ORIGINAL CONTRIBUTION

Rhinology 49: 331-336, 2011

# Correlation analyses of detection thresholds of four different odorants*

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SUMMARY

*The detection threshold task of the Sniffin' Sticks can be conducted using two different odorants – n-butanol or phenylethyl alcohol (PEA). Previous studies show contradictory results regarding the correlation analysis of the two odorants. The current study investigated the relationship between PEA and n-butanol with respect to previous findings and subject population. We compared four different odorants (PEA, n-butanol, isoamyl butyrate, isobutanol) in an olfactory detection threshold task depending on subject population. Test odorants were applied to 73 healthy subjects. The experiment was divided into two sessions performed on two different days. The correlation coefficient between individual thresholds of PEA and n-butanol was not significant when exclusively normosmic subjects were included, but significant when additionally hyposmic, older subjects were studied. Comparable results were found for the analysis of the odorants n-butanol and isoamyl butyrate. Correlation between n-butanol and isobutanol was significant, both for exclusively normosmic, and additionally older, hyposmic subjects. The analyses of all other odorants revealed no significant correlations. Results give explanations for previous contradictory findings regarding investigations of PEA and n-butanol in a detection threshold task, and indicate that a formal validation of the Sniffin' Sticks with PEA as odorant is required.*

*Key words: n-butanol, phenylethyl alcohol, isoamyl butyrate, isobutanol, sensitivity, Sniffin' Sticks*

## INTRODUCTION

It is a well known phenomenon that different odorants demonstrate different olfactory detection threshold values in healthy subjects. Human olfactory thresholds have been investigated in numerous studies [1-3], and compilations of threshold values have been published by several authors [4-8]. The threshold concentrations for odorant detection nonetheless vary greatly due to variations in the assessed concentrations of the odorants, differences in stimulus preparation, presentation, and differences in the psychophysical testing methods [9-12]. Furthermore, the variations in absolute threshold values for a given odorant can be caused by differences in age [13,14], receptor repertoire [15], current state of hunger [16], and hormonal status [17,18] of the subjects. To assess subjects' olfactory function numerous methods have been established [9-11]. In the German-speaking countries, the Sniffin Sticks' are in common use which are a well-established test battery for combined testing of olfactory function [12,19]. This test battery comprises three subtests, namely odour threshold, odour discrimination, and odour identification using pen-like odour-dispensing devices for odour presentation. Commercially available is the detection threshold test with two different odorants, n-butanol and phenylethyl alcohol (PEA). Both tests are identical regarding their odorant concentrations, assembling, and testing procedure; they differ only in the assessed odorants and solvents used to dilute the odorants. Both tasks are commonly applied in published studies. The detection threshold task with n-butanol as odorant is thoroughly validated [12,13,19], whereas the threshold test with PEA as odorant is hardly investigated [20-22]. To date, only two studies compared both odorants in the detection threshold test of the Sniffin' Sticks with contradictory findings [20,22]. Croy et al. [20] found a significant correlation between both odorants, whereas our previous study [22] revealed no significant correlation. We believe that this difference is of relevance before an application of the threshold test with PEA as odorant can be generally recommended. In their study, Croy et al. [20] included healthy controls as well as hyposmic subjects whereas we investigated only younger, normosmic subjects. Accordingly, we hypothesize that the contradictory findings have been mainly caused by the varying data sets. Therefore, in the present study, we conducted a comparison of the odorants PEA and n-butanol in the detection threshold task of the Sniffin' Sticks

*Received for publication: November 3, 2010; accepted: December 13, 2010

DOI:10.4193/Rhino10.263

with a subject population comparable with the data set of Croy et al. [20] and our previously published study [22]. Furthermore, we also acquired detection thresholds for two further odorants, isobutanol and isoamyl butyrate (IAB) to investigate possible correlations among the different odorants with respect to the subject population included in statistical analyses.

## MATERIAL AND METHODS

The entire study was approved by the Institutional Review Board of the Ludwig-Maximilians-University of Munich and was conducted according to the Declaration of Helsinki. All subjects provided their written informed consent.

### *Subjects*

A total of 73 healthy subjects (36 females) participated in this study. Similar to the age classification described in previous studies [13,23,24], we conducted the experiment with three different age classes: age class 1 included subjects aged 18 – 34 years (15 males, 13 females; age range 18 – 33 years; mean age 26.3 ± 3.6 years), age class 2 comprised subjects aged 35 – 54 years (12 males, 13 females; age range 35 – 51 years; mean age 42.2 ± 4.4 years), and class 3 contained subjects ≥ 55 years (10 males, 10 females; age range 55 – 85 years; mean age 59.7 ± 6.8 years). Using the olfactory screening test Sniffin' Sticks with the thoroughly validated n-butanol as odorant of the detection threshold task, all subjects were tested for their olfactory function by determining the TDI score (sum of threshold, discrimination and identification scores) [13,19,24]. We included exclusively normosmic subjects in age class 1 (mean TDI score 36.42 ± 3.27, range 31.50 – 43.75) and 2 (mean TDI score 34.54 ± 2.70, range 30.25 – 40.50). All subjects of age class 3, whether they were normosmic or hyposmic were included (mean TDI score 28.78 ± 5.05, range 19.00 – 38.75), because olfactory performance decreases with aging [13,14]. Twelve subjects of age class 3 were hyposmic (mean TDI score 25.46 ± 3.26, range 19.00 – 29.75) and eight subjects were normosmic (mean TDI score 33.75 ± 2.30, range 31.50 – 38.75). Mean age did not differ significantly between male (mean age 40.5 ± 14.0 years, n = 37) and female subjects (mean age 41.3 ± 14.9 years; n = 36), neither for all subjects ($t(71) = 0.22$, p = n.s.), nor for the three age classes analysed separately (age class 1: $t(26) = 0.66$, p = n.s.; age class 2: $t(23) = 0.57$, p = n.s.; age class 3: $t(18) = 0.13$, p = n.s.). All subjects were non-smokers and were not taking any medication known to interfere with sensory perception [25-27]. None of them reported any olfactory disturbances, neurological disorders, or metabolic diseases. All subjects including the hyposmic subjects of age class 3 reported not to suffer from allergic coryza and chronic rhinitis, not having any infections of the respiratory tract, and reported not having epistaxis during the three days before the testing session.

### *Stimulus material*

The experiment was conducted with four different odorants in an olfactory detection threshold test: n-butanol, phenylethyl alcohol (PEA), isoamyl butyrate (IAB), and isobutanol (2-methylpropyl alcohol). The threshold tests with the odorants n-butanol and PEA of the Sniffin' Sticks are commercially available (Burghart Instruments, Wedel, Germany), the threshold tests using IAB (> 99.5% purity, Th.Geyer, Renningen, Germany) and isobutanol (> 99% purity, Merck, Darmstadt, Germany) as odorants were self-made. These two odorants were chosen because of their structural similarity to n-butanol. N-butanol is structurally most similar to isobutanol, medium similar to IAB, and least similar to PEA. The Sticks were made in the same manner as described for the commercially available Sniffin' Sticks [12]. Both odorants were diluted in geometric series consisting of sixteen steps with a dilution ratio of 1:2; the highest concentration constituted 4 % (v/v). Each commercially available (unfilled) felt-tip pen (Burghart Instruments, Wedel, Germany) was filled with 4 ml of the odorant in the appropriate concentration, or with 4 ml of the solvent to prepare the blank sticks. The odorants IAB, isobutanol, and PEA were diluted in propylene glycol (> 99.5% purity, Fluka Chemie, Buchs, Switzerland), n-butanol was diluted in aqua conservata (demineralized water with antidegradants).

### *Experimental procedure*

Subjects' sensitivity was tested with four different odorants (n-butanol, isobutanol, PEA, IAB). Detection threshold was determined using a single-'staircase', three alternative forced choice procedure described by Doty [28]. Standard procedure for using the Sniffin' Sticks test battery is described by Kobal et al. [12] and Hummel et al. [19]. For more details on the procedure please consult our previously published study [22].

The four odours were tested in two sessions performed on two different days (2 tests/session). The interval between both testing sessions was between 24 hours and 10 days. Both sessions were conducted at the same time of the day. The order of the four threshold tests was pseudo randomized, and the ratio of sex was systematically counterbalanced for each age class.

After each threshold test subjects filled in a questionnaire. One session of the experiment lasted approximately 45 min including a 15 min break between both testing odorants to avoid olfactory adaptation.

### *Questionnaire*

A questionnaire consisting of seven parameters was employed to measure the perception of the odorants, the recipients' emotional state, and their current state of hunger. At the beginning of each testing day subjects rated their current state of hunger (0 = very hungry, 100 = not hungry at all); after each threshold test they rated the following parameters: emotional valence (0 = positive, 100 = negative), arousal (0 = aroused, 100 = calm), alertness (0 = very inattentive, 100 = very attentive), as well as the pleasantness (0 = pleasant, 100 = unpleasant), and familiarity (0 = not familiar, 100 = very familiar) of the assessed odorant, and the intensity (0 = very weak, 100 = very strong) of the pen containing the highest concentration.

Participants answered the questions using a visual analogue scale (VAS). They gave a response by placing a mark on a 100 mm horizontal line. VAS have been shown to measure even

minor changes in affect with high reliability and validity [29,30].

*Statistical analyses*

Data were statistically analysed using SPSS version 18.0 for Windows (SPSS Inc., Chicago, IL, USA). Normality of the data was tested using the Kolmogorov–Smirnov test. To explore differences between subjects' sensitivities of the four different odorants, and the parameters of the questionnaire between the different sessions, normally distributed data (detection threshold of isobutanol; ratings of the questionnaire) were submitted to repeated measure analyses of variance (ANOVA) or two-tailed Student's paired t-tests; not normally distributed data (detection threshold of n-butanol, IAB, and PEA) were submitted to Friedman tests and subsequent non-parametric Wilcoxon signed-rank tests. Correlation analyses were performed using Spearman's rank tests (not normally distributed data) or Pearson's correlation analysis (normally distributed data) to examine the relationship between the four different odorants in an olfactory detection threshold task, and between the odorants and subjects' age, respectively. Differences between the three age classes regarding the different parameters were tested using one-way analysis of variance (one-way ANOVA; normally distributed data), or Kruskal-Wallis test and subsequent Mann Whitney U tests (not normally distributed data). To analyse sex-differences data were submitted to independent two sample t-tests (normally distributed data) or Mann Whitney U tests (not normally distributed data). Results of the questionnaire were corrected for multiple testing using the Bonferroni method. The alpha level was set at 0.05.

## RESULTS

*Olfactory sensitivity*

Results of subjects' olfactory sensitivity demonstrated significant differences between the four odorants (Table 1), both when data were analysed for all subjects (age classes 1-3: $p < 0.001$), and when data were analysed for each age class separately (age class 1, 2, and 3, each with $p < 0.001$). All pair wise comparisons analysed for age classes 1-3 were significant with $p < 0.001$. Age class 1 and 2 showed also significant differences for each pair wise comparison (age class 1: all pair wise comparisons, each with $p < 0.001$ except IAB vs. isobutanol, $p = 0.038$; age class 2: all pair wise comparisons, each with $p \leq 0.001$ except n-butanol vs. PEA, $p = 0.005$, and IAB vs. isobutanol, $p = 0.028$). Age class 3 revealed significant differences between the four testing odorants for each pair wise comparison except IAB vs. n-butanol indicating similar threshold scores (age class 3: all pair wise comparisons, each with $p \leq 0.001$ except n-butanol vs. IAB, p = n.s., n-butanol vs. isobutanol, $p = 0.003$, and IAB vs. isobutanol, $p = 0.018$).

With regard to the parameter sex there were no significant differences in olfactory detection thresholds between men and women, neither for subjects all together, nor for the three age classes analysed separately (p = n.s.).

*Correlation analyses between four different odorants*

Spearman-rho correlation coefficients were calculated for each pair of odorants assessed in an olfactory detection threshold test. Results are shown in Table 2. Correlation analyses of the four different odorants revealed significant correlation coefficients for subjects' individual sensitivity scores of n-butanol vs. PEA, n-butanol vs. isobutanol, and n-butanol vs. IAB, when data were analysed for all subjects (age classes 1-3), but no significant correlations between detection thresholds of PEA vs. isobutanol, PEA vs. IAB, and IAB vs. isobutanol. When data were analysed for exclusively normosmic subjects (age classes 1 and 2), no significant correlations between the odorants were observed; only the correlation between n-butanol vs. isobuta-

Table 1. Results of olfactory sensitivities of four different odorants in a detection threshold test for all subjects (age classes 1 - 3), and for the three age classes separately (age class 1: 18 – 34 years; age class 2: 35 – 54 years; age class 3: ≥ 55 years). Reported are means ± standard deviations.

| | n-Butanol | Isobutanol | PEA | IAB |
|---|---|---|---|---|
| All subjects (n = 73) | 8.13 ± 2.15 | 5.06 ± 2.78 | 12.12 ± 3.86 | 6.60 ± 1.51 |
| Age class 1 (n = 28) | 8.85 ± 2.06 | 5.62 ± 2.87 | 13.26 ± 3.13 | 6.91 ± 1.03 |
| Age class 2 (n = 25) | 8.44 ± 1.81 | 5.55 ± 2.48 | 11.49 ± 4.29 | 6.82 ± 1.49 |
| Age class 3 (n = 20) | 6.75 ± 2.14 | 3.66 ± 2.67 | 11.31 ± 4.02 | 5.89 ± 1.91 |

Table 2. Results of the correlation analyses of four different odorants in a detection threshold test. Reported are Spearman-rho correlation coefficients and p-values for all subjects (age classes 1 - 3, 18 - 85 years), and for exclusively normosmic subjects between the ages of 18 and 54 years (age classes 1 + 2). * Spearman's rank test, significant with $p \leq 0.05$.

| | All subjects (n = 73) | | Age classes 1 + 2 (n = 53) | |
|---|---|---|---|---|
| | Spearman-rho | p-value | Spearman-rho | p-value |
| n-Butanol vs. PEA | 0.230 | 0.050* | 0.163 | 0.243 |
| n-Butanol vs. Isobutanol | 0.332 | 0.004* | 0.303 | 0.028* |
| n-Butanol vs. IAB | 0.249 | 0.034* | 0.039 | 0.780 |
| PEA vs. Isobutanol | 0.117 | 0.323 | 0.206 | 0.138 |
| PEA vs. IAB | 0.024 | 0.838 | 0.015 | 0.915 |
| IAB vs. Isobutanol | 0.015 | 0.898 | - 0.108 | 0.441 |

Table 3. Means ± standard deviations of the parameters of the questionnaire rated by the subjects at the beginning of each testing day (interval ≤ 10 days) or after each threshold test (n = 73).

| | Testing day 1 | Testing day 2 | n-Butanol session | Isobutanol session | PEA session | IAB session |
|---|---|---|---|---|---|---|
| Intensity | | | 73.92 ± 19.19 | 60.44 ± 23.01 | 71.32 ± 18.66 | 76.07 ± 18.81 |
| Pleasantness | | | 61.62 ± 23.66 | 52.92 ± 22.11 | 27.19 ± 21.11 | 38.19 ± 25.15 |
| Familiarity | | | 52.36 ± 23.43 | 55.08 ± 23.34 | 69.97 ± 16.87 | 55.70 ± 24.02 |
| Valence | | | 28.40 ± 21.06 | 29.73 ± 20.91 | 25.29 ± 20.28 | 25.30 ± 18.53 |
| Arousal | | | 78.21 ± 16.85 | 73.10 ± 19.63 | 77.90 ± 18.41 | 75.88 ± 18.94 |
| Alertness | | | 78.51 ± 15.24 | 77.64 ± 15.65 | 78.97 ± 14.90 | 78.07 ± 15.80 |
| Hungriness | 62.51 ± 21.88 | 64.71 ± 17.81 | | | | |

nol was significant.

*Olfactory sensitivity in the different age classes*

Analyses of differences between the three age classes demonstrated significant differences for the odorants n-butanol ($p = 0.001$), isobutanol ($F(2.70) = 3.73$, $p = 0.029$), and IAB ($p = 0.034$), whereas the sensitivity scores of PEA demonstrated no significant differences between the three age classes (p = n.s.). Analyses of the pair wise comparisons revealed significant differences between the age classes 1 vs. 3, and between classes 2 vs. 3 regarding IAB and n-butanol, but no significant differences for the other odorants. Participants of age class 3 showed significantly lower threshold scores for the odorants n-butanol and IAB when compared with the scores of the subjects of age class 1 and 2 (n-butanol: age class 1 vs. 3, $p = 0.001$; age class 2 vs. 3, $p = 0.009$; IAB: age class 1 vs. 3, $p = 0.017$; age class 2 vs. 3, $p = 0.028$). Pair wise comparisons of isobutanol revealed significant differences only between the age classes 1 vs. 3 (age class 1 vs. 3, $p = 0.046$; age class 2 vs. 3, and age class 1 vs. 2, each with p = n.s.). The comparisons between the age classes 1 and 2 revealed no significant differences between subjects' detection thresholds of all assessed odorants (p = n.s.).

Results revealed a consistent decline of detection thresholds with increasing age (Table 1). Correlation analyses between subjects' age and the different detection thresholds revealed significant results for the odorants n-butanol (Spearman-rho correlation coefficient = - 0.440, $p < 0.001$) and isobutanol (Pearson's $r = - 0.321$, $p = 0.006$). Data demonstrated no significant correlations between age and the odorants PEA (Spearman-rho correlation coefficient = - 0.217, p = n.s.) and IAB (Spearman-rho correlation coefficient = - 0.203, p = n.s.).

*Questionnaire*

Descriptive statistics of subjects' ratings of the questionnaire are shown in Table 3. Results revealed no significant differences between the two testing days regarding subjects' hungriness; they were not hungry during both days ($t (72) = 1.23$, p = n.s.). Subjects felt predominantly positive ($F (3.216) = 1.64$, p = n.s), attentive ($F (3,216) = 0.22$, p = n.s.), and calm during all threshold tests ($F (3.216) = 2.68$, p = n.s.). Subjects evaluated the odorant PEA as familiar, the odorants isobutanol, n-butanol, and IAB as neutral to slightly familiar; pair wise comparisons revealed significant differences only for the odorant PEA ($F (3.216) = 11.99$, $p < 0.001$; pair wise comparisons: PEA vs. n-butanol, PEA vs. isobutanol, and PEA vs. IAB, each with $p < 0.001$). PEA and IAB were estimated as pleasant, isobutanol as nearly neutral, and n-butanol as slightly unpleasant; statistical analyses displayed significant differences for all comparisons ($F (3.216) = 44.62$, $p < 0.001$; all pair wise comparisons, each with $p \leq 0.002$ except isobutanol vs. n-butanol, $p = 0.008$). Subjects rated the assessed odorants as slightly strong (isobutanol: mean 60.44, SD 23.01) to strong (PEA: mean 71.32, SD 18.66; n-butanol: mean 73.92, SD 19.19; IAB: mean 76.07, SD 18.81); pair wise comparisons revealed significant differences for isobutanol versus the three other odorants regarding the parameter intensity ($F (3.216) = 11.78$, $p < 0.001$; pair wise comparisons: isobutanol vs. n-butanol, isobutanol vs. PEA, and isobutanol vs. IAB, each with $p \leq 0.001$; IAB vs. PEA, PEA vs. n-butanol, and IAB vs. n-butanol, each with p = n.s.).

Analyses of sex-differences revealed significant differences only for the parameter intensity of n-butanol ($t (71) = 2.60$, $p = 0.011$); females (mean 79.61, SD 15.19) evaluated n-butanol as significantly more intense than males (mean 68.38, SD 21.17). All other ratings of the questionnaire were similar for both sexes (p = n.s.).

Analyses of differences between the three age classes demonstrated significant differences only for the parameter familiarity ($F (2.70) = 6.55$, $p = 0.002$) of isobutanol. Subjects of age class 2 estimated isobutanol as familiar (mean 67.68, SD 16.10), whereas subjects of age class 1 (mean 50.00, SD 23.11) and 3 (mean 46.45, SD 25.55) rated this odorant as nearly neutral (pair wise comparisons: age class 1 vs. 2, p = n.s.; age class 2 vs. 3, $p = 0.005$; age class 1 vs. 3, p = n.s.). All other parameters of the questionnaire revealed no significant differences between the three age classes (p = n.s.).

## DISCUSSION

The present study gives explanation for previous contradictory findings regarding the correlation between individual sensitivities of the odorants PEA and n-butanol in an olfactory detection threshold test of the Sniffin' Sticks [20,22]. A significant correlation between the sensitivities of PEA and n-butanol only occurred when not only normosmic subjects, but also hyposmic subjects were included in statistical analysis. In contrast, the analysis of exclusively normosmic subjects showed no significant correlation coefficient. Therefore, the contradictory

previous findings regarding these odorants are explainable. Croy et al. [20] included more elder subjects, and subjects with olfactory impairment in their correlation analysis, and found a significant correlation between the odorants PEA and n-butanol, whereas our previous study [22] revealed no significant correlation between the two odorants due to a differing subject population compared to the study of Croy et al. [20]. In our previous study we included only normosmic subjects through the age of 51 years, comparable with the analysis of age classes 1 and 2 of the current study. Therefore, we can confirm the findings of our previous study regarding the results of the correlation analysis and the results of the detection threshold scores of PEA and n-butanol. Detection thresholds of both odorants are similar to previously published studies [13,19,21,24,31,32], whereas the threshold scores of Croy et al. [20] were not in line with the findings in literature.

Comparable results were found for the comparison of the detection thresholds of n-butanol and IAB; correlation coefficients were significant for all subjects, but revealed no significant results when exclusively normosmic subjects were studied.

The analysis of the odorants n-butanol and isobutanol revealed significant correlations, both when data were analysed for all subjects, and when data were analysed for only normosmic subjects. These findings could be attributed to the fact that odorants with the same functional group activate the same olfactory receptor set [33-36]. As the repertoire of olfactory receptors is unique for every individual [15], detection thresholds differ between humans depending on the combination of receptors which are activated by the assessed odorant. Therefore, if two different odorants, like n-butanol and isobutanol, have the same functional group, subjects' detection thresholds of both substances are related to each other due to the same receptors which process the odorants [37]. This could lead to a strong relationship, and accordingly to a significant correlation even when only normosmic subjects are included.

This fact could also explain our findings of no significant correlations between subjects' sensitivities of the odorants PEA versus isobutanol, PEA versus IAB, and IAB versus isobutanol, neither for subjects all together, nor for subjects of age class 1 and 2. These three odorants have different structures and functional groups, and therefore, activate different receptors leading to a lack of correlations between the individual thresholds of these odorants, even if additionally hyposmic subjects are included in data analyses. Another reason might have been the generally small threshold values of the odorants IAB and isobutanol. As we assembled both dilution series in the same manner as the commercially available threshold tests (PEA and n-butanol), the utilized concentrations of IAB and isobutanol could have been too low for a representative detection threshold assessment, and therefore, not comparable with the results of the PEA threshold values which were generally higher. These findings indicate that each odorant in an olfactory detection threshold test should be validated on its own; odorant concentrations for assessing subjects' sensitivities could not be one-to-one translated to other odorants. We suggest that these assumptions can also be regarded to the odorants PEA and n-butanol of the detection threshold task of the Sniffin' Sticks. Our findings of generally higher threshold scores of PEA [22] compared to the other odorants, which were found even for elder subjects, and the lacking significant differences between the age classes, also indicate that a formal validation of the threshold task with PEA as odorant is necessary. On the contrary, thresholds of n-butanol, isobutanol, and IAB demonstrated significant differences between the age classes. It is well documented that advancing age impairs the ability to detect odorants due to considerable deteriorations of the olfactory neuroepithelium throughout the life span [13,14]. Subjects may have a better receptor repertoire for PEA resulting in higher threshold scores. Therefore, even low concentrated PEA sticks, and subjects of age class 3 could detect the odorant. In addition, correlation analysis between age and PEA threshold scores revealed no significant result. On the contrary, the odorants n-butanol and isobutanol showed a consistent decrease of detection thresholds with increasing age, and with significant correlation coefficients. The lack of significant correlations between subjects' age and their threshold scores of PEA and IAB could be attributed to the characteristic of olfactory receptors; both odorants have other functional groups than iso- and n-butanol. These findings indicate that odorants related to n-butanol and with the same functional group, may reflect subjects' olfactory performance better than other odorants, and therefore, might be more appropriate for assessing olfactory functions in the threshold test of the Sniffin' Sticks with respect to subjects' age.

Our study gives an explanation for previous contradictory findings regarding the odorants phenylethyl alcohol and n-butanol in the olfactory detection threshold task of the Sniffin' Sticks. We suggest that significant correlations between the individual threshold scores of the two odorants only occur when additionally to normosmic subjects also hyposmic subjects are included in statistical analyses. This has implications on the clinical use of the Sniffin' Sticks with PEA as odorant. It may be appropriate to modify the TDI scale for assessing the olfactory function if PEA instead of n-butanol is used for the threshold test. As long as the reliability of the PEA threshold test of the Sniffin' Sticks is not demonstrated, only the n-butanol test should be used to avoid false positive results of patients' olfactory performance. Furthermore, our data confirm previous findings that the Sniffin' Sticks with n-butanol as detection threshold task are an undisputable well established test, and excellently represents subjects' olfactory function throughout humans' life span. On the contrary, the threshold task with the odorant PEA is poorly validated, yet. The results of the comparisons of PEA with the odorants n-butanol, isobutanol, and isoamyl butyrate, and the findings regarding subjects' age and PEA account for a formal examination of this threshold task of the Sniffin' Sticks in further studies as it has already been done for the threshold task with n-butanol as odorant.

ACKNOWLEDGMENTS

Parts of this study were conducted in line with the dissertation of Tilman Frank at the Medical Faculty of the Ludwig-Maximilians-University of Munich (in preparation).

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