学位論文内容の要約

学位論文題目

Physiological study of the effects of cisplatin-induced conditioned sweet taste aversion on feeding and drinking behavior in rats

（シスプラチン誘発条件付け甘味嫌悪がラットの摂食・飲水行動に
与える影響に関する生理学的研究）

博士の専攻分野名称　　博士（歯学）　　氏名　魏 紫茉

When rats experience sweet solution intake and nausea induction at the same time, they develop a conditioned taste aversion and reduce their next intake of sweet solution. When rats are reared as laboratory animals, solid feed for rats is used, but this feed does not contain sweetening ingredients, and rats do not experience sweet taste throughout their lives under normal rearing. When such rats ingest sweetened solution for the first time, they initially show fear of the novel taste and ingest the sweetened solution with carefully tasting whether it is safe or not.

Since sweet taste is innately perceived as an energy source and pleasant information, sweet taste preference eventually instinctively becomes dominant, followed by a gradual increase in sweet solution intake. Since Garcia et al., 1955 proposed the concept of CTA, many researchers have conducted animal experiments using rats and mice to elucidate the neural mechanisms of conditioned taste aversion or avoidance. When rats acquire CTA by conditioning with a pair of sweet taste and nausea induction, they avoid the next intake of saccharin, despite being thirsty due to water deprivation. Most of these studies compare the intake of sweetened solutions at a certain time (15-30 minutes) after forcing experimental animals deprived of water for a considerable period of time. In other words, this is tantamount to assessing how much water of an unpleasant taste the experimental animals will consume even against thirst. Our question here is how much rats that have acquired a CTA for sweetness will consume an unpleasant sweet-tasting diet, even against hunger. Hunger sensation is a physiological motivation that triggers feeding behavior and is an important emotion essential for life maintenance. We hypothesized and tested in this study that acquisition of a conditioned aversion memory for a specific taste would lead to avoidance of ingestion of food.

Emetogenic drug-induced nausea (visceral discomfort) was used as an unconditioned stimulus to induce CTA to saccharin. In this study, emetine, which induces acute nausea, and cisplatin, an antineoplastic drug that induces acute and delayed nausea, were used.

Cisplatin-induced nausea and vomiting is one of the side effects of antineoplastic drugs, and antiemetic therapy is often given in parallel with cancer chemotherapy. Severe nausea and vomiting caused by the side effects of anticancer drugs not only reduce the quality of life, but also decrease the effectiveness of chemotherapy treatment due to reduced food intake caused by anorexia and a decrease in physical strength and natural healing power. In such cases, ondansetron or tropisetron, inhibitors of serotonin type 3 (5-HT_3) receptors, are often prescribed as antiemetic agents.

The present study was conducted to test the hypothesis that feeding behavior is suppressed by emetine- and cisplatin-induced conditioned taste aversion memory and that the mechanism involves neural information mediated by 5-HT_3 receptors. The ultimate goal of this study is to examine and discuss whether the central mechanism of CTA is associated with the pathogenesis of eating disorders.

Thirty-nine male Sprague-Dawley rats were used for the experiments. Their body weight was 180-220g at the beginning of every experiment period. Rats were individually kept in cages and the light/dark cycle was 12h. All animals were handled according to the Hokkaido University Animal Experiment Guidelines. Rats were feed by normal diet or self-made sweet diet. Sweet diet was prepared by mixing regular solid diet with an equal amount of 0.2% saccharin solution. The normal diet was also processed in the same way to match the texture.

We used modified CTA protocol in which the food deprivation was added. The experiment was run for 12 days. Throughout of the experiment, rats were acclimated to a total of 20 h 40 min of water deprivation and 22 h 40 min of food deprivation per day.

The CTA conditioning with 0.1% saccharin solution was started at 10:00 a.m. on experiment day 6 (the conditioning day). The unconditioned stimulus of emetine hydrochloride (5.54 mg/kg, 1% BW, i.p., n = 6) or cisplatin dihydrochloride (3mg/kg, 1% BW, i.p., n = 9) were applied at 10:20 a.m. In

the control group of rats, saline (1% BW, i.p., n = 6) was used instead of emetine or cisplatin. On the day 7 for recovery, rats drank distilled water instead of saccharin solution. On the test day 1 to 5 (experiment day 8‑12) , the rats were fed sweet diet instead of the normal diet for 20 min from every 10:00 a.m.. The amount of sweet diet intake during this period was measured. The rats' body weight, fluid intake and normal diet intake were also measured on each experimental day.

Some cisplatin-conditioned groups received ondansetron (0.1 mg/kg, 0.1% BW, i.p.), a 5-HT_3 receptor antagonist, 30 min before the conditioned stimulus (n=6) or just before the start of the test (n = 6). In addition, some cisplatin conditioned groups were also tested on normal feed during the 20 min of tests 1-5 (n = 6).

The mean consumption of sweet diet was analyzed with a one-way analysis of variance. The comparisons of each test days with conditioning day were performed with the Dunnett's multiple test. A p value less than 0.05 was considered statistically significant. Statistical analysis was performed with StatPlus (version 7.8.11).

The cisplatin-treated group showed a significant decrease in the body weight compared to the saline-treated group on test 1-5 ($F(5,65) = 8.326$, $P < 0.05$). While there was no significant weight loss in other groups. The cisplatin-treated group showed a significant decrease in fluid intake per body weight compared to the saline-treated group on test 1 and 4 ($F(5,65) = 2.654$, $P < 0.05$). The emetine-treated group showed a significant decrease in sweet diet intake on test 1-5 compared to saline-treated group ($F(5,50) = 5.159$, $P < 0.05$), and showed a significant decrease in sweet diet intake and sweet diet intake per body weight in the test 1-4 as compared to the conditioning day ($F(5,30) = 9.737$, $P < 0.05$ and $F(5,30) = 7.497$, $P < 0.05$). The cisplatin-treated group showed a significant decrease in sweet diet intake per body weight in the test 1and 2 as compared to the conditioning day ($F(5,48) = 2.55$, $P < 0.05$ and $F(5,48) = 4.639$, $P < 0.05$). We also found that the amount of normal diet intake was not significantly reduced in test 1-5. Pre-administration of ondansetron abolished the decrease in sweet diet intake in the cisplatin group. In the control group, sweet diet consumption was rather increased in the test 3-5 compared to conditioning day.

The emetine-induced CTA group showed no weight loss during the experimental period, while the cisplatin-induced CTA group showed significant weight loss compared to the conditioning day. It was found that ondansetron treatment prevented weight loss in the cisplatin-treated group. In addition, rats that acquired CTA to the sweet taste of saccharin showed suppressed feeding behavior even in the fasting state. Furthermore, it was suggested that this feeding inhibition may be mediated by 5-HT_3 receptors. These results suggest that CTA may alter feeding behavior and provide useful information for further research on the central mechanism of eating disorders.