

Studies on the sensory perception and oral function  
of aversive stimuli in food

-Abstract version-

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## Abstract

Oral sensations significantly influence eating behavior. Humans perceived tastes and somatic sensations in the oral cavity, and regulate their ingestion based on these stimuli. The rejection of bitter-tasting foods is essential for survival because many toxic compounds actually taste bitter. Therefore, a bitter taste is generally considered as a harmful signal to avoid the ingestion of a potentially toxic compounds. Naive mammals also innately express aversive responses to astringency, similarly bitter taste. Therefore, bitterness and astringency are typical aversive sensations. On the other hand, some components of bitterness and astringency make key flavor contributions to promote the palatability of foods and beverages such as beer, coffee and wild vegetables, as noted in adults. These contradictory behaviors suggest that humans decide whether to accept or reject the taste of bitter and astringent compounds based on certain criteria, and tolerate some components of these taste. However, the mechanism underlying the taste acceptance of aversive stimuli remains unclear. Therefore, in the present study, the factors that contribute to taste acceptance of aversive stimuli were explored from different perspectives depending on the target aversive stimuli. To discover clues to help clarify taste acceptance for bitterness, I performed tests to compare taste tolerance among various bitter compounds in humans and mice. Habituation mechanisms were explored based on the changes in tolerance associated with prolonged exposure to bitter stimuli in mice. Regarding taste acceptance of astringency, the study focused on dietary habits paired with the consumption of astringent beverages, and explored the oral functions with respect to astringent beverages during the course of a meal.

In chapter 1, the study investigated human taste tolerance and sensory cues leading to diverse taste tolerance of bitter compounds. Taste tolerance of eight bitter compounds, which are typically contained in foods, was evaluated by measuring detection and rejection thresholds. Tolerance was defined as the ratio of group rejection threshold to group detection threshold that

represents the hedonically acceptable range of the suprathreshold of tastants to evaluate differences in the acceptability of the taste of bitter compounds. The results revealed that the level of tolerance of each compound was variable. Caffeine and L-isoleucine showed a relatively wider acceptable range of the suprathreshold, suggesting that the taste of compounds promotes the palatability of foods and beverages at a concentration between detection threshold and rejection threshold. However, the acceptable range does not exceed 3-fold the detection threshold, suggesting that it is necessary to exercise caution when developing products containing bitter compounds for young adults. Secondly, the study focused on the association between tolerance and temporal characteristics. Two types of time-intensity profiles, indicating attenuation and accumulation characteristics of bitterness, of four bitter compounds were evaluated to examine the association between tolerance and temporal characteristics. However, tolerance did not depend on the attenuation and accumulation characteristics of bitterness. In addition, nutritive value, bitter taste receptors types and the slope of the dose-dependent preference curve did not contribute to the difference in tolerance. These results suggest that the criteria controlling tolerance of bitter compounds may be derived from a complex relationship between the taste quality and cognitive process.

In chapter 2, the study investigated effects of prolonged exposure to a bitter compound on both recognition and rejection behaviors to the same compound in mice. Paired measurements of rejection ( $R_jT$ ) and apparent recognition ( $aR_cT$ ) thresholds were made using brief-access two-bottle choice tests before and after taste aversion conditioning, respectively. First,  $R_jT$  was much higher than  $aR_cT$  for the bitter amino acids, L-tryptophan and L-isoleucine, which mice daily taste in their food, indicating strong acceptance of those familiar stimuli within the concentration range between  $R_jT$  and  $aR_cT$ . Next, I tested five other structurally dissimilar bitter compounds, to which mice were naive at the beginning of the experiments: denatonium benzoate, quinine-HCl, caffeine, salicin and epigallocatechin gallate.  $R_jT$  was moderately higher than  $aR_cT$  for all the compounds tested,

indicating the presence of innate acceptance to these various, unfamiliar bitter stimuli in mice. Lastly, a 3-week forced exposure increased R<sub>j</sub>T for all the bitter compounds except for salicin, demonstrating that mice acquire tolerance to a broad array of bitter compounds after long-term exposure to them. These results using mice reveal that bitter acceptance is a general response to low concentrations of bitter substances and tolerance to bitter stimuli can be induced by long-term presentation, although the underlying mechanisms remain unknown. The study also provides preference data for two amino acids, L-tryptophan and L-isoleucine, that have not been tested in mice, suggesting that mice find them as bitter as humans do because they evoked aversive behavioral responses and, similarly to many other known bitter stimuli, stronger nerve responses in the glossopharyngeal nerve than in the chorda tympani nerve have been reported in mice. Because this study was limited to female mice, further studies are necessary to extend these findings to male mice. This work is an initial step toward understanding how bitter tolerance develops in mice, and possibly mammals in general.

In chapter 3, the present study was conducted to identify key components in oolong tea that attenuate oral fat sensations. I showed through a sensory study that oolong tea consumption significantly reduces oral fat sensations as compared with water. According to the correlation analysis, oral fat sensations negatively correlated to astringency, suggesting that oral fat sensations is attenuated by the stimulation of astringency. Interfacial tension was measured to evaluate the emulsifying properties of teas and its components. The interfacial tension values for oolong tea and tea-leaf saponins were significantly lower when compared with other teas and major tea components. Furthermore, an emulsion made from tea-leaf saponins and corn oil was stable after 24 h. Moreover, polyphenol enhanced oolong tea showed significantly decreased oral fattiness when compared with oolong tea. Thus, tea-leaf saponins appear to be key components in reducing oral fat sensation. Given these results, two factors explaining the effectiveness of the pairing oolong tea with fatty food

were identified. The first factor is the perceptual cleaning effects that astringent stimuli have to attenuate the texture of fat based on tribology in the mouth. The second factor is the physical cleaning effect, in which oolong tea helps to remove fat from the mouth due to its marked emulsifying properties. This background may help determine the pairing of oolong tea and fatty food during meals. By both perceptual and physical elimination of excess fat sensations due to the continuous consumption of oolong tea during meals, oolong tea may exhibit a “refreshing effect” when consuming fatty foods.

In the present studies, it appears that taste tolerance of bitter compounds is variable in each compound in humans. It also appears that tolerance is independently modulated to the attenuation and accumulation characteristics of bitterness, nutritive value, bitter taste receptor types, or slope of the dose-dependent preference curve. As the results of mice behavior studies, it reveals that bitter acceptance is a general response to low concentrations of bitter substances and tolerance to the bitter stimuli can be induced by long-term presentation. Although the underlying mechanisms of tolerance of bitter compounds to be determined, two separate pathways whether to be associated with or without sensory adaptation are suggested. Regarding taste acceptance of astringency, two oral functions which are “the perceptual cleaning effects by astringent stimuli” and “the physical cleaning effects” are suggested to contribute the palatability of astringent beverages during the course of a meal.