Monell Chemical Senses Center

Annual Progress Report: 2012 Formula Grant

Reporting Period


Formula Grant Overview

The Monell Chemical Senses Center received $234,200 in formula funds for the grant award period January 1, 2013 through December 31, 2013. Accomplishments for the reporting period are described below.

Research Project 1: Project Title and Purpose

*Early Stage Recordings of Cognitive Odor Processing* – The purpose of this project is to establish a novel measure of olfactory responses occurring at the first input to the human brain, the olfactory bulb (OB). This innovation would not only enable further explorations of the role fulfilled by the olfactory bulb in the cognitive and perceptual processing of odors but would also be easily implemented as an everyday clinical tool. A technique allowing measures of human OB signals will greatly aid future olfactory-related translational work and establish a new paradigm for studies of human olfactory processing.

Anticipated Duration of Project

1/1/2013 – 12/31/2013

Project Overview

The goal of the proposed research is to establish in humans the first non-invasive measure of olfactory bulb (OB) responses to odor stimuli. We hypothesize that signals obtained via recordings from extranasal electrodes at the nasal bridge represent responses from the olfactory bulb (OB), so-called Electro-Bulbograms (EBG). If our hypothesis is confirmed, this innovation would not only enable further explorations of the role fulfilled by the OB in the human olfactory system, but would also be easily implemented as an everyday clinical tool. The specific aims of this project are to determine whether the recorded putative EBG signal originates from the olfactory receptors by using paradigms demonstrated to clearly alter OB responses, but not olfactory receptor responses, in other animals. Moreover, we will determine whether the putative EBG signal originates from the OB or from cortical structures by using paradigms demonstrated to alter signal in piriform cortex but not the OB. The specific aims of this project are:

**AIM 1** - To test the alternative hypothesis that the Electro-Bulbogram (EBG) response originates from receptor processing. Nutritional state and food/non-food odor distinction have both been demonstrated to alter the signaling of the OB in non-human animals, but no effect has been
demonstrated for receptor firing. We will test the hypothesis that the OB signal in response to a food odor is modulated by the individual’s nutritional state. In two separate sessions, subjects will be exposed to either ‘food associated odors’ or ‘non-food associated odors,’ or be hungry or satiated. Based on the literature demonstrating nutritional state-dependent modulation of the OB signal, we expect to find a clear difference between the two odors, as well as nutritional state, where differences would indicate an OB signal. A lack of significant differences would suggest that the signal originates from olfactory receptors.

AIM 2 - To test the alternative hypothesis that the EBG response originates from cerebral processing. Human olfactory cortex is known to demonstrate rapid habituation to repeated odor exposure whereas the OB displays no reduced firing. Thus, a significant decline of the signal due to repetitive odor presentation would indicate that the OB signal has a major cortical source. Signals will be recorded during two sessions on two consecutive days; a ‘Response session’, during which odor will be delivered using long inter-stimulus-interval (ISI), and a ‘Habituation session’, during which odors will be delivered using short ISI. We hypothesize that: 1) there will be no significant difference in the OB response between the two sessions, 2) there will be a significant and clear difference for all scalp components between sessions in that all peak amplitudes from the Habituation session will be either reduced or non-existent.

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Expected Research Outcomes and Benefits

The proposed work will establish a novel, non-invasive measure of human olfactory bulb (OB) responses to odor stimuli, and allow assessment of neural responses from one of the early stages of human olfactory processing using an inexpensive, non-invasive, and temporally-precise recording method. For the first time, we would be able to acquire data from the complete human olfactory system. If established, these measures would enable detailed investigations into the role of the OB in a wide variety of clinical disorders known to affect olfactory processing, such as neurodegenerative (Alzheimer’s disease, Parkinson’s disease, and multiple sclerosis) and eating disorders as well as schizophrenia, to mention only a few.

Summary of Research Completed

The milestone for this reporting period is:
AIM 1 (Study 1) will be completed. The scientific outcome will be to define the EBG response and to exclude the possibility that the recorded response originates from receptor or other external responses.

Aim 1 consisted of one experiment. Prior to the experiment we conducted two pilot studies to select and optimize odor stimuli for the experiment. In this progress report, we are reporting complete data from two separate pilot studies and partial data (14 out of 20 subjects analyzed) from the experiment.

In Pilot Study 1, we determined iso-intense odor concentrations of two odors that previous pilot studies (performed under previous NIH grants and therefore not reported within this progress report) identified as being perceived as being food odor related. In Pilot Study 2, we determined that our paradigm were able to produce clear scalp event-related potentials (ERPs) as well as clear responses from the putative olfactory bulb electrodes.

Pilot Study 1

Aim: In Pilot Study 1, we assessed whether the two target odors we selected were perceived as iso-intense and iso-pleasant by our common testing population.

Methods: A total of 14 healthy individuals (8 women; mean age 22.3) participated in Pilot Study 1. The two odors selected were both complex odor mixtures to account for odorant-dependent effects that the olfactory bulb is known to be sensitive for. Because Experiment 1 modulates nutritional state (hungry vs. satiated) as independent variable, the two odors also varied in nutritional load. One of the two odors is commonly perceived to originate from a low energy food source (orange) whereas the other odor is commonly perceived to originate from a high energy food source (peanut). The orange odor consisted of 30% volume/volume (v/v) concentration of orange extract (Orange Oil, Sigma Aldrich) diluted by mineral oil (Sigma Aldrich) and the peanut odor consisted of 35% v/v concentration of peanut extract (roasted peanut odor, Takasago Inc.) diluted in mineral oil (Sigma Aldrich); each odor was presented using amber 150 milliliter (ml) glass bottles with a total of 10ml in each. A total of 14 participants rated the two odors for perceived intensity and pleasantness using computerized visual analogue scales ranging from ‘Very Weak’ to ‘Very Intense’ for the intensity ratings and ‘Very Unpleasant’ to Very Pleasant’ for the pleasantness ratings. Each participant provided both ratings for each odor four times and the geometric mean of their ratings were used for statistical analyses. Ods were presented using a fully automatic, computer operated, olfactometer with a known high temporal processing (20ms odor rise-time) using a total flow of 3 liters per minutes (l/m). Each odor presentation lasted for a total of 1 second. Statistical differences were assessed using repeated-measures analyses of variance (ANOVA).

Results: There were no statistical differences between the two odors samples in either their perceived intensity \[F(1,13) = 1.25, \text{ p ns.}\] or their perceived pleasantness \[F(1,13) = 1.9, \text{ p ns.}\]. These data demonstrate that the odors we aim to use are good stimuli in respect of perceptual values.
Pilot Study 2

Aim: In Pilot Study 2, we determined that our paradigm was able to produce clear scalp event-related potentials (ERPs) as well as clear responses from the putative olfactory bulb electrodes. The former was assessed to validate our stimulus section and temporal presentation because scalp ERP recordings are known to be very sensitive to presentation methods with poor temporal accuracy and the latter was assessed to assure that our dependent measures performed well.

Methods: A total of 5 healthy individuals (4 women; mean age 24.3) participated in Pilot Study 2. Olfactory bulb responses and EEG signals were simultaneously and continuously recorded with an Active-Two system (BioSemi, Amsterdam, NL) using 32 active electrodes. The olfactory bulb response was recorded from 4 additional active electrodes placed slightly above and at the extension of the eyebrows. Signals were recorded with a sampling rate of 512 hertz (Hz) and analog filtered (0.06 and 100 Hz). All data were pre-processed using EEGLAB, a MatLab signal analyses toolbox, and data were segmented into epochs from -500 to 1000 milliseconds (ms), relative to the onset of the odor stimulus, determined using a photon ionizing detector. Extended infomax independent component analyses were applied to the concatenated single trials, and independent components representing common electroencephalography (EEG) artifacts, such as eye blinks, were identified and removed. Data were then re-referenced to the average mastoid signal, the baseline (-200 to 0 ms) was subtracted, and a 30 Hz low-pass filter was applied. Subsequently, evoked responses were computed by averaging all valid responses using a standardized algorithm within the EEGLAB software. Valid deflections in the signal were identified using multiple-comparisons-adjusted t-tests, per sampling-point, against the known baseline. Differences between conditions were assessed with adjusted t-tests of peak amplitudes. For the olfactory bulb responses, the four electrodes were merged into one response of interest to account for variations in individual electrodes. For the scalp ERP analyses, ERPs for single electrodes were merged into three regions of interest (ROIs), namely, Anterior, Central, and Posterior ROIs, to avoid a loss of statistical power and reduce variance.

Identical odors as presented above for Pilot Study 1 were used. Similarly to what was described above for Pilot Study 1, odors were presented using the same olfactometer with the same settings.

Results: The analyses of the olfactory bulb component demonstrated that our paradigm could indeed invoke a clear response that is not co-located in time with the scalp ERP response (Figure 1). Moreover, the analyses of the scalp ERP component over the electrode cluster centered over the Pz electrode, electrode positions known in past studies to display the most robust odor related ERP response, indicated that our stimulus paradigm could readily produce clear ERP responses (Figure 2). Taken together, this pilot study confirms that our methods and stimulus presentation paradigms are sound and are able to reliably detect our responses of interest.

Experiment 1

Aim: In Experiment 1, we tested our alternative hypothesis that the putative olfactory bulb responses originate from receptor processing. This alternative hypothesis is based on the basic assumption that nutritional state is not able to modulate odor receptor responses.
Methods: A total of 14 healthy individuals (7 women; mean age 21.9) have participated in Experiment 1 as of 6/30/2013. Olfactory bulb responses and EEG signals were simultaneously and continuously recorded as described above with the exception that measures were obtained in two separate sessions while participants were exposed to the two separate odors in each session; each odor was repeated a total of 40 times. In the nutrition-deprived session, participants commenced testing at 8am and were not allowed to ingest nothing but water from 8pm through the start of testing 12h later. In the satiated session, a standardized breakfast, based on caloric intake per measured body weight, was served shortly before EEG preparations begin (attachment of electrodes, etc.). The order of sessions was counterbalanced. All other measures, settings, and analyses were according to what is described above for Pilot Study 2.

Results: We initially analyzed whether there were any nutrition-based differences in perceived intensity or pleasantness of the odor stimuli. There were no nutrition-based difference in either perceived pleasantness or perceived intensity, as assessed by repeated measures of ANOVA [F(1,13)=1.4991, p=0.24 and F(1,13)=1.3316, p=0.26, respectively], see Figure 3. These results mean that differences in our electrophysiological measures cannot be directly explained by perceptual differences in our stimuli.

We thereafter assessed whether there was a difference in the olfactory bulb response between the two nutritional states. These analyses demonstrated that there was a clear, significant difference in the response between nutritional states (Figure 4). To date, our measures conclusively support our a priori assumption that nutritional state would modulate the putative olfactory bulb responses, thus being in line with the assumption that these signals are indeed measures of true olfactory bulb responses.
Figure 1: Putative olfactory bulb responses, sampled from the four ‘bulb’ electrodes, in Pilot Study 2. Data from the two odors combined for maximum power. Note the large negative deflection around 200ms after odor onset, a time window where odor ERPs are not commonly developed.

Figure 2: Scalp ERP response, sampled from the Pz electrode cluster, from the two odors combined in Pilot Study 2. Note the large positive deflection around 620ms after odor onset typical for odor-evoked ERPs.
**Figure 3:** Average perceptual ratings (n=14) of perceived odor intensity and pleasantness within Experiment 1, divided per nutritional state. Error bars in graph represents standard-error of the means. There were no statistical differences between the two nutritional states for either perceived intensity or pleasantness.

![Graph showing average perceptual ratings of odor intensity and pleasantness](image)

**Figure 4:** A) Putative olfactory bulb responses, pooled over the four ‘bulb’ electrodes, in Experiment 1. Data from the two odors combined for maximum power and each nutritional condition displayed separately. Note the large difference in negative deflection around 200ms after odor onset, a time window where odor ERPs are not commonly are displayed. B) Mean olfactory bulb response amplitude displayed for each nutritional condition separately. Error bars in graph represents standard-error of the means. Blue bar and line indicates mean response during testing session when participants felt satiated and red bar indicates mean response when participants felt hungry.

![Graph showing olfactory bulb responses](image)
Research Project 2: Project Title and Purpose

Bitter Taste-Induced Nausea – Bitter taste stimuli can directly elicit nausea. Usually nausea is the negative experience that accompanies toxin-induced illness. The problem lies in that some toxins are also medicines and their induced nausea is a disincentive to medical compliance, especially among children who cannot ingest encapsulated drugs. Moreover, a wide variety of gastrointestinal disorders are frequently accompanied by bitter taste reflux from gastric contents and by chronic nausea. The proposed work will determine the extent to which bitter taste evokes nausea in healthy people. This knowledge in a normative state might lead to new preventions for patients suffering from nausea due to gastric reflux or direct stimulation of bitter taste.

Anticipated Duration of Project

1/1/2013 – 12/31/2013

Project Overview

Bitter taste is the evolutionary marker of poisoning, which in turn is a prime elicitor of nausea. Thus we are asking whether bitter taste is a privileged inducer of nausea. The proposed work will determine (1) the extent to which bitter stimuli evoke nausea in healthy people, and (2) the role of perceived bitterness in nausea elicitation. In other words we will determine whether we can separate bitter perception from nausea elicitation. Bitter stimuli may indeed be able to stimulate toxin receptors that contribute to nausea in the absence of bitterness perception. In Aim 1, we will test whether different bitter tasting compounds are equally nauseating when intensity matched. Subjects will be presented with strong bitter taste stimuli in an oral stimulation protocol without swallowing. Nausea will be measured by self-assessment on a modified Muth Nausea Profile (MNP) questionnaire and by the physiological measures of electrogastrography (EGG). Four bitter solutions (SOA, denatonium benzoate, quinine sulfate (QSO4) and Phenylthiocarbamide (PTC)) will be tested for their nauseogenic effects, and compared to an equi-intense control solution (NaCl).

In Aim 2, we will test whether highly concentrated but weakly perceived bitter compounds elicit nausea to determine whether we can separate conscious bitter perception from the nausea eliciting effects of stimulation. We will manipulate perceived bitterness independently of concentration either by peripheral inhibition (using a bitter taste inhibitor: sodium glutamate) or by cognitive suppression (using mixtures of sweeteners and bitter tasting compounds). The same protocol and measurements as in Aim 1 will be used.

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Expected Research Outcomes and Benefits

This work has important clinical relevance. These data will help determine the basic protective physiological function of bitter taste and its elicitation of nausea as a warning against subsequent ingestion of noxious stimuli and spoiled foods. Since nausea can be a debilitating side-effect of many cancer therapies and drug treatments, and because patients with gastroparesis suffer nausea chronically as a consequence of their disorder, an improved understanding of normative nausea reflexes will provide insight into the cause of these nausea side effects and pathologies. Many diabetic patients experience gastroparesis caused by excessive blood sugar, which inhibits gastric motility via normal feedback reflexes. The bitter gastric reflux resulting from gastroparesis is believed to be a cause of their chronic and sometimes debilitating nausea. The data obtained from this work may benefit these patients directly as we may be able to single out people at higher risk for nausea side-effects, to determine whether certain food experiences are likely to exacerbate low level ongoing nausea, and to advise vulnerable patient populations to avoid certain types of foods that are strongly bitter.

Summary of Research Completed

For the funded period of 1/1/2013-6/30/2013 in our proposal we anticipated that the purchase and staff training of the new equipment (Electrogastrograph) would be completed, that pilot studies would be finished and that recruiting and testing of Aim 1 study would be under way. We are pleased to say that we are on schedule.

We acquired a Research 101 Electrogastrography Machine from the company 3CPM, based in Maryland. An instructor from the company came to Monell and trained the research staff. By the end of the fiscal year (June 30), we had recruited 26 participants (pilot studies + Aim 1 study). Among those 26 participants:
7 subjects completed the pilot studies.
6 subjects completed Aim 1 study.
1 subject was being tested for Aim 1 study.
10 were excluded from the studies after the first session (construction of psychophysical curves) as their taste functions did not show monotonic increases of taste intensity with increasing concentration.

Pilot studies:
The Aim 1 study investigates whether different bitter tasting compounds are equally nauseating when intensity matched. Whereas Aim 2 study will test whether the same highly concentrated bitter compounds are still nauseogenic when weakly perceived, due to the addition of compounds that interfere with bitter taste perception. In order to be able to address both aims, we needed first to determine the correct parameters (time of oral exposure and compound concentrations) to establish which bitter stimuli would be nauseating when intensity matched (Aim 1) and we need
to establish how to decrease perceived bitterness via the use of blockers or masking tastes (Aim 2).

In a previous study we were successful at inducing nausea in some subjects with a solution of 0.8 mM SOA (Sucrose Octaacetate) presented in 6 medicine cups containing 10 mL of solution each. The solution in each cup was slowly swished around for 30 seconds then expectorated for a total of 3 min oral exposure. This was therefore our starting point to develop the protocols of this proposal.

We first conducted informal testing in the lab to match the bitterness of 0.8 mM SOA with the 3 other bitter compounds. We found that 4 μM denatomiun, 3 mM PTC (Phenylthiocarbamide) and 1.5 mM quinine were approximately equi-bitter to 0.8 mM SOA. At those concentrations, the bitterness intensity of the taste compounds could be decreased by the presence of a mixture of bitter blockers (15 mM Zinc Sulfate + 50 mM MSG (mono sodium glutamate)) or 2 M sucrose but was still moderately perceived. At lower concentrations, but still high taste intensity, the perceived bitterness could be more efficiently inhibited. Another set of equi-bitter solutions with lower concentrations was then established: 0.4 mM SOA, 1 μM denatomiun, 1.5 mM PTC and 0.75 mM quinine.

**The design of the pilot study was as follows:**
First session: construction of psychophysical curves with the stimuli and concentrations listed in Table 1
Test 1: testing of one bitter compound for its nauseogenic effects
   Taste compound exposure: lower concentration, 6 cups (= 3 min exposure)
Test 2: testing of the same bitter compound for its nauseogenic effects
   Taste compound exposure: lower concentration, 20 cups (= 10 min exposure)
Test 3: testing of the same bitter compound for its nauseogenic effects
   Taste compound exposure: higher concentration, 6 cups (= 3 min exposure)
Test 4: testing of a negative control (water or 0.4 mM NaCl (sodium chloride)), 6 cups
Tests 1, 2, 3 and 4 were conducted in random order and in separate sessions. Sessions were scheduled with a minimum of 3 days apart.

7 subjects participated in the pilot study. Each subject tested only one bitter compound. Thus, each bitter compound was tested by two subjects, except PTC (only one). For the negative control (Test 4) 3 subjects were presented with 0.4 mM NaCl and 4 subjects were presented with water. Like in our previous study, the solution in each cup (10 mL) was slowly swished around for 30 seconds then expectorated. For the short exposure tests (6 cups/3 min) the subjects also gargled the first two cups for 5 seconds. For the long exposure (20 cups/10 min) the subjects gargled the solution every 5th cup for 5 seconds.

The nauseogenic effects of the stimuli were measured by self-assessment on a modified Muth Nausea Profile (MNP) questionnaire and by the physiological measures of electrogastrography (EGG) throughout the session. At the end of the sessions, subjects were asked to fill out a MNP questionnaire and to rate the perceived taste intensity of the stimuli they tested.
Results of the psychophysical curves for the taste stimuli (Figure 1):
The curves of total taste intensity were constructed for the compounds of interest to enable us to deliver stimuli that are in the same perceived intensity range for all subjects. They also helped to identify and exclude from the study the subjects who were very weakly sensitive or unable to correctly judge the taste intensity of the compounds of interest.

The obtained psychophysical curves indicated that for all the taste stimuli of the study (4 bitter compounds and NaCl), the “lower concentrations” solutions (corresponding to exposure/cup #5 in the psychophysical curves) were all perceived by the participants as being strong to very strong and thus also confirmed that at those concentrations the taste stimuli are approximately matched in intensity.

Results of the MNP questionnaire and taste intensity ratings after oral exposure of a strong taste stimulus (Figure 2):
The MNP questionnaire asks participants about the perception of common nausea symptoms (15 symptoms scored from 0 to 9) felt during and after the exposure to the stimuli. Mean MNP ratings were taken as an index of the severity of nausea experienced.

The results show that the taste of the bitter compounds presented at higher concentrations was perceived as being slightly stronger than the taste of the same compounds presented at lower concentrations, for an identical time exposure (3 min), as well as for a longer one (10 min). However, the lower concentration bitter solutions were reported as being slightly more nauseating when exposed for a longer period in the mouth. Thus, the time of exposure, even more than the concentration level, seemed to affect the degree of self-reported nausea elicited by bitter compounds, at least when they are all perceived as being strongly bitter.

Results of the EGG after oral exposure of a strong taste stimulus (Figure 3):
EGG data were collected via electrodes placed on the abdomen skin of the subjects throughout the session (recordings of 10 min baseline, stimulus exposure period then 25 min post-exposure). Frequencies of interest recorded in the EGG are 1.0–2.5 cpm (1–2 cpm _ bradygastrias), 2.5–3.75 cpm (3 cpm _ normal range), and 3.75–10.0 cpm (3.75–10 cpm _ tachygastrias). The change in EGG power from the baseline period to the manipulation period (during and after bitter stimulus exposure) is calculated for bradygastria, normal and tachygastria EGG power. Increase in bradygastria and tachygastria (= dysrhythmia) EGG power ratio has been correlated to the elicitation of nausea in numerous studies.

The EGG results presented in Figure 3 confirm the results obtained with the MNP questionnaire: the time of stimulus exposure has a higher impact on nausea elicitation (here increase of dysrhythmia) than the stimulus concentration, when all the solutions are perceived as being strongly bitter.

We thus decided to use the equi-bitter solution set at lower concentrations with 10 min oral exposure (20 cups) for the study as these conditions appear adapted to the realization of both Aims 1 (nausea induction) and 2 (perceived bitterness reduction) of the proposal.
Note: after the pilot studies we decided not to match individually the bitterness of the 4 bitter compound solutions for each subject as mentioned in the proposal because it was too difficult for subjects to taste and compare so many strongly bitter solutions. Moreover, we observed that on average the perceived taste intensity of the 4 bitter solutions tested, as well as the NaCl solution, were rated very similarly (see these results in Figure 5 for Aim 1 study).

Aim 1 Study:
The Aim 1 study follows the pilot study protocol, but here the subjects are tested with all the taste stimuli, presented in random order, (0.4 mM SOA, 1 µM denatonium, 1.5 mM PTC, 0.75 mM quinine, 0.4 mM NaCl, distilled water) for a total of 10 min oral exposure. Six subjects completed the study so far.

Results of the MNP questionnaire and taste intensity ratings after oral exposure of a strong taste stimulus:
The results of the taste intensity ratings obtained in the study so far confirmed the results of the pilot study showing that the taste stimuli tested are intensity matched (Figures 4 and 5). Figure 4 underlines the fact that although the intensity of 0.4 mM NaCl and 1.5 mM PTC solutions were rated lower than the other solutions during the psychophysical curves construction, both were perceived as intense as the other stimuli after repeated exposures (20) during the EGG sessions. It is also worth noting that 2 subjects who would have been classified as weakly sensitive to PTC, according to the psychophysical data collected during the first session, rated the PTC solution as being strong to very strong during the EGG sessions (multiple exposures). Figure 5 displays also the MNP ratings attributed to the different stimuli, which are low for all of them. Thus, the subjects tested thus far did not report significant nausea by the stimuli tested.

Results of the EGG after oral exposure of a strong taste stimulus:
The early results presented in Figure 6 would indicate that PTC and quinine are slightly more nauseogenic than denatonium and SOA. A higher gastric activity was recorded after exposure of these first two stimuli. Oral exposure to high concentration of NaCl appears to also increase bradygastria. Nevertheless, the subjects tested thus far did not report being clearly nauseated by the taste stimuli. The on-going testing with many more subjects will clarify these first results.
Table 1: Stimuli and concentrations used for the construction of individual psychophysical curves. Each concentration was tested in duplicate.

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Figure 1: Psychophysical curves constructed during the pilot study with the stimuli and concentrations listed in Table 1.
Figure 2: Averaged MNP scores and taste intensity ratings given after oral exposure of taste stimuli (pilot study). “6xLOW” corresponds to the exposure of 6 cups of either 0.4 mM SOA, 1 uM denatonium, 0.75 mM of quinine or 1.5 mM PTC; “6xHIGH” corresponds to the exposure of 6 cups of either 0.8 mM SOA, 4 uM denatonium, 1.5 mM of quinine or 3 mM PTC; “20xLOW” corresponds to the exposure of 20 cups of either 0.4 mM SOA, 1 uM denatonium, 0.75 mM of quinine or 1.5 mM PTC; “water” corresponds to 6 cups of water and “NaCl” corresponds to 6 cups of 0.4 mM NaCl.

Figure 3: Averaged EGG power ratios for the frequencies of interest (bradygastria, normal, and tachygastria) after oral exposure of taste stimuli (pilot study). The blue numbers correspond to the averaged MNP scores given for each testing conditions. The black line indicates the level at which no change is observed between pre and post-stimulation (ratio=1).
Figure 4: Comparison of the perceived intensity of the same taste stimuli presented either during the psychophysical curve construction (rating after 1 cup (# 5&11), 10 ml, sip and spit) or during the EGG sessions (rating after 20 cups, 10 ml each, sip, swishes for 30 sec and spit, with gargle every 5\textsuperscript{th} cup) in Aim 1 study.

Figure 5: Averaged MNP scores and taste intensity ratings given after oral exposure of taste stimuli (Aim 1 study)
Figure 6: Averaged EGG power ratios for the frequencies of interest (bradygastria, normal, and tachygastria) after oral exposure of taste stimuli (Aim 1 study). The green numbers correspond to the averaged MNP scores given for each testing conditions. The black line indicates the level at which no change is observed between pre and post-stimulation (ratio=1).