emesis produced by optimal emetic doses of copper sulfate, nicotine, and motion in musk shrews. Horn, 2009 has investigated the effects of vagotomy on cisplatin-induced Fos expression in the rat brain that in areas known to play a role in emesis in other species [60]. He has shown that vagotomy reduced cisplatin-induced Fos expression in the caudal and middle levels of the NTS and the central amygdala, suggesting a defined portion of Fos expression dependent on vagal input,

with a majority of this response determined by either direct action of cisplatin or humoral factors on the CNS. In this way, the conflicting results are obtained by the difference of the vomiting substance used as an unconditioned stimulus and the difference of the animal species.

Rotation is also an emetogenic stimulation as shown in motion sickness. Previous studies of dog emesis have demonstrated the critical functional relevance of the area postrema, but not the abdominal visceral afferents, to the rotation-induced emetic responses [61, 62]. Brizzee et al., 1980 have reported that area postrema lesioned squirrel monkeys were refractory to the rotation that can produce motion sickness-induced emesis in the intact monkeys [63]. This is consistent with prior studies by Wang and Chinn in dogs [61, 62]. In contrast, area postrema lesions in rats did not affect or enhance the rotation-induced CTA [64, 65].

Vansant and Baker, 1976 have reported that postoperative nausea and vomiting occurred ten times more frequently when vagotomy was included in the treatment of reflux esophagitis in 311 patients treated by the Hill posterior gastropexy technique of hiatal hernia repair [66]. Vagotomy with anti-reflux surgery was performed in 159 patients (51%), and vagotomy was not included in 152 patients (49%).

Nausea often precedes vomiting or occurs concurrently with vomiting, but nausea and vomiting are phenomena of completely different nature [54]. While nausea is a perception, vomiting is a reflex movement to expel the contents of the gastrointestinal tract. Rats are animals that do not vomit, but they can acquire CTA due to the induction of nausea by administration of emetine. This point strengthens the distinction between nausea and vomiting. Nausea can thus be compared to pain in terms of being a perception in contrast to a biological reflex process. In the case of vomiting induced by stimulation to the gastrointestinal tract, visceral discomfort and the perception of nausea occurs in advance of the vomiting response. Even if one does not vomit, prolonged nausea can elicit various autonomic symptoms such as dizziness, headache, and significant restriction of food intake and drinking. These elements may contribute to eating disorders such as anorexia nervosa, so elucidating the central mechanism of nausea perception may improve the diagnosis and treatment of such disorders.

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References

[1] J. Hojer, W.G. Troutman, K. Hoppu, A. Erdman, B.E. Benson, B. Megarbane, R. Thanacoody, R. Bedry, E.M. Caravati, T. American academy of clinical, C. European Association of poison, T. clinical, position paper update: ipecac syrup for gastrointestinal decontamination, Clin. Toxicol. (Phila) 51 (2013) 134–139.
[2] K.P. Bhargava, P.C. Gupta, O.M. Chandra, Effect of ablation of the chemoreceptor trigger zone (CT zone) on the emetic response to intraventricular injection of apomorphine and emetine in the dog, J. Pharmacol. Exp. Ther. 134 (1961) 329–331.
[3] T. Endo, M. Nemoto, T. Ogawa, H. Tamakai, N. Hamaue, M. Hirafuji, Y. Takeda, M. Hasegawa, Y. Fugii, M. Minami, Pharmacological aspects of ipecac syrup (TJN-119)-induced emesis in ferrets, Res. Commun. Mol. Pathol. Pharmacol. 108 (2000) 187–200.
[4] T. Sato, Y. Hirai, S. Su, W. Zimo, N. Yasuura, T. Inui, M. Funahashi, Involvement of the area postrema and the nucleus tractus solitarius in the emetogenic action of emetine in rats, J. Oral. Biosci. 62 (2020) 310–314.
[5] D.J. Reynolds, N.A. Barber, D.G. Grahame-Smith, R.A. Leslie, Cisplatin-evoked induction of c-fos protein in the brainstem of the ferret: the effect of cervical vagotomy and the anti-emetic 5-HT3 receptor antagonist granisetron (BRL 43694), Brain Res 565 (1991) 231–236.
[6] T. Yamamoto, T. Shimura, N. Sako, S. Azuma, W.Z. Bai, S. Wakisaka, C-fos expression in the rat brain after intraperitoneal injection of lithium chloride, Neuroreport 3 (1992) 1049–1052.
[7] T. Yoshizawa, M. Funahashi, Effects of methyl methacrylate on the excitability of the area postrema neurons in rats, J. Oral. Biosci. 62 (2020) 306–309.
[8] R.J. Contreras, T. Kosten, E. Bird, Area postrema: part of the autonomic circuitry of caloric homeostasis, Fed. Proc. 43 (1984) 2966–2968.
[9] R.C. Ritter, G.L. Edwards, Area postrema lesions cause overconsumption of palatable foods but not calories, Physiol. Behav. 32 (1984) 923–927.
[10] D. van der Kooy, Area postrema: site where cholecystokinin acts to decrease food intake, Brain Res 295 (1984) 345–347.
[11] R.R. Miselis, T.M. Hyde, R.E. Shapiro, Area postrema and adjacent solitary nucleus in water and energy balance, Fed. Proc. 43 (1984) 2969–2971.
[12] M. Iovino, M. Papa, P. Monteleone, L. Steardo, Neuroanatomical and biochemical evidence for the involvement of the area postrema in the regulation of vasopressin release in rats, Brain Res 447 (1988) 178–182.
[13] A.V. Ferguson, P. Smith, Autonomic mechanisms underlying area postrema stimulation-induced cardiovascular responses in rats, Am. J. Physiol. 261 (1991) R1–R8.
[14] H.L. Borison, S.C. Wang, Physiology and pharmacology of vomiting, Pharmacol. Rev. 5 (1953) 193–230.
[15] B. Costall, A.M. Domeney, R.J. Naylor, J.B. Owera-Atepo, J.A. Rudd, F. D. Tattersall, Fluphenazine, ICS 205-930 and dl-fenfluramine differentially antagonise drug-induced emesis in the ferret, Neuropharmacology 29 (1990) 453–462.
[16] H.L. Borison, Area postrema: chemoreceptor circumventricular organ of the medulla oblongata, Prog Neurobiol 32 (1989) 351–390.
[17] C.J. Price, T.D. Hoyda, A.V. Ferguson, The area postrema: a brain monitor and integrator of systemic autonomic state, Neuroscientist 14 (2008) 182–194.
[18] P.L. Andrews, C.J. Davis, S. Bingham, H.I. Davidson, J. Hawthorn, L. Maskell, The abdominal visceral innervation and the emetic reflex: pathways, pharmacology, and plasticity, Can, J. Physiol. Pharmacol. 68 (1990) 325–345.
[19] J. Hawthorn, K.J. Ostler, P.L. Andrews, The role of the abdominal visceral innervation and 5-hydroxytryptamine M-receptors in vomiting induced by the cytotoxic drugs cyclophosphamide and cis-platin in the ferret, Q. J. Exp. Physiol. 73 (1988) 7–21.
[20] H. Fukui, M. Yamamoto, S. Sato, Vagal afferent fibers and peripheral 5-HT3 receptors mediate cisplatin-induced emesis in dogs, Jpn. J. Pharmacol. 59 (1992) 221–226.
[21] M. Mutoh, H. Imanishi, Y. Torii, M. Tamura, H. Saito, N. Matsuki, Cisplatin-induced emesis in Suncus murinus, Jpn. J. Pharmacol. 58 (1992) 321–324.
[22] S.C. Wang, H.L. Borison, Copper sulphate emesis; a study of afferent pathways from the gastrointestinal tract, Am. J. Physiol. 164 (1951) 520–526.
[23] D. van der Kooy, L.Y. Koda, Organization of the projections of a circumventricular organ: the area postrema in the rat, J. Comp. Neurol. 219 (1983) 328–338.
[24] R.E. Shapiro, R.R. Miselis, The central neural connections of the area postrema of the rat, J. Comp. Neurol. 234 (1985) 344–364.
[25] H. Tsuboi, Y. Hirai, H. Maezawa, K. Notani, N. Inoue, M. Funahashi, Effects of treadmill exercise on the LiCl-induced conditioned taste aversion in rats, Physiol. Behav. 138 (2015) 1–5.
[26] Y. Okusha, Y. Hirai, H. Maezawa, K. Hisadome, N. Inoue, Y. Yamazaki, M. Funahashi, Effects of intraperitoneally administered L-histidine on food intake, taste, and visceral sensation in rats, J. Physiol. Sci. 67 (2017) 467–474.
[27] M. Kalia, J.M. Sullivan, Brainstem projections of sensory and motor components of the vagus nerve in the rat, J. Comp. Neurol. 211 (1982) 248–265.
[28] S.J. Marsh, C.E. Stansfeld, D.A. Brown, R. Davey, D. McCarthy, The mechanism of action of capsaicin on sensory C-type neurons and their axons in vitro, Neuroscience 23 (1987) 275–289.
[29] R. Norgren, G.P. Smith, A method for selective section of vagal afferent or efferent axons in the rat, Am. J. Physiol. 267 (1994) R1136–R1141.
[30] P. Moriarty, R. Dimaline, D.G. Thompson, G.J. Dockray, Characterization of cholecystokininA and cholecystokininB receptors expressed by vagal afferent neurons, Neuroscience 79 (1997) 905–913.
[31] Y. Date, N. Murakami, K. Toshinai, S. Matsukura, A. Niijima, H. Matsuo, K. Kangawa, M. Nakazato, The role of the gastric afferent vagal nerve in ghrelin-induced feeding and growth hormone secretion in rats, Gastroenterology 123 (2002) 1120–1128.
[32] V. Ryu, Z. Gallaher, K. Czaja, Plasticity of nodose ganglion neurons after capsaicin- and vagotomy-induced nerve damage in adult rats, Neuroscience 167 (2010) 1227–1238.
[33] H.L. Borison, Effect of ablation of medullary emetic chemoreceptor trigger zone on vomiting responses to cerebral intraventricular injection of adrenaline, apomorphine and pilocarpine in the cat, J. Physiol. 147 (1959) 172–177.
[34] B.D. Berger, C.D. Wise, L. Stein, Area postrema damage and bait shyness, J. Comp. Physiol. Psychol. 82 (1973) 475–479.
[35] S. Ritter, J.J. McGlone, K.W. Kelley, Absence of lithium-induced taste aversion after area postrema lesion, Brain Res 201 (1980) 501–506.
[36] J.D. Coil, R. Norgren, Taste aversions conditioned with intravenous copper sulfate: attenuation by ablation of the area postrema, Brain Res 212 (1981) 425–433.
[37] M. Gallo, M. Arnedo, A. Aguero, A. Puerto, Electrical intracerebral stimulation of the area postrema on taste aversion learning, Behav. Brain Res. 30 (1988) 289–296.
[38] M. Gallo, M. Arnedo, A. Aguero, A. Puerto, The functional relevance of the area postrema in drug-induced aversion learning, Pharmacol. Biochem. Behav. 35 (1990) 543–551.
[39] A.D. Miller, S. Nonaka, Mechanisms of vomiting induced by serotonin-3 receptor agonists in the cat: effect of vagotomy, splanchnicectomy or area postrema lesion, J. Pharmacol. Exp. Ther. 260 (1992) 509–517.
[40] C.M. Spencer, L.A. Eckel, R. Nardos, T.A. Houpt, Area postrema lesions attenuate LiCl-induced c-Fos expression correlated with conditioned taste aversion learning, Physiol. Behav. 105 (2012) 151–160.

[41] L.A. Parker, Behavioral conditioned responses across multiple conditioning/testing trials elicited by lithium- and amphetamine-paired flavors, Behav. Neural. Biol. 41 (1984) 190–199.
[42] L.A. Parker, Taste reactivity responses elicited by reinforcing drugs: a dose-response analysis, Behav. Neurosci. 105 (1991) 955–964.
[43] D. van der Kooy, L.Y. Koda, J.F. McGinty, C.R. Gerfen, F.E. Bloom, The organization of projections from the cortex, amygdala, and hypothalamus to the nucleus of the solitary tract in rat, J. Comp. Neurol. 224 (1984) 1–24.
[44] R.R. Miselis, Shapiro, R.E., Hyde, T.M. The area postrema. In: P. Gross, ed. Circumventricular Organs and Body Fluids, vol. II. Boca Raton FL: CRC Press; 1987. p. pp. 185–207.
[45] M. Funahashi, Y. Mitoh, T. Akaike, R. Matsuo, Variety of morphological and electrophysiological properties of area postrema neurons in adult rat brain slices, Neurosci. Res. 54 (2006) 43–48.
[46] T. Endo, M. MInami, Y. Monma, M. Yoshioka, H. Saito, H. Parvez, Vagotomy and ondansetron (5-HT3 antagonist) inhibited the increase of serotonin concentration induced by cytotoxic drugs in the area postreama of ferrets, Biogen. Amin. 8 (1992) 79–86.
[47] D.O. Carpenter, D.B. Briggs, A.P. Knox, N.L. Strominger, Radiation-induced emesis in the dog: effects of lesions and drugs, Radiat. Res. 108 (1986) 307–316.
[48] D.O. Carpenter, D.B. Briggs, A.P. Knox, N. Strominger, Excitation of area postrema neurons by transmitters, peptides, and cyclic nucleotides, J. Neurophysiol. 59 (1988) 358–369.
[49] J. Garcia, W.G. Hankins, K.W. Rusiniak, Behavioral regulation of the milieu interne in man and rat, Science 185 (1974) 824–831.
[50] M. Nachman, P.L. Hartley, Role of illness in producing learned taste aversions in rats: a comparison of several rodenticides, J. Comp. Physiol. Psychol. 89 (1975) 1010–1018.
[51] R.A. Wise, R.A. Yokel, H. DeWit, Both positive reinforcement and conditioned aversion from amphetamine and from apomorphine in rats, Science 191 (1976) 1273–1275.
[52] E. Ionescu, O. Buresova, Failure to elicit conditioned taste aversion by severe poisoning, Pharmacol. Biochem. Behav. 6 (1977) 251–254.
[53] H.J. Grill, R. Norgren, The taste reactivity test. I. Mimetic responses to gustatory stimuli in neurologically normal rats, Brain Res 143 (1978) 263–279.
[54] L.A. Parker, Conditioned flavor avoidance and conditioned gaping: rat models of conditioned nausea, Eur. J. Pharmacol. 722 (2014) 122–133.
[55] B.M. Rabin, W.A. Hunt, J. Lee, Interactions between radiation and amphetamine in taste aversion learning and the role of the area postrema in amphetamine-induced conditioned taste aversions, Pharmacol. Biochem. Behav. 27 (1987) 677–683.
[56] L.A. Parker, S.A. Rana, C.L. Limebeer, Conditioned nausea in rats: assessment by conditioned disgust reactions, rather than conditioned taste avoidance, Can. J. Exp. Psychol. 62 (2008) 198–209.
[57] H. Fukui, M. Yamamoto, S. Sasaki, S. Sato, Possible involvement of peripheral 5-HT4 receptors in copper sulfate-induced vomiting in dogs, Eur. J. Pharmacol. 257 (1994) 47–52.
[58] C.C. Horn, E.J. Richardson, P.L. Andrews, M.I. Friedman, Differential effects on gastrointestinal and hepatic vagal afferent fibers in the rat by the anti-cancer agent cisplatin, Auton. Neurosci. 115 (2004) 74–81.
[59] C.C. Horn, K. Meyers, A. Lim, M. Dye, D. Pak, L. Rinaman, B.J. Yates, Delineation of vagal emetic pathways: intragastric copper sulfate-induced emesis and viral tract tracing in musk shrews, Am. J. Physiol. Regul. Integr. Comp. Physiol. 306 (2014) R341–R351.
[60] C.C. Horn, Brain fos expression induced by the chemotherapy agent cisplatin in the rat is partially dependent on an intact abdominal vagus, Auton. Neurosci. 148 (2009) 76–82.
[61] S.C. Wang, H.I. Chinn, Experimental motion sickness in dogs; functional importance of chemoceptive emetic trigger zone, Am. J. Physiol. 178 (1954) 111–116.
[62] S.C. Wang, H.I. Chinn, A.A. Renzi, Experimental motion sickness in dogs: role of abdominal visceral afferents, Am. J. Physiol. 190 (1957) 578–580.
[63] K.R. Brizzee, J.M. Ordy, W.R. Mehler, Effect of ablation of area postrema on frequency and latency of motion sickness-induced emesis in the squirrel monkey, Physiol. Behav. 24 (1980) 849–853.
[64] K.P. Ossenkopp, Area postrema lesions in rats enhance the magnitude of body rotation-induced conditioned taste aversions, Behav. Neural Biol. 38 (1983) 82–96.
[65] R.L. Sutton, R.A. Fox, N.G. Daunton, Role of the area postrema in three putative measures of motion sickness in the rat, Behav. Neural Biol. 50 (1988) 133–152.
[66] J.H. Vansant, J.W. Baker, Jr., Complications of vagotomy in the treatment of hiatal hernia, Ann. Surg. 183 (1976) 629–635.