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Reduced Palatability in Pain-Induced Conditioned Taste Aversions

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Abstract

The current study investigated whether internal pain-inducing agents can modulate palatability of a tastant in the same way as illness-inducing agents (e.g., lithium chloride). Similar to traditional conditioned taste aversion (CTA) experiments, during conditioning the rats were exposed to a saccharin solution followed by intraperitoneal injections of either gallamine (Experiment 1) or hypertonic sodium chloride (NaCl; Experiments 1 and 2). In addition to the total amount consumed, the time of each lick was recorded for lick pattern analysis. The results showed that both gallamine and hypertonic NaCl caused suppression in saccharin intake. Importantly, both lick cluster size and initial lick rate (the measures of taste palatability) were reduced as well. This pattern of results suggests that these pain-inducing agents reduce the hedonic value of the associated tastant and thus CTA is acquired. The current finding serves as evidence supporting the view that CTA is a broadly tuned mechanism that can be triggered by changes in internal body states following consummatory experience.

Keywords

Lick cluster size; Lithium chloride; Gallamine; Hypertonic sodium chloride; Rats

1. Introduction

Animals, humans included, avoid eating foods that have previously caused them to experience gastrointestinal malaise. That is, the taste of the food (conditioned stimulus, CS) becomes associated with the aversive post-ingestive effects of food consumption (unconditioned stimulus, US) and a conditioned taste aversion (CTA) is acquired (for reviews see edited volumes by [5,8,43,52]). Not only is intake of the CS suppressed but also, as shown with both taste reactivity (e.g., [10,16,33,48,49,58]) and lick pattern analysis (e.g., [3,4,27,40]), the CS itself is devalued. That is, the palatability of the CS is reduced consequent to pairings with the US. This latter effect ensures that we do not accidentally or mistakenly consume the toxic food because it is now disgusting and quickly ejected from the mouth. Thus, one can argue that a reduction in taste palatability drives the suppression in consumption of the illness-inducing food.

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The present research investigated whether a change in palatability occurs when a taste CS is paired with a US that causes internal pain. A number of studies have found that external pain, induced with footshock, can suppress intake of the associated taste CS (e.g., [30,51]). However, the results of a study by Brett ([11]; discussed in [29]) clearly show that peripheral pain does not cause CTAs. In Brett's experiment, rats, receiving a foot shock at the mid-point of 60-sec taste trials, learned to suppress CS intake prior to the shock but drank avidly thereafter. Thus, it appears that taste can serve as a signal for the external pain of footshock but that the taste is not devalued.

The role of internal pain remains largely unresolved, however. Indeed, we are aware of only one study that investigated the palatability of a taste CS followed by an internal pain US (specifically lower gastrointestinal pain caused by lactose malabsorption; [51]). In that study, palatability was assessed using taste reactivity measures, a technique involving analysis of stereotypical orofacial and somatic responses evoked by taste stimuli [32-34]. Although most taste reactivity studies use a procedure in which a small, controlled volume of a tastant is infused directly into the mouth via a cannula implanted in the rat's cheek, the task employed by Pelchat et al. required voluntary ingestion of 40% lactose, which served as both the CS and US. The results show that lactose suppressed intake but had no influence on taste palatability (assessed by the number of rats showing aversive orofacial and somatic responses on the test trial). Because no aversive responses were detected the pattern of results was interpreted as evidence that the internal pain-caused by lactose malabsorption produces conditioned taste avoidance not CTA.

There are, however, a number of reasons why this interpretation of the Pelchat et al. [51] result cannot be accepted with confidence. First, intake of lactose on the first conditioning trial ranged from 0.3 ml to 15.0 ml, indicating wide individual variability in experience with the CS and, perhaps more importantly, the US properties of lactose. Second, because lactose is not very soluble, the 40% solution was presented to the subjects at 35 °C. In turn, to prevent the temperature of the CS from becoming a salient cue during conditioning, all water presented to the thirsty rats during the experiment was also presented at 35 °C. That is, an unconventional experimental procedure was employed with all the attendant concerns about the comparability and generality of the obtained results. Third, intake on the test trial achieved a group mean of less than 0.5 ml and it is not clear, in the absence of intraoral delivery, how much lactose made contact with the mouth or was spilled from the spout. That is, low intake volumes may have influenced detection of aversive taste reactivity responses. For these reasons, the procedure and test stimulus may not have been optimal to evaluate whether internal pain influences taste palatability. Accordingly, the current study sought to reexamine this issue using a procedure more closely resembling that of the traditional LiCl-induced CTA.

In the present study we used 0.1% saccharin as the CS in each experiment where it was paired with gallamine or hypertonic sodium chloride (NaCl) in our standard voluntary intake procedure. Like curare, gallamine blocks cholinergic transmission at the neuromuscular junction [15] thereby inducing paralysis and pain in muscle tissue. Hypertonic NaCl is well established as a laboratory model of visceral pain in rats (e.g., [31]). These two agents were administered intraperitoneally thus providing a better comparison to studies with LiCl as the US. Experiment 1 examined whether gallamine or hypertonic NaCl can significantly reduce CS palatability along with volume consumed. As a positive control, LiCl was included as a US along with a no-US (isotonic saline) control group. Experiment 2 focused on hypertonic NaCl as the US and involved parametric changes intended to increase CS-US contiguity and thus sensitivity to the detection of changes in palatability of the associated saccharin CS.

Given the use of a voluntary drinking procedure, lick pattern analysis was used to assess the conditioned changes in the palatability of the taste CS. The raw data from such an analysis supply lick counts and inter-lick interval times which reveal that rats, during trials that typically last 30 min, produce sustained runs of rapidly occurring licks (termed clusters) that are separated by pauses of varying durations [1,12,23]. Thus, a number of measures can be extracted including: total licks, initial and overall lick rates, number of clusters, inter-cluster interval and lick cluster size. However, not all of these variables reflect taste palatability [17]. Total licks and number of clusters are each highly correlated with amount consumed but not the concentration of the tastant [9,25,56,59]. Similarly, inter-cluster intervals, which show a skewed distributed, do not seem to code palatability [19,23]. Indeed, long inter-cluster intervals (ranging from 5 s to over 100 s) indicate that the rat has stopped licking and is engaged in other behaviors such as grooming or exploring. Cluster size, however, is highly controlled by sensory input from the taste solution [20,21]. For example, for a palatable taste like sucrose, there is a monotonic relationship between concentration and cluster size - as the former increases or decreases so too does the latter. It should also be noted that larger cluster size does not necessarily imply a larger volume consumed. There is, in fact, an inverted U-shaped function between volume consumed and the concentration of palatable solutions [18,23,59,60]. On the other hand, for an unpalatable tastant like quinine, cluster size decreases monotonically as concentration increases [36,61].

Another line of evidence that supports the use of cluster size as an index of taste palatability comes from studies in which manipulations that influence taste palatability produce comparable effects on both cluster size and taste reactivity responses. For instance, benzodiazepines are considered to stimulate feeding by enhancing taste palatability as revealed by an increased frequency in the occurrence of ingestive taste reactivity responses (e.g., [50,53,57]) and an increase in the size of lick clusters (e.g., [35]). As noted already, illness-induced with LiCl decreases the palatability of the associated taste CS as determined with taste reactivity methodology and cluster size analysis. Overall, these findings, which in many instances have been used as cross-validation for the two approaches, along with those in the preceding paragraph support the view that cluster size is a reliable measure of taste palatability during voluntary intake tasks (e.g., [18,22,23,35,39,59]; for a review see [26]).

Finally, it has been suggested that initial lick rate also represents the affective reactions to a tastant (e.g., [20,47]). However, this measure may have less utility than cluster size. For example, initial lick rate is defined by an arbitrary break (e.g., 1 min or 3 min) determined by the experimenter and not by a pause in licking. Furthermore, given that this measure is taken from a relatively small portion of the whole drinking period, the influence of experimental manipulations on initial lick rate may, in some circumstances, be obscured by ceiling effects in lick rate [59]. Nonetheless, initial lick rate is considered a relatively pure measure of the sensory properties of the taste solution because the short sample period minimizes post-ingestive feedback [22], which, of course, is one of the merits of using taste reactivity methodology to assess palatability. Therefore, in addition to cluster size, initial lick rate was included as an additional measure of taste palatability in the current study.

2. Experiment 1

This experiment examined whether the internal pain-inducing USs gallamine and hypertonic NaCl, like the illness-inducing US LiCl, reduces palatability as well as CS intake. To prevent the rapid development of floor effects in CS consumption, the LiCl US (0.075 M) was half our standard concentration and hypertonic NaCl (1.0 M) was half the concentration employed by Sakai and Yamamoto [55]. To our knowledge, two studies [37,40] have investigated whether gallamine can suppress intake of a taste CS. In the first of these studies, Ionescu and Buresova [37] reported that all rats given a 40-mg/kg dose of gallamine (which

failed to induce taste suppression in a one-trial procedure) had to be placed on artificial respiration for the duration (1 - 2 hr) of muscular paralysis. A preliminary study in our laboratory of gallamine-induced taste suppression using 10 and 20 mg/kg found that the lower dose was sufficiently high to suppress CS intake without generating lethal consequences in the absence of support from artificial respiration. Accordingly, the 10-mg/kg dose of gallamine US was used in the present, multiple-conditioning trial experiment.

2.1. Materials and methods

2.1.1. Subjects—A total of 40 experimentally naïve male Sprague-Dawley rats (Charles River Laboratory, Wilmington, MA) were used as subjects. The rats were individually housed in hanging steel cages (Acme Metal Product, Chicago, IL) with free access to food and water in a colony room that was illuminated for 12 hr each day beginning at 7:00 am. Prior to the behavioral treatments, the subjects were acclimated to a water deprivation schedule that permitted 15 min access each morning in the drinking chambers and 15 min access each afternoon in the home cages; food was always available in the home cages. The University of Illinois at Chicago Animal Care and Users Committee approved all procedures employed in the present study. At all times, rats were treated according to guidelines recommended by the American Psychological Association [2] and the National and the Institutes of Health [45].

2.1.2. Apparatus—Eight identical drinking chambers (Med Associates, St. Albans, VT; 30.5 cm × 24.0 cm × 29.0 cm) were employed, each housed in a sound-attenuating cubicle fitted with a ventilation fan. The sidewalls of each chamber were made of aluminum, the front wall, back wall and ceiling were clear Plexiglas and the floor consisted of 19 stainless rods. A house light and a white noise generator (providing a masking noise level of 80 dB) were mounted at the top of the left wall in the chamber. Fluid was presented in a retractable sipper tube accessible through an oval hole (1.3 cm × 2.6 cm) in the middle of the right wall. In its extended position, the tip of sipper tube was ~3 mm outside the wall. A lickometer circuit allowed monitoring of the time of each lick with a resolution of 10 milliseconds. Event control and data collection from the drinking chambers were carried out on-line with a computer running Med-PC software (Med Associates).

2.1.3. Procedure—The rats were counterbalanced into four groups (n = 10/group) according to baseline water intake on the day before the first conditioning trial, when their body weights ranged from 337 g to 435 g. On the each conditioning trial, the rats were given 15 min access to 0.1% saccharin in the drinking chambers followed, 5 min later, by an intraperitoneal injection of isotonic (0.9%) NaCl (Group Control), 0.075 M lithium chloride (Group LiCl), 10 mg/kg gallamine hydrochloride (Group Gallamine) and hypertonic (1.0 M) NaCl (Group hNaCl); the dose for each US injection was 0.5 ml/100 g body weight. All groups, except for Group LiCl, received five CS-US conditioning trials and a single CS only test trial, each trial spaced 72 hr apart. Since the CS intake of Group LiCl was completely suppressed after three CS-US pairings, the LiCl injections were thereafter omitted such that this group received three CS only trials. Water was available for 15 min in the drinking chambers on each of the intervening mornings.

2.1.4. Data analysis—Three dependent variables were analyzed: volume consumed, lick cluster size, and initial lick rate. Intake was measured by weighing the bottles before and after each drinking session at a resolution of 0.1 g. Lick cluster size was defined as a run of licks with inter-lick intervals less than 0.5 s. While a 1 s criterion has also been used in literature, the 0.5 s inter-lick interval was chosen to provide comparability with previous studies investigating the effects of drug USs on palatability shift (e.g., [3,27,42]). Initial lick rate was determined as the total licks occurring during the 3 min following the first lick.

Statistical analyses were conducted with analysis of variance (ANOVA) using US as the between-subject variable and Trial as the within-subject variable. Significant main effects or interactions were followed by post hoc comparisons (i.e., simple main effect with adjusted error terms taken from the overall ANOVA). The alpha level was set at $p < .05$. All statistical analyses were conducted using Statistica software (StatSoft, Inc. Tulsa, OK).

2.2. Results and Discussion

Water consumption data from the day before first conditioning trial were analyzed with separate one-way ANOVAs that found no significant group effects for amount consumed, lick cluster size, or initial lick rate ($ps > .05$).

The saccharin intake data from the four groups of Experiment 1 are displayed in Figure 1A. It is apparent from inspection of the figure that all groups consumed the same amount of the saccharin CS on Trial 1. Thereafter, as expected, saccharin intake of the control group increased and quickly reached asymptote. Similarly, the performance of Group LiCl conformed to expectations with the rats requiring only two conditioning trials to achieve maximal levels of saccharin suppression across Trials 3 - 6. With regard to the two USs of most theoretical interest, Group Gallamine reduced intake across trials whereas Group hNaCl showed little variation from trial to trial. These impressions were substantiated with statistical analyses which initially revealed a significant main effect of US, $F(3, 36) = 17.34$, $p < .001$, a significant main effect of Trial, $F(5, 180) = 11.64$, $p < .001$, and a significant US x Trial interaction, $F(15, 180) = 10.23$, $p < .001$. Post hoc comparisons confirmed that there were no between-group intake differences on Trial 1 ($p > .05$) and that Group Control increased intake from Trial 1 to Trial 2 ($p < .05$) and that intake did not vary between Trials 2 - 6 ($ps > .05$). Group LiCl decreased intake from Trial 1 to Trial 2 to Trial 3 ($ps < .05$) and showed no change across Trials 3 through 6 ($ps > .05$) when intake was completely suppressed. Intake of the saccharin CS in Group Gallamine numerically decreased across each successive trial and there was a significant difference between Trial 1 and Trials 4, 5, and 6 ($ps < .05$). However, the consumption levels of Group hNaCl were maintained at similar amount across the six taste trials ($ps > .05$).

The lick cluster size (see Figure 1B) analysis followed the same pattern as amount consumed. Accordingly, the initial ANOVA showed significant main effects of US, $F(3, 36) = 3.54$, $p < .05$, and of Trial, $F(5, 180) = 3.42$, $p < .01$, as well as a significant US x Trial interaction, $F(15, 180) = 4.40$, $p < .001$. Post hoc comparisons revealed there were no differences between groups on Trial 1 ($ps > .05$). Lick cluster size in Group Control increased from Trial 1 to Trial 2 ($p < .05$) and was stable across subsequent trials ($ps > .05$). The rats in Group LiCl decreased their cluster size from Trial 1 to Trial 2 ($p < .05$) and did not differ thereafter ($ps > .05$). Group Gallamine gradually reduced lick cluster size across trials and a significant difference was found between Trial 1 and each of Trials 4 - 6 ($ps < .05$). For Group hNaCl, there were no significant differences between any two successive trials ($ps > .05$), except Trials 5 - 6 ($p < .05$). Consistent with lick cluster size, initial lick rate (see Figure 1C) was suppressed in rats receiving contingent injections of LiCl and gallamine; neither physiological saline nor hypertonic NaCl influenced performance. A two-way ANOVA found significant main effects of Group, $F(3, 36) = 23.23$, $p < .001$, and Trial, $F(5, 180) = 35.79$, $p < .001$, as well as a significant Group x Trial interaction, $F(5, 180) = 11.79$, $p < .001$. Post hoc comparisons reveal that LiCl suppressed initial lick rate after one conditioning trial ($p < .001$) whereas two pairings were necessary for gallamine to significantly reduce performance ($p < .05$). On the other hand, the initial lick rates of the Control and hNaCl groups were constant across all six trials ($ps > .05$).

As demonstrated in the present results, gallamine, like LiCl, suppressed CS intake as well as cluster size and initial lick rate. That is, gallamine caused a decrease in the palatability of the

associated taste CS, an effect that we interpret as evidence of the acquisition of a CTA not conditioned taste avoidance (which requires that palatability is not influenced by association with the US). On the other hand, the hypertonic NaCl US appeared to have little influence on either volume consumed or lick cluster size.

3. Experiment 2

Although the gallamine results of Experiment 1 are clear, the findings obtained with hypertonic NaCl are ambiguous. That is, despite the apparent failure of hypertonic NaCl to suppress fluid intake relative to the Trial 1 level, there was no indication of an habituation of taste neophobia in this group (unlike Group Control that showed a significant elevation of saccharin intake and cluster size on Trial 2 relative to Trial 1). Does the failure to recover from taste neophobia indicate the development of a mild taste aversion in Group hNaCl? This view is also suggested by the significant reduction of lick cluster size on Trial 6 relative to Trial 5. We reasoned that the parameters of Experiment 1 (specifically 15-min CS duration and 5-min inter-stimulus interval) may not have been optimal for the detection of CTA induced with hypertonic NaCl. In this context it might be noted that the behavioral reaction to an injection of hypertonic NaCl was very noticeable. That is, injections of hypertonic NaCl caused an apparent immediate visceral pain reaction expressed as severe contractions of the abdominal musculature, writhing, and scratching at site of the injection (see also [14,31]) which, in the present experiment, terminated abruptly after ~15 s. This observation suggests that the duration of the visceral pain effects of hypertonic saline is brief (certainly with respect to the action of the other USs tested in Experiment 1) which, of itself, may influence the effectiveness of that US to support taste aversion learning. Based on this reasoning, the procedure of Experiment 2 increased CS-US contiguity by using a shorter CS duration (5 min) and a shorter CS-US delay (2 min) than those employed in Experiment 1 (respectively, 15 min and 5 min). In our experience, short duration CSs tend to produce ceiling effects that can obscure the detection of group differences in some measures of performance. Accordingly, to increase sensitivity to the detection of group differences, two 15-min taste only test trials were given after four CS-US conditioning trials.

3.1. Materials and methods

3.1.1. Subjects and apparatus—The subjects were 16 experimentally naïve male Sprague-Dawley rats that were housed and maintained as described in Experiment 1. The drinking chambers were those previously used in Experiment 1. The day before the first conditioning trial the rats ranged in weight between 377 to 486 g.

3.1.2. Procedure—The procedure was similar to that of Experiment 1 with the following exceptions. On each conditioning trials, the rats received 5 min access to 0.1% saccharin and, 2 min later, were given an intraperitoneal injection of isotonic saline (Group Control; $n = 8$) or hypertonic NaCl (Group hNaCl; $n = 8$). The two CS only test trials were each 15 min in duration. As in Experiment 1, trials were spaced 72 hr apart.

3.1.3. Data analysis—The behavioral data collected from both the conditioning trials and CS-only test trials were analyzed in the same way as described in Experiment 1.

3.2. Results and Discussion

Baseline water performance was analyzed with separate one-way ANOVAs and revealed no significant group differences for each dependent variable (i.e., intake, lick cluster size, and initial lick rate) on the day before the first conditioning trial ($ps > .05$).

Examination of the data from the conditioning trials (see Figure 2, left panels) suggests that the modification of parameters were effective in that intake and initial lick rate, but not cluster size, of the saccharin CS were reduced by contingent injections of the hypertonic NaCl US. Supporting this view there was, for intake (Figure 2A), a significant US X Trial interaction, $F(3, 42) = 10.11$, $p < .001$, and a significant main effect of Trial, $F(3, 42) = 5.65$, $p < .01$; the main effect of US was not significant, $F(1, 14) = 3.80$, $p = 0.07$. Post hoc analyses revealed that the US X Trial interaction was due to significantly lower CS intake ($ps < .05$) in Group hNaCl relative to Group Control on Trials 3 and 4 only. For lick cluster size (Figure 2B) there was no significant main effect of US ($F < 1$), or Trial ($p > .20$), and no significant US X Trial interaction ($F < 1$). Analysis of the initial licks rate (Figure 2C) found a significant main effect of Trial, $F(3, 42) = 6.71$, $p < .001$, and a significant Group x Trial interaction, $F(3, 42) = 6.65$, $p < .001$; the main effect of Group was not significant, $F(1, 14) = 2.90$, $p > .05$. Post hoc comparisons revealed that although Group Control maintained a high stable level of performance, probably reflecting a ceiling effect, the initial lick rate of Group hNaCl gradually decreased across trials and achieved significance on Trials 3 and 4 ($ps < .05$). This latter result indicates that the hypertonic NaCl US reduced taste palatability.

This conclusion was further confirmed by analysis of the data from the taste only test trials (when access was extended to 15 min from the 5 min used on the conditioning trials) which revealed significant reductions in intake as well as lick cluster size and initial lick rate in Group hNaCl relative to Group Control. Thus, for volume consumed (Figure 2D), analysis found significant main effects of US, $F(1, 14) = 9.09$, $p < .01$, and Trial, $F(1, 14) = 5.62$, $p < .05$, but no significant interaction ($p > .20$). Importantly, for both lick cluster size (Figure 2E) and initial lick rate (Figure 2F), analysis revealed a significant main effect of US, $F(1, 14) = 9.72$, $p < .01$, and $F(1, 14) = 7.69$, $p < .05$, respectively; neither the main effect of Trial nor the US X Trial interaction was significant for either measure of palatability ($ps > .18$).

To confirm that the group differences found on the 15-min taste test trials were genuine conditioning effects specific to the CS rather than a general elevation in fluid intake occasioned by the increased duration of the trials, statistical analyses were conducted on performance from the two 15-min water trials that intervened between the taste trials (see Table 1). For volume consumed the ANOVA revealed a small (~1 ml) but nonetheless significant main effect of Trial, $F(1, 14) = 6.37$, $p < .05$, but no significant main effect of US or US X Trial interaction ($ps > 0.40$). There were no significant main effects or interaction for lick cluster size or initial lick rate ($ps > 0.10$) during the two 15 min water trials that intervened between the two taste only test trials.

With regard to the absence of a group difference in cluster size during the 5-min trials of this experiment, we note that one outlier was responsible for the across-trial increase in cluster size for Group hNaCl; cluster size decreased over these trials for the other rats in this group. However, on the 15-min test trials, where more samples of clusters are obtained, there was a reduction in individual variability and a significant group difference in taste palatability was exposed. Given the between-subject variability that can occur in lick pattern analysis (also noted in [59]), we advocate the use of larger groups when short-duration trials are employed.

In future research it will be important to determine whether the critical between-experiment difference was the reduction in CS duration or the shorter CS-US delay. However, irrespective of the resolution of this issue, we interpret the overall pattern of significance on the taste test trials as support for the view that, given appropriate parameters, hypertonic NaCl is an effective US that causes a reduction in the palatability of the associated taste CS. As with the gallamine US in Experiment 1, we take the reduction of palatability of the saccharin CS as evidence that hypertonic NaCl can induce CTA.

4. General Discussion

The current study sought to investigate whether the conditioned taste suppression induced by internal pain-inducing agents is qualitatively different from that found with illness-based USs (e.g., LiCl). Using lick pattern analysis, both gallamine (Experiment 1) and hypertonic NaCl (Experiment 2) suppressed CS intake and, importantly, also reduced the size of the lick clusters as well as the initial rate of licking directed at the associated tastant (i.e., these USs each caused a conditioned reduction in taste palatability). As defined in the Introduction, CTA acquisition involves a reduction in palatability. Thus, we interpret the current results as evidence that these internal pain-inducing agents, similar to a LiCl US, support the acquisition of CTAs rather than conditioned taste avoidance.

It is worth noting that hypertonic NaCl did not always induce CTAs. That is, although no evidence of suppression was revealed in Experiment 1 when the inter-stimulus interval was 5-min (the functional CS-US interval was probably longer because rats normally stopped drinking before the end of 15-min CS trial), this pain-inducing US did support CTA when the inter-stimulus interval was reduced to 2-min in Experiment 2. This finding reminds us that the optimal parameters for CTA acquisition can vary as a function of the nature of the USs (for a review see [54]). In the case of hypertonic NaCl, the pain immediately following US injections required multiple trials and greater CS-US contiguity to induce a CTA. As suggested by these results, perhaps it is not too surprising that some toxins (e.g., malonate, strychnine; [37,44]) are considered ineffective in generating CTAs because the experimental parameters, such as a single conditioning trial or inter-stimulus interval, may not have been optimal.

Because gallamine and hypertonic NaCl can support CTA acquisition without causing nausea/illness, an immediate issue arises concerning how these pain-inducing USs produce CTAs. In the typical fear conditioning experiment, a stimulus (e.g., tone or light) is paired with a peripheral pain US (e.g., footshock), and the CS evokes behavioral and autonomic responses that are motivated by conditioned fear (e.g., [24,28,38,46]). Could conditioned fear be evoked by an internal pain-paired taste CS that in turn reduces the hedonic value of the tastant? One of our recent findings provides some support for this hypothesis. Lin, Amodeo, Arthurs, and Reilly [41] examined whether palatability changes as a function of taste novelty/familiarity. Taste neophobia is manifest as a reduction in consumption of a novel tastant because of fear of the unknown post-ingestive consequences that might, of course, be fatal [6,7,13]. In the absence of consequent illness, the initial wariness toward the new tastant habituates such that it is now perceived as safe and familiar. Lin, Amodeo et al. [41] found that in addition to an increase in the amount consumed, recovery from neophobia was also accompanied with a concomitant elevation in lick cluster size (i.e., palatability increased as neophobia habituated). This result encourages the view that fear, occasioned by a novel tastant, can modulate (i.e., reduce) taste palatability. Building on this analysis, we entertain the view that conditioned fear can devalue the pain-associated tastant. If this hypothesis is correct, then one might predict that anxiolytics (e.g., chlordiazepoxide) would attenuate the acquisition of pain-induced CTAs by reducing the severity of the perceived pain induced by the US, as that found in studies using a conditioned suppression paradigm with a light or tone CS and a footshock US (e.g., [62]). This line of reasoning might be taken one step further. If CTAs can be induced by nausea/illness as well as an internal pain-inducing USs then one might predict a double dissociation in terms of the influence of anti-emetic and anxiolytic treatments. That is, a CTA induced with LiCl might be reduced by an anti-emetic (e.g., ondansetron) but unaffected by an anxiolytic (e.g., chlordiazepoxide), whereas the opposite outcomes might be expected for a CTA induced with an internal pain-inducing US like gallamine.

Using lick pattern analysis, it has been demonstrated that CTAs (defined by a reduction in the palatability of the associated taste CS) are not only induced by USs that cause nausea/illness (e.g., LiCl; [3,4,27,40]) but also can be acquired when pain-inducing agents (gallamine and hypertonic NaCl in the present report) serve as the US. The CTA mechanism protects animals against the ingestion of a wide range of toxins that presumably can have numerous modes of actions and disparate pharmacological effects. To be effective, then, this mechanism must be very broadly tuned – it is better to make a misattribution about the toxicity of a food (that, at worst, mistakenly limits feeding options) than to fail to make a correct attribution that might have fatal consequences. Pain-inducing USs, at appropriate doses, have severe internal aversive consequences that, given appropriate parameters, can, as the present study shows, cause a reduction in the palatability of the associated tastant. A feral animal that is experiencing severe internal pain consequent to the ingestion of a novel food is in a highly vulnerable condition that might compromise survival. A mechanism that ensures the animal will avoid consumption of the food that caused such pain would be highly beneficial. We believe that the broadly tuned CTA mechanism serves this function.

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Research Highlights

Gallamine and hypertonic NaCl cause internal pain but not nausea/malaise.

Gallamine and hypertonic NaCl reduced palatability of the associated tastant.

Internal pain causes CTAs comparable to those based on emesis-related agents.

Figure 1.

Experiment 1: Mean ($\pm SE$) conditioned stimulus (0.1% saccharin)-directed performance across the five conditioning trials and one taste only test trial in rats receiving contingent injections of isotonic NaCl (Control), lithium chloride (LiCl), gallamine, or hypertonic NaCl (hNaCl). Note that due to rapid learning, the LiCl Group received two conditioning trials followed by four taste only trials. A: Intake; B: Cluster size; C: Initial lick rate.

Figure 2.

Experiment 2: Mean ($\pm SE$) conditioned stimulus (0.1% saccharin)-directed performance across the four 5-min conditioning trials (Panels A, B and C) and the two 15-min taste only test trials (Panels D, E and F) in rats receiving contingent injections of isotonic NaCl (Control) or hypertonic NaCl (hNaCl). Panels A & D: Intake; Panels B & E: Lick cluster size; Panels C & F: Initial lick rate.

Table 1

Data, mean ($\pm SE$), for intake, lick cluster size, and initial lick rate from the two 15-min water trials that intervened between the two 15-min taste only test trials in Experiment 2 for the Control subjects injected with isotonic NaCl and the rats injected with hypertonic NaCl (hNaCl).

Group	Intake	Lick Cluster Size	Initial Lick Rate
Control			
Water 1	13.70 (0.58)	106.21 (23.73)	1034.50 (37.53)
Water 2	15.04 (1.17)	103.78 (11.54)	1038.13 (45.80)
hNaCl			
Water 1	15.49 (0.78)	165.34 (46.93)	1111.00 (41.97)
Water 2	16.21 (1.45)	107.24 (18.77)	1080.00 (38.45)