ORIGINAL CONTRIBUTION

The Leicester semi-automated olfactory threshold test – a psychophysical olfactory test for the 21st century*

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SUMMARY

Objective: Develop a useful and cost-effective olfactometer for routine clinical use by providing a standardised threshold test for patients with olfactory disorders presenting in the ENT clinic.

Method of study: A prospective study of olfactory thresholds in 48 healthy volunteers on 2 consecutive occasions, undergoing quantitative testing with an olfactometer. Further studies of 10 subjects performing 20 tests and 100 subjects performing a single test were performed. An olfactometer was designed to deliver a semi-automated threshold test for an odour. It contains 8 logarithmic dilutions of an odour along with a control valve operated by software from a laptop computer. Common potential variables for olfactory threshold testing were considered including peak inspiratory flow rate. The odours used were phenethyl alcohol (PEA) and eucalyptol (EUC). Subjects were asked to perform 2 tests within 1 month of each other and the mean threshold score for each was calculated to derive a test-retest score.

Main Results: Consistent olfactory thresholds for PEA were achieved with a mean concentration of 10-4. Test-retest reliability score (r_x) for the olfactometer was $r_x = 0.78$ (95% CI 0.67 to 0.89).

Principal Conclusions: The Leicester Olfactometer provides a simple and cost-effective method of reliably assessing olfactory thresholds in the outpatient clinic.

Key words: olfaction disorders, sensory thresholds, smell, outpatient

INTRODUCTION

A plethora of quantitative and qualitative test kits for the assessment of olfactory function have been devised around the world. Some of the more publicised ones include the University of Pennsylvania Smell Identification Test (UPSIT) [1,2] and the Sniffin' Sticks [3-6], with other less well publicised tests such as the Combined Olfactory Test (COT) [7,8], Connecticut Chemosensory Research Test (CCCRT) [9], Barcelona Smell Test – 24 (BAST-24) [10], and Japanese Odour Sticks [11] receiving less coverage in the medical literature. The tests listed above all comprise psychophysical tests and while there are now more objective tests available in the form of olfactory Event Related Potentials (oERPs) [12,13], with functional Magnetic Resonance Imaging (fMRI) around the corner [14], these remain time consuming, costly and the realm of specialist chemosensory centres. In otorhinolaryngological practice, assessment of olfactory function is often neglected outside of these specialist centres, and the evidence for this was only too pertinent in a recent survey of British otorhinolaryngologists [15]. The reasons for this include time, cost and a lack of therapeutic strategies. This is in contrast to the other main sensory modality that falls into the realm of otorhinolaryngologists – namely hearing. Certainly in the UK setting of the National Health Service, the cost of equipment is an important consideration when a single UPSIT kit costs about £14 and the Sniffin' Sticks cost over £400 with a shelf life of 6 months. The ideal olfactory test for this kind of environment in general otorhinolaryngological clinics should be cost-effective, easy to use and have longevity. In the latter case this should mean that the odours can be replenished at a minimal cost to the individual department.

Aside from cost, a test that can be administered predominantly by the patient with little or no input from a clinician saves valuable time in the clinic and allows the patient to see the

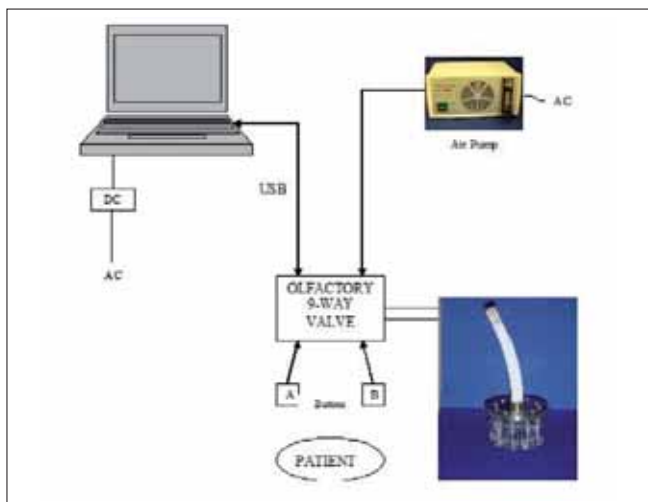Figure 1. The Mark 2 Leicester Olfactometer (LOM2).

Figure 2. Diagrammatic annotation of the LOM2 apparatus.

doctor with the result of their olfactory test much like a patient with hearing loss would have a prior audiogram.

In considering the best format for a modern psychophysical test, careful consideration was given to potential variables that could adversely affect the results achieved. Dawes gave consideration to some of these potential problems in 1998 when he listed temperature, solvent odour and bottle opening dilution effect amongst other potential sources of error in olfactometry [16]. For a robust test format it was necessary to give these variables due consideration. However the main purpose of this paper is to report the reliability data for the Leicester Olfactometer.

MATERIALS AND METHODS
This study was approved by the local ethics committee.

Olfactometer
The original core piece of apparatus, known here as the Mark I Leicester Olfactometer (LOM1), was conceived as a device containing nine bottles, 8 for dilutions of the odorant and 1 for the control (solvent only) and attached to this was a manual valve 9 tap system. This was subsequently superseded by the Mark II Leicester Olfactometer (LOM2). Problems with the LOM1 included the tubing matrix creating audible noise at the olfactometer spout and the awkwardness of the manual valves and thus a new design was conceived. The LOM2 (Figure 1) was created as a circular design so that all bottles were equidistant from the spout. Also due to the unique design of the plates in the head of the olfactometer the apparatus became much easier to maintain. Along with this an automated valve system was designed that could be controlled via a USB connection to a laptop computer. This allowed not only computer controlled randomisation of the odour presentation, but allowed the subject to control delivery of the stimulus themselves.

Measurements
Odour thresholds were initially determined for phenethyl alcohol (PEA), using a two-alternative forced choice task (2-AFC). The subject's task was to find out which of the two puffs of air smelled of the odorant, which had been presented at the beginning of the test as the highest of the 8 concentrations. Employing a staircase paradigm, pairs of stimuli were presented to the patient every 20-30 s. Correct identification of the puff of air that contained the odorant in three successive trials triggered a reversal of the staircase to the next lower concentration, whereas a single incorrect identification triggered the reversal of the staircase to the next higher concentration. From a total of seven reversals, the mode of the last four staircase reversal points was used as a threshold estimate. This technique was derived from that employed by Doty et al. in their PEA threshold test [17]. As per their technique initial testing with the LOM1 started in the middle of the range at 10^{-6} (vol/vol); the range of PEA dilutions being from 10^{-2} (strongest) to 10^{-9} (weakest). Subsequent testing with the LOM2 commenced at the lowest concentration (10^{-9}) as is practised with Sniffin' Sticks.

The test proceeds by a series of audible prompts from the laptop computer and the subject is positioned with their nose near to the spout. With the first prompt the subject is expected to first press the left-hand button to deliver the first stimulus and then lean forward and sniff at the spout with the second prompt. This is then repeated for the second stimulus with the right-hand button, following which a warble tone indicates a need for the subject to press the button which they think delivered the stimulus. Duration of the test is normally approximately 10 minutes. The assembled unit also required an air supply and the whole set of equipment is shown diagrammatically in Figure 2. The stimuli are delivered birhinally in front the nose; there is no mask or attachment between the device and the patient and thus this best mimics orthonasal olfaction (Figure 3).

Figure 3. Demonstration of the use of the LOM2 by the 3rd author.

Other modifications introduced with the LOM2 included the forced choice format whereby if a subject was completely unsure about which of the two stimuli contained the odour they were asked to press both delivery buttons and this was scored by the computer as an incorrect answer. This latter alteration helped to prevent subjects artificially trying to detect an odour way below their threshold level when guessing could show apparent olfactory ability at this level. Secondly the scoring of the last four reversals was taken as the mean not a mode of the correct data plots providing greater precision.

Whilst the bulk of the apparatus evolved, one component remained constant: the odour bottles (Figure 4). These bottles have a wick covering a central perforated core with each containing 3 ml of the odorant/solvent mixture. Airflow is directed downwards into the bottle through the wick soaked in the liquid. Odorised air is then conducted out of a tube exiting from the top of bottle to the olfactory presenting funnel. The tubes to the funnel are all separate from each other so that there is no cross contamination of odour dilutions. With the

apparatus setup determined it was then possible to examine the variables for olfactory testing. Temperature and humidity were assessed using a handheld thermohygrometer, peak inspiratory flow rate with a Youlton flow meter and patient scores for sense of smell, nasal symptoms, mood and alertness were recorded using visual analogue scores [18]. A suitable solvent was determined by a comparative study of potential solvents including propylene glycol and mineral oil [19]. Other variables examined were the effect of smoking, local air pollution and the presence of recently applied body sprays [20-22] and the methodology used to examine these variables is described in our previous work [18,20,23]. A typical threshold trace with the LOM2 is seen in Figure 5.

Subjects
Healthy volunteers were recruited for the three phases of evaluation described below and the inclusion criteria included being aged between 18 and 60 and considering themselves to have no significant deficits in olfactory ability. The exclusion criteria for these volunteers were principally the presence of active sinonasal disease on history or examination or of any olfactory disorders.

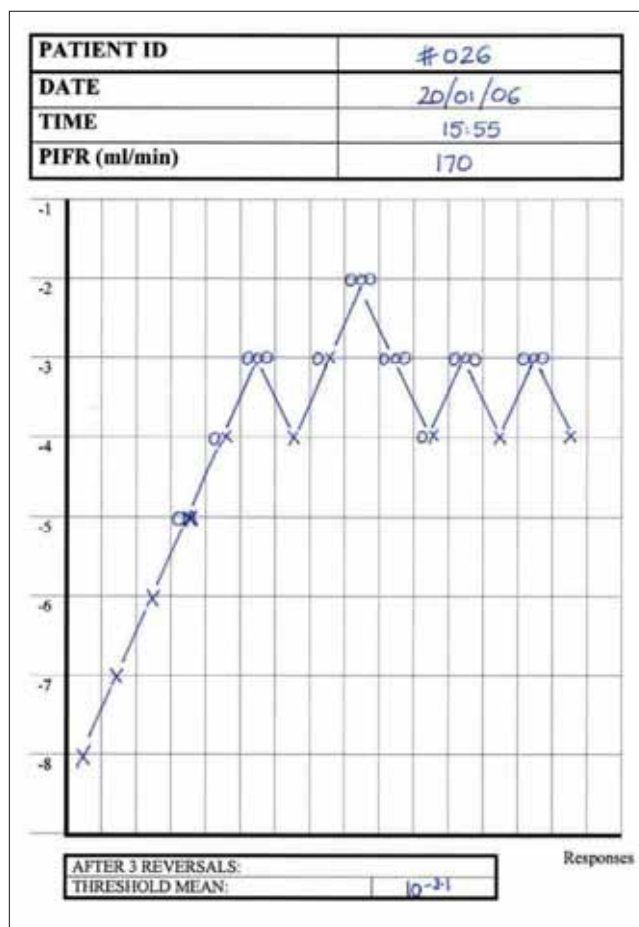Figure 4. Odour bottle (with wick removed for clarity).

Figure 5. A typical threshold trace

Reliability study
For the purposes of assessing reliability of the LOM2, threshold data on 48 healthy subjects was achieved for the odours PEA and eucalyptol (EUC). These results have been achieved using the methods described above and were analysed with intra-class correlation coefficient using Stata SE 9.1 (StataCorp LP, Texas, USA). Data was also collected on 10 subjects undergoing 20 repeated tests over 3 months, giving a total of 400 tests (200 PEA and 200 EUC).

Reference range study
Two groups of 100 healthy subjects underwent an olfactory test with the LOM2 on one occasion only for one of the odours PEA or EUC. A normal reference range was calculated from this data.

Factors influencing olfactory thresholds
The variables of age, sex, smoking status, temperature, relative humidity and peak inspiratory flow rate were correlated with the results to further determine their influence on the olfactory thresholds achieved. Unadjusted relationships between possible influencing factors and threshold mean were investigated using Pearson's correlation for continuous factors, the t-test for gender and analysis of variance for smoking status. Adjusted relationships were investigated using a multiple regression analysis. All analyses were carried out using Stata 9.1 SE software.

RESULTS
Reliability study
The mean score for PEA for the 48 subjects was $10^{-3.8}$ and $10^{-3.94}$ for the first and second test respectively (Table 1, Figure 6). The mean score for EUC was $10^{-2.52}$ and $10^{-2.67}$ respectively (Table 1, Figure 7). The test-retest reliability score was derived

Table 1. Threshold means for PEA and EUC groups.

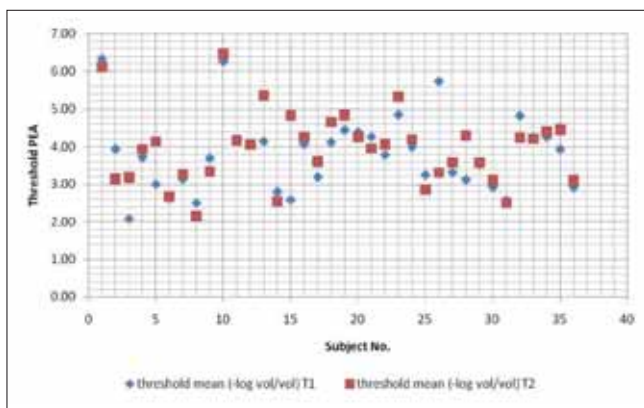| Odour	threshold mean (-log vol/vol)	threshold mean (-log vol/vol)
PEA	3.799861	3.944666
EUC	2.51821	2.66523

Figure 6. Correlation of threshold means for PEA (test-retest data).

using an intra-class correlation coefficient which was $r_x = 0.78$ (95% CI 0.67 to 0.89).

Reference range study
The other studies of healthy subjects have found the mean threshold for PEA to be between $10^{-3.97}$ and $10^{-4.02}$ log vol/vol and for EUC between $10^{-3.31}$ and $10^{-3.56}$ log vol/vol.

Factors influencing olfactory thresholds
Of all the factors investigated no statistical association could be found with either the unadjusted (Table 2) or adjusted results (results not shown).

DISCUSSION
The test-retest reliability score means that the LOM2 is comparable to existing olfactory tests; in fact it is second only to the UPSIT among the most popular of the current olfactory test kits available. The analysis of variables against the threshold mean did not show any significant influences, underlining our previous work [18,20,23] and once again confirming that these variables do not need to be controlled in a normal room environment. Whilst the authors recognise the need for more data collection, the baseline data in normal subjects to date gives us a benchmark against which future testing in patients

Table 2. Unadjusted analysis of possible influencing factors.

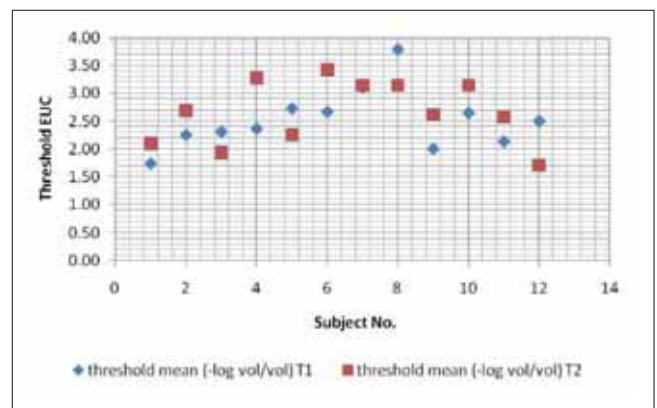| Variable	Level	N	Mean (SD) or Correlation	p-value
Gender	Male	41	4.25 (1.36)	0.0930
	Female	62	3.87 (0.94)	0.9841
Smoking status	Non-smoker	76	4.01 (1.18)	
	Ex-smoker	7	4.09 (1.12)	
	Smoker	20	4.02 (1.00)	
Age		103	-0.07	0.4964
Nasal symptoms		87	-0.01	0.9124
Sense of Smell		87	0.01	0.9416
Mood		87	0.07	0.4997
Tiredness		87	-0.11	0.3284
PIFR		104	0.18	0.0705
Temperature		104	0.00	0.9731
Humidity		104	0.11	0.2888

Figure 7. Correlation of means for EUC (test-retest data).

with olfactory deficits can be compared. Given the promising reliability seen even in a small number of patients, we are confident that the LOM2 provides an easy and reliable means of quantitative olfactory testing in the ENT Clinic. The initial cost for building the apparatus (including the air supply) is £ 3000 (plus the cost of a laptop computer if a dedicated machine is required), but after this initial outlay the only costs are for replenishing the odours which are no more than £ 20 per year. Whilst this is not currently a commercial product, the purpose of discussing costs here is purely to demonstrate that the apparatus whilst initially expensive, allows for correction of this by requiring only replenishment of the solutions and minimal maintenance over its life. This is in contrast to the commercially available products that have a shelf life and may also be single use only. There will also be an additional saving in terms of time as a resource in clinic, as the test does not require a clinician to administer it and with further modifications may enable the patient to even test themselves.

Assessment of olfactory thresholds allows quantification of any olfactory deficits somewhat akin to determining a frequency threshold in audiometry. As it stands the LOM2 does not allow for qualitative olfactory assessment as possible with the UPSIT [1] or Combined Olfactory Test [8] or discrimination as also possible with Sniffin' Sticks [3]. This deficiency is however the subject of ongoing research and development in our centre and other adaptations will include further automation of the test and the inclusion of half log steps in the threshold testing. The odours PEA and EUC were chosen primarily due to their "purity" as olfactory stimulants at the concentrations used here, however they are also readily miscible with the solvent mineral oil and PEA itself has long enjoyed prominence as an odour in olfactory research [17,24,25]. Ideally it would be useful to test a range of odours for threshold to capture the olfactory spectrum, but this is time consuming and usually coupling other olfactory testing modalities with threshold testing such as in the Sniffin' Sticks is sufficient to provide a reliable test. Once the above modifications have been made to the LOM2, then the aim will be to provide a commercially available tool to allow ENT departments and other units seeking olfactory test devices to utilise this modern form of olfactory testing.

CONCLUSIONS

The LOM2 is a reliable quantitative test that is user-friendly for both the subject and operator, as well as bringing the format of the test into the 21st century and provides a long term cost-effective option, both in terms of time and money for Otorhinolaryngology departments to provide olfactory testing.

REFERENCES

1. Doty RL, Shaman P, Dann M. Development of the University of Pennsylvania Smell Identification Test: a standardized microencapsulated test of olfactory function. Physiol Behav. 1984; 32: 489-502.
2. Doty RL, Shaman P, Kimmelman CP, Dann MS. University of Pennsylvania Smell Identification Test: a rapid quantitative olfactory function test for the clinic. Laryngoscope. 1984; 94: 176-178.
3. Hummel T, Sekinger B, Wolf SR, Pauli E, Kobal G. 'Sniffin' sticks': olfactory performance assessed by the combined testing of odor identification, odor discrimination and olfactory threshold. Chem Senses. 1997; 22: 39-52.
4. Hummel T, Kobal G, Gudziol H, Mackay-Sim A. Normative data for the "Sniffin' Sticks" including tests of odor identification, odor discrimination, and olfactory thresholds: an upgrade based on a group of more than 3,000 subjects. Eur Arch Otorhinolaryngol. 2007; 264: 237-243.
5. Kobal G, Hummel T, Sekinger B, Barz S, Roscher S, Wolf S. "Sniffin' sticks": screening of olfactory performance. Rhinology. 1996; 34: 222-226.
6. Wolfensberger M, Schnieper I, Welge-Lussen A. Sniffin' Sticks: a new olfactory test battery. Acta Oto Laryngologica. 2000; 120: 303-306.
7. Philpott CM, Rimal D, Tassone P, Prinsley PR, Premachandra DJ. A study of olfactory testing in patients with rhinological pathology in the ENT clinic. Rhinology. 2008; 46: 34-39.
8. Robson AK, Woollons AC, Ryan J, Horrocks C, Williams S, Dawes PJ. Validation of the combined olfactory test. Clin Otolaryngol Allied Sci. 1996; 21: 512-518.
9. Cain WS, Gent J, Catalanotto FA, Goodspeed RB. Clinical evaluation of olfaction. Am J Otolaryngol. 1983; 4: 252-256.
10. Cardesin A, Alobid I, Benitez P, et al. Barcelona Smell Test - 24 (BAST-24): validation and smell characteristics in the healthy Spanish population. Rhinology. 2006; 44: 83-89.
11. Saito S, Ayabe-Kanamura S, Takashima Y, et al. Development of a Smell Identification Test Using a Novel Stick-Type Odor Presentation Kit. Chem Senses. 2006; 31: 379-391.
12. Hummel T, Klimek L, Welge-Lussen A, et al. Chemosensory evoked potentials for clinical diagnosis of olfactory disorders. HNO. 2000; 48: 481-485.
13. Rombaux P, Mouraux A, Bertrand B, Guerit JM, Hummel T. Assessment of olfactory and trigeminal function using chemosensory event-related potentials. Neurophysiol Clin. 2006; 36: 53-62.
14. Yousem DM, Williams SC, Howard RO, et al. Functional MR imaging during odor stimulation: preliminary data. Radiology. 1997; 204: 833-888.
15. McNeill E, Ramakrishnan Y, Carrie S. Diagnosis and management of olfactory disorders: survey of UK-based consultants and literature review. J Laryngol Otol. 2007; 121: 713-720.
16. Dawes PJ. Clinical tests of olfaction. Clin Otolaryngol Allied Sci. 1998; 23: 484-490.
17. Doty R, Gregor T, Settle R. Influence of intertrial interval and sniff bottle volume on phenyl ethyl alcohol odor detection thresholds. Chem Senses. 1986; 11: 259-264.
18. Philpott C, Goodenough P, Robertson A, Passant C, Murty G. The effect of temperature, humidity and peak inspiratory nasal flow on olfactory thresholds. Clin Otolaryngol Allied Sci. 2004; 29: 24-31.
19. Philpott C, Goodenough P, Wolstenholme C, Murty G. What solvent for olfactory testing? Clin Otolaryngol Allied Sci. 2004; 29: 667-671.
20. Philpott CM, Wolstenholme CR, Goodenough PC, Clark A, Murty GE. Which variables matter in smell tests in the clinic? J Laryngol Otol. 2007; 15: 1-5.
21. Robinson AM, Gaskin JA, Philpott CM, et al. What is the short term effect of perfumes on olfactory thresholds? J Laryngol Otol. 2007; 121: 755-758.

22. Goonetilleke P, Goodenough P, Philpott CM, Murty GE. Is pollution a key variable in olfactory assessment? Otolaryngol Head Neck Surg. 2007; 137: P45.
23. Philpott C, Wolstenholme C, Goodenough P, Clark A, Murty G. A comparison of subjective perception with objective measurement of olfaction. Otolaryngol Head Neck Surg. 2006; 134: 488-490.
24. Doty RL, McKeown DA, Lee WW, Shaman P. A study of the test-retest reliability of ten olfactory tests. Chem Senses. 1995; 20: 645-656.
25. Cain WS, Krause RJ. Olfactory testing: rules for odor identification. Neurological Res. 1979; 1: 1-9.

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ANNOUNCEMENT

Advanced Course in Septorhinoplasty

M. Scheithauer, G. Rettinger

University Ulm, Germany
March 08-11, 2010

This traditional course will be held in English language. Based on the experience of more than 35 courses, endoscopic demonstrations of endonasal procedures have been developed to a high standard. Open and closed approaches will be performed. This course is laid out for experienced surgeons who like to learn different solutions for typical and difficult problems.

Teachers:
Marc Scheithauer, Gerhard Rettinger (Head) and consultants of the ENT-Department University Ulm

Invited Teachers:
D. Simmen (Switzerland), A.-J. Tasman (Switzerland), H. Gassner (Germany) and K. Ingels (The Netherlands)

Topics of the course are:
- Septal reconstruction, closure of septal perforation
- Surgery of the hump-, prominent- and deviated nose
- Functional and aesthetic surgery of the nasal tip and base
- Special problems: Saddle nose, nasal syndromes, transplants and implants, revision surgery and more…

The course provides:
- Lectures, Live surgery , cadaver dissections (demonstration and hands on **[limited availability]**)

The course is the first segment of an entire **week of surgery** composed of
- Functional and aesthetic rhinosurgery (**Advanced Course**): March 08-11, 2010
- Course on Facial Plastic Surgery: Scar revision and reconstruction of skin defects: March 12, 2010

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