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# A double-blind study of the influences of eszopiclone on dysgeusia and taste function

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ABSTRACT

Taste disturbance is a common, but poorly understood, side effect of a large number of medications. This double-blind study examined the frequency, intensity, and quality of taste disturbances related to the widely used hypnotic sleep aid eszopiclone (ESZ; Lunesta®), as well as their associations with age, sex, body mass index (BMI), time of day, phenyl thiocarbamide (PTC) taste sensitivity, and ESZ saliva and blood levels. Sixty six percent of 24 female subjects and 53% of 15 male subjects reported dysgeusic sensations, mostly bitter/metallic, during the drug administration (respective placebo figures 17% and 7%). No meaningful relationships were found between the frequency or the intensity of the sensations and age, BMI, or PTC taste sensitivity. Dysgeusia was more intense and longer lasting in women than in men, stronger in the morning than in the evening, and positively correlated with drug plasma and saliva levels. In women, intensity ratings decreased across treatment days. Taste test measures were marginally, at best, influenced by ESZ. This study demonstrates, for the first time, that the dysgeusia associated with ESZ is systemically influenced by a number of factors, including sex, time since drug administration, and both blood and saliva levels of the drug.


## 1. Introduction

Numerous drugs alter the ability to taste. In some cases, drugs produce strange tastes in the absence of a taste stimulus. Among offending medications are antimicrobial, antihypertensive, and anti-hyperlipidemic agents, including such commonly used medications as atorvastatin (Lipitor®), losartan (Cozar®), pravastatin (Pravachol®), lisinopril (Zestril®), simvastatin (Zocor®) and terbinafine (Lamisil®) (Doty et al., 2008). In the majority of such cases, taste-related side effects have been reported in less than 5% of patients. An exception is the hypnotic sleep aid eszopiclone (ESZ; Lunesta®), a short-acting non-benzodiazepine. In one double-blind placebo-controlled study, 26.1% of patients taking this medication spontaneously reported an unpleasant taste (Krystal et al., 2003). The percentage of patients taking the placebo who similarly reported an unpleasant taste was 5.6%. This specific Class IV drug differs from a number of other hypnotics in that it is among the few sleeping agents approved by the FDA for long-term use.

From a societal perspective, hypnotics are an important class of drugs, since approximately 2.5% of the United States population is prescribed such medication for the treatment of insomnia each year (Balter et al., 1992). Because of side effects, just over one fourth of this group will remain on hypnotic medication for a period greater than four months. It is not known what proportion of patients taking such drugs discontinue treatment because of taste-related side effects, although one study reported that 1.7% of subjects discontinued treatment solely for this reason (Krystal et al., 2003).

The goal of this study was to specifically identify the taste side effects associated with the use of ESZ. Among the questions asked were the following: What aberrant taste qualities are most commonly experienced in individuals taking this agent (e.g., sweet, bitter, salty, metallic, sour)? Does the frequency or intensity of such aberrant sensations change during the period of drug administration? Is such frequency or intensity influenced by body mass index (BMI), gender, or other factors? Is there a correlation between blood or saliva levels of ESZ and the presence or intensity of the dysgeusic sensations? Are quantitative measures of taste perception, such as the ability to detect or identify electrical or chemical taste stimuli, altered by ESZ? Do persons sensitive to the bitter taste of phenylthiocarbamide (PTC), a largely inherited trait, experience taste side effects related to ESZ differently than those who are not sensitive to this agent?

## 2. Materials and methods

### 2.1. Subjects

Sixteen male (mean age ± SD: 35.7 ± 12.6) and 25 female (mean age ± SD: 32.7 ± 10.63) non-smoking healthy adults served as subjects. The data from two were omitted from analysis since no evidence of ESZ was found in their blood or saliva during the ESZ treatment period, suggesting non-compliance with the study protocol. Thus, the final study group was comprised of 15 men (mean age ± SD: 35.9 ± 13.8) and 24 women (mean age ± SD: 32.8 ± 10.33). None reported having problems sleeping. Most

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were recruited from fliers displayed on kiosks at the University of Pennsylvania and Drexel University campuses. Subjects had to pass a physical examination to be eligible for enrollment. Exclusion criteria for participation included: history of chronic rhinosinusitis, liver disease, hypertension, head trauma resulting in concussion or loss of consciousness, drug or alcohol abuse, chemosensory dysfunction, or neurological dysfunction. Pregnant women, as well as individuals who operated heavy machinery or who were engaged in shift work or hazardous occupations, were not allowed to participate. Pregnancy status was confirmed via urinalysis prior to and midway through the study. Subjects gave written informed consent in accord with the requirements of the University's Office of Regulatory Affairs. This research complied with the standards of ethical research set by the 1964 Declaration of Helsinki. Each subject was paid $325 dollars for participation.

### 2.2. Experimental design

In this double-blind study, each subject received two weeks of ESZ (Lunesta®, 3 mg q.d.) and two weeks of placebo (Lunesta® minus the active ingredient ESZ) in randomized order with an interspersed week-long washout period. Questionnaires concerning the drug efficacy and presence, magnitude, and perceived quality of taste side effects were filled out on a daily basis and returned to the investigators at the end of each week. The subjects indicated whether the quality of the taste aberration, when present, fell into one or more of the following categories: metallic, chalky, bitter, salty/bitter, sour/bitter, sweet/bitter, salty, salty/sour, salty/sweet, sweet, sweet/sour, sour, and sharp. The intensity and hedonic quality of the taste side effects were established using visual analog (intensity) and category (hedonic) rating scales.

Blood and saliva samples for analysis of ESZ were obtained at the same time and day each week. Taste function was measured following the drug and placebo periods using quantitative taste tests described in the next section. Thus, aside from initial screening and orientation, each subject was in the protocol for 5 weeks (2 weeks of initial treatment condition, 1 week washout, 2 weeks of second treatment condition). Sepracor, Inc., the manufacturer of Lunesta®, provided the placebo and drug medications in a blinded fashion to the study investigators. The basic paradigm is shown for two subjects in Fig. 1.

At the beginning of the study, subjects were given a week's supply of pills. They were instructed to self-administer one pill immediately before retiring each evening. They completed a standardized questionnaire each night prior to this time which contained items related to sleep habits and incidences of potential drug side effects, most notably abnormal taste or smell sensations, that were experienced during the day.

After this initial week, the subjects returned to the Center to receive their next week's supply of pills, turn in their questionnaires, receive new questionnaires, and provide blood and saliva samples (Fig. 1). Subjects were requested to take their sleep medication in the evening within 9 to 10 h of their scheduled study visits. On the morning of their visits, they brushed their teeth before eating breakfast, and once again after eating breakfast. They were instructed not to use mouthwash on these mornings and not to eat or drink anything after breakfast until after their Center visit that morning.

### 2.3. Taste tests

Taste function was measured at the end of the drug and placebo periods using three different tests. In the *first* test, 8-cm-long strips of filter paper with 2 $cm^2$ distal sections impregnated with either sucrose (sweet; 0.05, 0.2 and 0.4 g/mL), NaCl (salty; 0.016, 0.1 and 0.25 g/mL), citric acid (sour; 0.05, 0.165 and 0.3 g/mL), or quinine hydrochloride (bitter; 0.0004, 0.0024 and 0.006 g/mL) were placed on each side of the anterior third of the tongue in a systematically counterbalanced manner. The taste strips are described elsewhere (Mueller et al., 2003). A total of 48 trials were obtained (4 tastants × 3 concentrations × 2 tongue regions × 2 repetitions). On each trial, the subject indicated whether a given stimulus tasted sweet, sour, salty or bitter by pointing to answers on a chart before protracting the tongue and rinsing the mouth with deionized water. Additionally, the perceived pleasantness/unpleasantness and intensity of each stimulus was rated on 9-point rating scales, with the extremes being "like extremely"/"dislike extremely" and "very weak"/"very strong". The *second* test employed electrogustometry (Miller et al., 2002;Loucks and Doty 2004). Small (μA) electrical currents were applied using a Rion TR-06 electrogustomer to four separate regions of the tongue via a 5 mm diameter stainless steel disk electrode. These regions were the left and right sides of the anterior (tongue tip ~ 1 cm from midline) and posterior (lateral margin ~ 1.5 cm from edge) tongue surface. The lowest current discernable from a standard (3.2 μA) was measured using a staircase threshold procedure similar to that described elsewhere for chemical taste thresholds (Doty et al., 2001). The *third* test, administered at the beginning of the last test day of week 2 and of week 5, established the "taster status" of the subjects to phenythiocarbamide (PTC). In this test, we determined whether a subject could detect the bitter taste of PTC present on a 3.80 × 1.43 cm strip of filter paper impregnated with 0.007 mg of PTC (Carolina Biological Supply Company, Burlington, NC). Detecting or not detecting this agent on the two test occasions was used as the index of PTC taster status.

### 2.4. Blood and saliva collection and analyses

Saliva samples were obtained using two methods. In the first, subjects administered a saliva collection swab (Quantisal, Immunalysis Corporation, Pomona, CA) the night before and prior to getting out of bed on each morning of their study visits (two separate samples). The evening saliva-containing tubes were refrigerated until transported to the Center by the subject the next morning. In the second, subjects provided a saliva sample by repeatedly expectorating into a 50 mL sterile collection vial when first arriving to the Center. The blood was collected by anticubital puncture upon the arrival to the Center and centrifuged at 4 °C within 30 min of collection. Both the plasma and saliva samples were stored at −70° until analysis.

| Subject | Week 1 | | Week 2 | | Week 3 | | Week 4 | | Week 5 | |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | Eszopiclone (ESZ) | Q B | Eszopiclone (ESZ) | Q T C B | Wash out | Q B | Placebo | Q B | Placebo | Q T C B |
| 2 | Placebo | Q B | Placebo | Q T C B | Wash out | Q B | Eszopiclone (ESZ) | Q B | Eszopiclone (ESZ) | Q T C B |

**Fig. 1.** Study paradigm illustrated for two subjects. Q = questionnaire return (questionnaires filled out daily); T = taste testing; C = PTC taste assessment; B = blood and saliva collection. See text for details.

To determine concentrations of ESZ in the saliva and plasma samples, 200 µL of each sample was added to 50 µL of 0.41 M citrate buffer along with 25 µL of zopiclone-$d_8$ internal standard solution. 100 µL of extraction buffer (saturated ammonium phosphate in water) and 2.0 mL of extraction solvent (1:4:5 hexanes:dichloromethane: methyl-t-butyl-ether) was added. The sample was then vortexed for a minimum of 5 min and centrifuged. The aqueous layer was then frozen in an isopropanol freezer bath and the organic layer transferred to a clean test tube and evaporated under nitrogen gas. After adding 200 µL of reconstitution solution (1.0 mM ammonium formate, 0.02% formic acid in 90:10 acetonitrile:water), the sample was vortexed and transferred to a 96 well plate. The samples were analyzed on a nonchiral high performance liquid chromatographic system equipped with a triple quadruple tandem mass spectrometer detector. Separation of ESZ from extracted matrix materials was accomplished using a Thermo-BETASIL Silica 100 analytical column (50 × 3.00 mm, 5 µm) at ambient temperature. A gradient was used starting with a mobile phase flow rate of 0.5 mL/min with 10% mobile phase A and 90% mobile phase B from 0 to 1.5 min. From 1.5 to 2.0 minutes the composition changed to 70% mobile phase A and 30% mobile phase B. From 3.5 to 3.6 min the flow rate changed to 1.0 mL/min. From 4.0 to 4.1 min the composition changed back to 10% mobile phase A and 90% mobile phase B. From 5.0 to 5.5 min the flow rate changed back to 0.5 mL/min. Mobile phase A consisted of 10 mM ammonium formate and 0.2% formic acid in water. Mobile phase B consisted of 0.5 mM ammonium formate and 0.01% formic acid in 95:5 acetonitrile:water. For the saliva samples collected at the Center, the peak area ratio of ESZ to the internal standard was used in calculating analyte concentrations in saliva using a natural logarithmic transformation of a quadratic least-squares regression algorithm. For the plasma, the same procedure was used without the logarithmic transformation. For the saliva collected at home using the Quantisal swab, a linear transformation of the quadratic least-squares regression algorithm was employed.

## 3. Results

### 3.1. Dysgeusia prevalence, quality, and intensity

Twenty-four of the thirty nine subjects (62%) in the study group reported experiencing dysgeusia during the ESZ treatment period, compared to 5 (13%) in the placebo condition (McNemar's test, $p<0.001$). The number of subjects that reported dysgeusia did not vary significantly as a function of sex ($X^2=0.69$ df = 1, $p=0.41$), PTC tasting ability ($X^2=0.60$, df = 1, $p=0.44$), age quartile ($X^2=1.73$, df = 3, $p=0.63$), or BMI quartile ($X^2=5.58$, df = 3, $p=0.13$).

Of the 24 subjects reporting dysgeusia under the ESZ condition, 8 (33%) reported a single dysgeusia quality, 10 (42%) reported two such qualities, 4 (17%) reported three such qualities, and 2 (8%) reported four such qualities. The predominant non-mutually exclusive qualities that were reported were bitter (67%; 16/24), metallic (54%; 13/24), sour/bitter (38%, 9/24), and sour (17%, 4/24). The classification of the taste quality did not change during the course of drug administration.

A sex difference was present in the number of days that dysgeusia was reported while taking ESZ. Women who experienced dysgeusia reported its presence on an average (median) of 13 of 14 days, whereas their male counterparts reported it on an average of 8.5 of 14 days (Mann–Whitney $U=28.5$, df = 1, $p=0.032$). Women also reported dysgeusia for more consecutive days than did men (13 vs 6 days; $U=30.5$, df = 1, $p=0.043$). While, overall, women rated the intensity of the dysgeusia under the drug condition higher than did men, the intensity ratings decreased significantly over the time period in which they were receiving the drug (Friedman $X^2=20.45$, $p=0.002$) (Fig. 2). This was not true for the men ($X^2=9.03$, $p=0.17$). Additionally, women experienced a dysgeusia for longer periods of the day than did men. Thus, men were more likely to rate their dysgeusia as lasting an hour or less, while women were more likely to rate their dysgeusia as lasting several hours or more ($X^2=8.9$, df = 3, $p=.031$).

A general decline occurred in dysgeusic symptoms over the course of the day. Twenty-two of the 24 (92%) subjects who experienced a dysgeusia reported experiencing it in the morning or when waking. Only 11 of those 22 subjects also reported experiencing a dysgeusia in the evening or before going to sleep. Additionally, 14 of 24 (58%) subjects specifically acknowledged a decline in their dysgeusic discomfort as the day progressed.

In their daily questionnaires, subjects were asked to localize where in the oral cavity the dysgeusia originated. In a non-mutually exclusive response array, fourteen of 24 subjects (58%) who experienced a dysgeusia said they believed the dysgeusia came from their tongue with 9 of those 14 specifically noting the back or sides of the tongue. Other popular designations were the throat (8 of 24 subjects), the roof of the mouth (9 of 24), and the saliva (9 of 24).

No meaningful correlations were present between the number of hours slept each night and the intensity, duration, or number of days of dysgeusia (Spearman $rs<0.10$, $ps>0.60$). As would be expected, subjects reported sleeping more under the drug than under the placebo condition [respective median hours (IQRs) = 8.21 (1.59) and 7.71 (1.68); Wilcoxin $Z=2.23$, $p=0.026$].

### 3.2. Relationship of dysgeusia intensity to plasma and saliva levels of ESZ

Spearman correlations were computed to determine whether plasma and saliva levels of ESZ were related to the rated intensity of the dysgeusic sensations. The coefficients are shown in Table 1, along with unadjusted and α-adjusted Bonferroni p values. Q-Saliva was the saliva collected at home in the morning using the Quantisal collection device and C-saliva was the saliva obtained at the Center. It is apparent that the intensity of the dysgeusia was related to the blood and saliva levels of ESZ and that the magnitude of the associations was stronger for week 1 than week 2. No meaningful correlations were present between the number of hours slept each night and either the plasma or saliva levels of ESZ ($rs<0.20$, $ps>0.20$).

Since the elapsed time between the collection of the plasma and saliva samples and their analysis after storage at −70 °C extended, in some cases, beyond dates for which analyte stability measures were available, we also computed correlations only for samples that fell within our known validation parameters; i.e., plasma stored up to 276 days ($n=17$) and saliva stored up to 496 days ($n=36$). No stability data were available

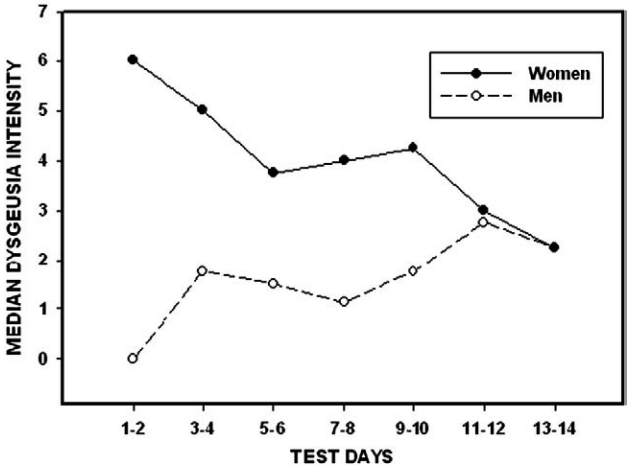


**Fig. 2.** Median intensity ratings of dysgeusia by gender in ESZ condition across test days (two-day average). The decrease in intensity ratings in women is statistically significant; the apparent increase in the intensity of dysgeusia in men is not.

**Table 1**
Spearman correlations between (a) saliva and plasma levels of ESZ sampled at the end of each of two weeks of successive drug administration and (b) measures of perceived dysgeusia during treatment weeks 1 and 2.

| Week 1 | Eszopiclone (ESZ) | | |
|---|---|---|---|
| | Plasma ($n=39$) | C-Saliva ($n=39$) | Q-Saliva ($n=39$) |
| Intensity of dysgeusia | .57<br>(.00005)<br>(.0003) | .49<br>(.0006)<br>(.004) | .50<br>(.0005)<br>(.003) |
| Week 2 | Eszopiclone (ESZ) | | |
| | Plasma ($n=39$) | C-Saliva ($n=39$) | Q-Saliva ($n=38$) |
| Intensity of Dysgeusia | .32<br>(.02)<br>(NS) | .33<br>(.02)<br>(NS) | .32<br>(.02)<br>(NS) |

Numbers in brackets indicate uncorrected (top) and Bonferroni corrected (bottom) one-tail $p$ values. $p$ values > 0.05 signified as NS. Q-Saliva is saliva collected by the subject at home using the Quantisal collection device. C-Saliva is saliva collected upon arrival at the Center. $n=39$ for all groups except the Q-saliva group for week 2, where saliva assays for one subject were corrupted, resulting in $n=38$.

for the Q-Saliva under the time and storage conditions employed in this study, although the Q- and C-Saliva levels were positively correlated (Spearman $r$s for week 1 and week 2 = 0.67 and 0.74, respectively; $p$s < 0.0001). The same general relationships as noted in Table 1 were present between dysgeusia intensity and both the blood and saliva ESZ levels for collections that fell within our validation parameters. Thus, week 1 dysgeusia intensity ratings correlated 0.76 with the ESZ plasma levels ($n=17$, one-tailed $p=.001$; Bonferroni corrected $p=0.006$), 0.55 with C-saliva levels ($n=36$, one-tailed $p=.0002$; Bonferroni corrected $p=0.001$) and 0.32 with Q-saliva levels ($n=36$, one-tailed $p=.025$, Bonferroni corrected $p$ ns). Corresponding $r$ values among the week 2 measures were 0.34 for plasma ($n=17$, $p$ ns), 0.31 for C-saliva ($n=36$, one-tailed $p=0.029$, Bonferroni corrected $p$ ns), and 0.26 for Q-saliva ($n=36$, one-tailed $p=0.057$, Bonferroni $p$ ns).

### 3.3. Taste tests

#### 3.3.1. Identification of taste quality

The number of trials on which correct identification of the appropriate quality (i.e., sweet, sour, bitter or salty) occurred for each tastant was subjected to both parametric (ANCOVA) and non-parametric (Wilcoxin signed-ranks test) analyses. The latter analyses were performed separately for each sex, as well as for the sexes combined, across the drug and placebo conditions. In the ANCOVA, the within subject factor was drug condition (ESZ, placebo) and the between subject factors were sex (male, female) and PTC taster status (yes, no). Age and BMI served as covariates. To increase the number of trials for each tastant to 12, the side (left, right) and tastant concentration (3 levels) data were combined. Preliminary analyses showed no influence of tongue side on the correct identification test measure. Since the dependent measure was skewed to the left, a square root transformation on the reflected distribution (i.e., 12.1 − number of correct trials) was performed to normalize the data. The age distribution was log transformed since the age distribution was skewed to the right. The same general findings occurred regardless of whether transformed or non-transformed data were employed.

ESZ had no discernable effect on the number of correct responses for any tastant for either the parametric or non-parametric analyses. In all cases, age was a meaningful covariate in the ANCOVAs [sucrose $F(1,33)=11.45$, $p=0.002$, $\eta^2=0.35$; NaCl $F(1,33)=6.73$, $p=0.014$; $\eta^2=0.21$; citric acid $F(1,33)=15.53$, $p=0.081$; $\eta^2=0.10$; quinine $F(1,33)=6.73$, $p=0.014$; $\eta^2=0.20$), reflecting an age-related decrease in correct responses. No meaningful influences of sex, PTC taster status, or BMI on the test measures were present.

#### 3.3.2. Intensity ratings

The drug and placebo intensity ratings for each of the four tastants were subjected to separate Wilcoxin signed-ranks tests at each tastant concentration. Additionally, the ratings were subjected to the same parametric ANCOVA as noted above, with the addition of tastant concentration (low, medium, high) as a factor. In no case was there a meaningful influence of ESZ on the intensity rating measures. Tastant concentration was a significant factor in all ANCOVA analyses [sucrose $F(2,66)=25.47$, $p<0.001$, $\eta^2=0.77$; NaCl $F(2,66)=9.64$, $p<0.001$; $\eta^2=0.29$; citric acid $F(2,66)=20.44$, $p<0.001$; $\eta^2=0.62$; quinine $F(2,66)=9.82$, $p<0.001$; $\eta^2=0.30$], reflecting the expected increase in rated intensity as stimulus concentration increased.

#### 3.3.3. Hedonic ratings

The same analyses as performed on the intensity ratings were performed on the taste hedonic ratings. As with the intensity ratings, no meaningful influences of ESZ were observed. Women rated the NaCl stimuli, overall, as less pleasant than did men [respective male and female means (SDs) = 5.28 (0.62) and 5.70 (0.59); sex main effect $F(1,33)=10.63$, $p=0.003$, $\eta^2=0.32$]. Citric acid was also rated as less pleasant by women than by men [respective male and female means (SDs) = 5.31 (0.71) and 5.85 (0.81); sex main effect $F(1,33)=4.36$, $p=0.045$, $\eta^2=0.13$]. Stimulus concentration was significantly related to the pleasantness ratings for sucrose [$F(2,66)=6.23$, $p=0.004$, $\eta^2=0.20$] and citric acid {$F(2,66)=6.13$, $p=0.036$, $\eta^2=0.11$], reflecting an increase in sucrose pleasantness ratings and a decrease in citric acid pleasantness ratings as stimulus concentration increased.

#### 3.3.4. Electrogustometric thresholds

There was no evidence that ESZ influenced the electrogustometric threshold measures. However, the threshold of most patients was at or below the lowest current that the electrogustometer could deliver (6.4 μA) relative to the comparison stimulus (3.2 μA). The number of subjects whose thresholds were at this point was essentially the same under the drug and placebo conditions, implying no drug-related deficit (i.e., elevation) in the threshold test measure (Table 2). The data were too skewed to perform parametric analyses. If one conservatively accepts 30 μA as a cut-off point between "normal" and "elevated" thresholds, then all thresholds were normal for all subjects on both sides of the anterior tongue under the drug and placebo conditions. When the left- and right-side posterior tongue data were combined, the frequency of "normal" thresholds was the same under the drug and placebo conditions (i.e., 29/39).

### 3.4. Relationship of taste intensity ratings to saliva and plasma levels of ESZ

Spearman correlations were computed between the taste stimulus intensity ratings for the highest and lowest stimulus concentrations and the week 2 saliva and plasma levels of ESZ (values closest in time to the taste testing). Although 21 of the 24 coefficients were in the negative direction (binomial $p=.0001$), implying that as circulating levels of the drug were higher when stimulus intensity ratings were lower, only three correlations were statistically significant when no correction for inflated α was made and no coefficient was statistically significant when the Bonferroni α correction was applied (Table 3). The three correlations occurred for the low concentration stimuli,

**Table 2**
Number of subjects who detected the lowest current that the electrogustometer could deliver (6.4 μA) relative to a comparison stimulus (3.2 μA) under drug and no drug conditions.

| Condition | Front of tongue | | Back of tongue | |
|---|---|---|---|---|
| | *Left* | *Right* | *Left* | *Right* |
| Eszopiclone | 35 (89.7%) | 31 (79.5%) | 15 (38.5%) | 17 (43.6%) |
| Placebo | 34 (87.2%) | 34 (87.2%) | 16 (41.0%) | 19 (48.7%) |

**Table 3**
Spearman correlations between the rated intensity of two concentrations of sweet, salty, sour, and bitter-tasting stimuli and levels of ESZ in saliva and plasma.

| Drug levels | Taste stimulus | Eszopiclone (ESZ) | |
|---|---|---|---|
| | | Stimulus concentration | |
| | | High | Low |
| Q-saliva | Sucrose | −.29 | −.45 (.0032) (ns) |
| | NaCl | −.15 | −.43 (.005) (ns) |
| | Citric Acid | .13 | −.10 |
| | Quinine | −.24 | −.27 |
| C-saliva | Sucrose | −.27 | −.31 |
| | NaCl | −.33 | −.15 |
| | Citric acid | .03 | −.10 |
| | Quinine | −.20 | −.39 (.012) (ns) |
| Plasma | Sucrose | −.13 | −.30 |
| | NaCl | −.19 | −.29 |
| | Citric acid | .23 | −.13 |
| | Quinine | −.19 | −.29 |

First number in parentheses indicates uncorrected $p$ value; ns indicates lack of significance of $p$ value after Bonferroni adjustment for multiple tests. See text for details.

which would be expected to be more sensitive to drug effects than high concentration stimuli. This concept is supported by the observation that in all cases but one the negative correlations were larger for the low than for the high stimulus concentration (binomial $p = 0.003$).

## 4. Discussion

This study is the first to specifically evaluate the nature of the dysgeusia associated with ESZ and, to our knowledge, the first to demonstrate an association between the intensity of any drug-related dysgeusia and saliva and blood drug levels. It is also the first to examine the relationship between quantitative measures of taste function and such drug concentrations. Its findings accord well with suggestions that many taste-related drug side effects are likely mediated via salivary or vascular routes (e.g., Schiffman et al., 1998; 2000a; 2000b). Importantly, its findings imply that gender may play a key role in influencing the nature and expression of drug-related dysgeusias.

The predominant dysgeusic sensations associated with ESZ were bitter and metallic, in accord with earlier, less definitive, work (Balter et al., 1992). The tendency of a few subjects to use sour in describing the ESZ-related taste side effects likely reflects the use of taste quality descriptors in a non-uniform manner, rather that meaningful variation in the elicited taste qualities. It is well known, for example, that a significant proportion of the population confuses sour and bitter taste sensations (O'Mahony et al., 1979). The reason why bitter and metallic are the predominant ESZ-related taste side effects is not known, although it should be pointed out that most medications with taste side effects produce solely or primarily these taste qualities. This includes, for example, tricyclic antidepressants, antimicrobials, anti-inflammatories, antihypertensives and antihyperlipidemics (Agro et al., 1998;Zervakis et al., 2000;Schiffman et al., 2000a; 2000b;Doty et al., 2003). It should be emphasized that, relative to other tastants, bitter-tasting chemicals exhibit an extraordinarily high level of structural diversity, making simple structure/activity relationships elusive (DuBois et al., 2008). This reflects, in part, the fact that at least 30 different types of G-protein-coupled receptors are responsible for bitter taste, in contrast to less than three for other primary taste qualities. Unlike olfaction, where a given receptor cell expresses only one type of receptor, each taste cell responsive to bitter-tasting compounds expresses many or all of the members of the T2R bitter taste receptor family (Mueller et al., 2005).

Nearly two-thirds (62%) of the participants of this study experienced dysgeusia during the ESZ treatment period, compared to 12% during the placebo period. This proportion is more than double that of a large double-blind study of the efficacy of ESZ where 26.1% of 593 subjects reported an unpleasant taste while on ESZ, as compared to 5.6% of 195 controls (Krystal et al., 2003). Two factors may explain this difference. First, and arguably most salient, the adverse taste sensations in the Krystal et al. study were spontaneously reported by study participants in the absence of specific inquiries as to taste side effects, an approach that likely underestimates the true incidence of such effects, as shown in several studies. For example, the incidence of dry mouth for extended release oxybutynin chloride based on studies in which patients are specifically asked about symptom occurrence is around 61%, compared to about half this number in clinical trials where no such question is posed (Anderson et al., 1999;Versi et al., 2000;Armstrong et al., 2005). Second, our non-smoking healthy subjects were not chosen for sleep problems, unlike the Krystal et al study, and rarely used medications, conceivably making them less acclimated to the effects of bitter-tasting drugs. However, sleep disturbances, per se, are not known to be associated with taste disturbances, and we found no association between sleep quality, in terms of hours of evening sleep, and the intensity or duration of dysgeusia.

Among those subjects who experienced a drug-related dysgeusia, women reported experiencing a more frequent and intense dysgeusia than did men, a finding not previously reported. Thus, irrespective of its quality, the dysgeusia occurred, on average, on 13 of the 14 drug days for the women, compared to only 8 days for the men. Even though the female ratings of the intensity of the dysgeusia decreased significantly over the drug period, they were above the level reported by men and even by 14 days of drug usage had not subsided to zero. It is unknown whether this phenomenon generalizes to other drugs with bitter taste side effects or if the mitigation extends beyond the time of drug administration. This sex difference was largely independent of BMI and body weight, since the effect was present even when these measures were used as covariates. Its basis is not known, although it should be noted that women have a greater sensitivity to some tastants than men (Doty 1978) and are typically more facile than men in using language for describing chemosensory experiences (Doty and Cameron 2009).

Unlike the intensity of the perceived dysgeusia, the rated intensities of the sweet, sour, bitter and salty taste stimuli were only weakly correlated with saliva or plasma levels of ESZ, albeit largely in the expected negative direction (Table 3). These weaker associations could reflect several factors. First, the function of the taste system, as measured psychophysically, may be less influenced by the drug than the expression of dysgeusia. It is well documented in clinical studies, for example, that dysgeusia often occurs in the absence of measurable changes in gustatory function (Deems et al., 1988). Second, the taste tests we employed may be relatively insensitive to subtle drug effects. The electrical thresholds were likely compromised by a basement effect, and suprathreshold sensory tests are generally less sensitive than threshold tests to the influences of age, sex, and other factors. The finding of higher negative correlations between the intensity ratings and the ESZ saliva and plasma levels at the lower than at the higher stimulus concentrations would seem to lend support to this concept (Table 3). Third, it is conceivable that the circulating levels of ESZ or its metabolites might dampen sensitivity to one or more taste qualities, thereby reducing the breadth of responses and, in turn, constricting the range of numbers upon which correlation coefficients depend. If this were the case, however, one might have expected to have seen drug-related decrements on the taste measures and somewhat smaller correlations for the bitter than the sour, salty, and sweet stimuli, none of which occurred (Table 3).

The exact time of the onset of the dysgeusia relative to the time of drug administration was not determined in this study. While most subjects reported experiencing the taste side effect on the first day of drug administration, albeit not immediately after taking the drug, some did not. In an earlier study of 10 subjects, peak plasma concentrations of a 7.5 mg dose of zopiclone occurred within an hour after drug

administration, implying rapid absorption (Caille et al., 1984). In the Caille et al study, zopiclone concentrations were found to be much higher in saliva than in plasma within 15 min of administration, implying rapid distribution and resulting in an average area under the concentration-time curve of 905.3 ng h $ml^{-1}$ for saliva compared to 387.2 ng h $ml^{-1}$ for plasma. The saliva/plasma ratio ($R_{sp}$) they obtained (2.3) was very close to that predicted on the basis of pH and the dissociation constant of ESZ by the following formula:

$$R_{sp} = \frac{1 + 10^{pKa - pH\ saliva}}{1 + 10^{pKa - pH\ plasma}} = \frac{1 + 10^{6.7-6.5}}{1 + 10^{6.7-7.4}} = \frac{2.50}{1.20} = 2.2$$

In light of the correlations between the intensity of the perceived dysgeusia and both saliva and plasma levels of ESZ during drug treatment (Table 1), it is conceivable that ESZ produces dysgeusia via both oral and hematic routes. It is well established that the taste bud is both an interoreceptor and an exteroreceptor, capable of being stimulated not only by agents that enter the taste pore after being solubilized in saliva, but also by agents that stimulate taste buds via capillary diffusion, a phenomenon termed 'intravascular taste' (Bradley 1973). Interestingly, the effects of tastants introduced into the lingual vascular system and to the surface of the tongue are additive. Moreover, when responses to an oral tastant are adapted by continuous stimulation of a tastant, those induced by vascular infusion of the same stimulus remain active, suggesting the involvement of different sets of receptors (Bradley 1973). Given that an intravascular stimulus, unlike an oral stimulus, must pass through the capillary wall, differential capillary permeability of blood-borne stimuli may explain some taste-related dysgeusias. It is known, for example, that capillaries are much more permeable to NaCl than to sucrose and glucose.

Our finding of an age-related change in the identification of the quality of the tastants independent of ESZ is in accord with a large literature on this topic (Fikentscher et al., 1977;Doty 1978;Schiffman et al., 1979;Hyde and Feller 1981;Weiffenbach et al., 1982;Matsuda and Doty 1995;Schiffman et al., 2002). The fact that PTC taster status was not meaningfully associated with our test measures is noteworthy, given reports that PTC tasters are generally more sensitive to the bitter tastes of a range of tastants such as nicotine, saccharine/sucrose, and quinine (Enoch et al., 2001;Chang et al., 2006;Pronin et al., 2007). This suggests the possibility that the bitter receptor types involved in ESZ-induced dysgeusia may not be the bitter receptors involved in the perception of PTC, although other explanations seem more probable. Thus, the PTC tasters did not rate quinine or any of the other tastants employed in this study as more intense than PTC non-tasters, conceivably reflecting the relatively small sample sizes and the sensitivity of our taste measures. Additionally, we measured PTC tasting ability to only one concentration of PTC. Like other taste measures, PTC sensitivity follows a continuum, albeit bimodal, and is not truly a bipartite categorization of taste function. Hence, a more thorough assessment of this relationship using PTC threshold values, for example, may have proven more efficacious. That being said, the PTC strips used in this study have proven successful in other studies in which PTC effects have been reported, suggesting that they were not the primary basis for our lack of PTC-related effects (Joiner and Perez, 2004;Moberg et al., 2007).

In summary, the present study demonstrates that ESZ induces a dysgeusic condition in a large number of people. The intensity of the dysgeusia is positively correlated with both saliva and blood levels of ESZ, suggesting induction via stimulation of the taste buds. The dysgeusia is typically bitter/metallic in nature, stronger in women than in men, stronger in the morning than in the evening, and relatively independent of PTC taster status and age. ESZ had minimal influences on the taste function measures employed in this study. This research represents the first direct demonstration of an association, for any drug, between the intensity of a drug-related dysgeusia and the levels of the drug in either saliva or blood.

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## References

Agro AS, Garner ET, Wright III JW, Caballeros I DE, Villeda B, Seidlin M. Clinical trial of ototopical ofloxacin for treatment of chronic suppurative otitis media. Clin Ther 1998;20:744–59.
Anderson RU, Mobley D, Blank B, Saltzstein D, Susset J, Brown JS. Once daily controlled versus immediate release oxybutynin chloride for urge urinary incontinence. OROS Oxybutynin Study Group. J Urol 1999;161:1809–12.
Armstrong RB, Luber KM, Peters KM. Comparison of dry mouth in women treated with extended-release formulations of oxybutynin or tolterodine for overactive bladder. Int Urol Nephrol 2005;37:247–52.
Balter MB, Uhlenhuth EH, Kupfer DJ, Dement WC, Gelenberg AJ, Jonas JM, et al. New epidemiologic findings about insomnia and its treatment. J Clin Psychiatry 1992;53:34–42.
Bradley RM. Electrophysiological investigations of intravascular taste using perfused rat tongue. Am J Physiol 1973;224:300–4.
Caille G, Du Souich P, Spenard J, Lacasse Y, Vezina M. Pharmacokinetic and clinical parameters of zopiclone and trimipramine when administered simultaneously to volunteers. Biopharm Drug Dispos 1984;5:117–25.
Chang WI, Chung JW, Kim YK, Chung SC, Kho HS. The relationship between phenylthiocarbamide (PTC) and 6-n-propylthiouracil (PROP) taster status and taste thresholds for sucrose and quinine. Arch Oral Biol 2006;51:427–32.
Deems DA, Doty RL, Settle RG, Snow JB. Chemosensory dysfunction — analysis of 750 patients from the University-Of-Pennsylvania-Smell-And-Taste-Center. Chem Senses 1988;13:683.
Doty RL. Gender and reproductive state correlates of taste perception in humans. In: McGill TE, Dewsbury DA, Sachs BD, editors. Sex and Behavior: Status and Prospectus. New York: Plenum Press; 1978. p. 337–62.
Doty RL, Cameron EL. Sex differences and reproductive hormone influences on human odor perception. Physiol Behav 2009;97:213–28.
Doty RL, Bagla R, Morgenson M, Mirza N. NaCl thresholds: relationship to anterior tongue locus, area of stimulation, and number of fungiform papillae. Physiol Behav 2001;72:373–8.
Doty RL, Philip S, Reddy K, Kerr KL. Influences of antihypertensive and antihyperlipidemic drugs on the senses of taste and smell: a review. J Hyperten 2003;21:1805–13.
Doty RL, Shah M, Bromley SM. Drug-induced taste disorders. Drug Saf 2008;31:199–215.
DuBois GE, DeSimone J, Lyall V. Chemistry of gustatory stimuli. In: Basbaum AI, Kaneko A, Shepherd GM, Westheimer G, editors. The Senses: A comprehensive Reference. Oxford: Elsevier; 2008. p. 27–74.
Enoch MA, Harris CR, Goldman D. Does a reduced sensitivity to bitter taste increase the risk of becoming nicotine addicted? Addict Behav 2001;26:399–404.
Fikentscher R, Roseburg B, Spinar H, Bruchmuller W. Loss of taste in the elderly: sex differences. Clin Otolaryngol Allied Sci 1977;2:183–9.
Hyde RJ, Feller RP. Age and sex effects on taste of sucrose, NaCl, citric acid and caffeine. Neurobiol Aging 1981;2:315–8.
Joiner Jr TE, Perez M. Phenylthiocarbamide tasting and family history of depression, revisited: low rates of depression in families of supertasters. Psychiatr Resid 2004;126:83–7.
Krystal AD, Walsh JK, Laska E, Caron J, Amato DA, Wessel TC, Roth T. Sustained efficacy of eszopiclone over 6 months of nightly treatment: results of a randomized, double-blind, placebo-controlled study in adults with chronic insomnia. Sleep 2003;26:793–9.
Loucks CA, Doty RL. Effects of stimulation duration on electrogustometric thresholds. Physiol Behav 2004;81:1–4.
Matsuda T, Doty RL. Regional taste sensitivity to NaCl: relationship to subject age, tongue locus and area of stimulation. Chem Senses 1995;20:283–90.
Miller SL, Mirza N, Doty RL. Electrogustometric thresholds: relationship to anterior tongue locus, area of stimulation, and number of fungiform papillae. Physiol Behav 2002;75:753–7.
Moberg PJ, Mcgue C, Kanes SJ, Roalf DR, Balderston CC, Gur RE. Phenylthiocarbamide (PTC) perception in patients with schizophrenia and first-degree family members: relationship to clinical symptomatology and psychophysical olfactory performance. Schizophr Res 2007;90:221–8.
Mueller C, Kallert S, Renner B, Stiassny K, Temmel AF, Hummel T. Quantitative assessment of gustatory function in a clinical context using impregnated "taste strips". Rhinology 2003;41:2–6.
Mueller KL, Hoon MA, Erlenbach I, Chandrashekar J, Zuker CS, Ryba NJP. The receptors and coding logic for bitter taste. Nature 2005;434:225–9.

O'Mahony M, Goldenberg M, Stedmon J, Alford J. Confusion in the use of the taste adjectives 'sour' and 'bitter'. Chem Senses 1979;4:301–18.
Pronin AN, Xu H, Tang H, Zhang L, Li Q, Li X. Specific alleles of bitter receptor genes influence human sensitivity to the bitterness of aloin and saccharin. Curr Biol 2007;17:1403–8.
Schiffman SS, Hornack K, Reilly D. Increased taste thresholds of amino acids with age. Am J Clin Nutr 1979;32:1622–7.
Schiffman SS, Graham BG, Suggs MS, Sattely-Miller EA. Effect of psychotropic drugs on taste responses in young and elderly persons. Ann NY Acad Sci 1998;855:732–7.
Schiffman SS, Zervakis J, Graham BG, Westall HL. Age-related chemosensory losses: effect of medications. In: Givens P, Pardes D, editors. Chemistry of Taste. Washington DC: Amer Chem Assoc; 2002. p. 94-108.
Schiffman SS, Zervakis J, Suggs MS, Budd KC, Iuga L. Effect of tricyclic antidepressants on taste responses in humans and gerbils. Pharmacol Biochem Behav 2000a;65:599–609.
Schiffman SS, Zervakis J, Westall HL, Graham BG, Metz A, Bennett JL. Effect of antimicrobial and anti-inflammatory medications on the sense of taste. Physiol Behav 2000b;69:413–24.
Versi E, Appell R, Mobley D, Patton W, Saltzstein D. Dry mouth with conventional and controlled-release oxybutynin in urinary incontinence. The Ditropan XL Study Group. Obstet Gynecol 2000;95:718–21.
Weiffenbach JM, Baum BJ, Burghauser R. Taste thresholds: quality specific variation with human aging. J Gerontol 1982;37:372–7.
Zervakis J, Graham BG, Schiffman SS. Taste effects of lingual application of cardiovascular medications. Physiol Behav 2000;68:405–13.