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Plasma 1,8-cineole correlates with cognitive performance following exposure to rosemary essential oil aroma

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Abstract

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Objective

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The mode of influence of the aromas of plant essential oils on human behaviour is largely unclear. This study was designed to assess the potential pharmacological relationships between absorbed 1,8-cineole following exposure to rosemary aroma, cognitive performance and mood.

Methods

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Twenty healthy volunteers performed serial subtraction and visual information processing tasks in a cubicle diffused with the aroma of rosemary. Mood assessments were made pre and post testing, and venous blood was sampled at the end of the session. Pearson correlations were carried out between serum levels of 1,8-cineole, cognitive performance measures and change in mood scores.

Results

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Here we show for the first time that performance on cognitive tasks is significantly related to concentration of absorbed 1,8-cineole following exposure to rosemary aroma, with improved performance at higher concentrations. Furthermore, these effects were found for speed and accuracy outcomes, indicating that the relationship is not describing a speed–accuracy trade off. The relationships between 1,8-cineole levels and mood were less pronounced, but did reveal a significant negative correlation between change in contentment and plasma 1,8-cineole levels.

Conclusion

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These findings suggest that compounds absorbed from rosemary aroma affect cognition and subjective state independently through different neurochemical pathways.

Keywords: aroma, cholinergic, cineole, cognition, essential oil, mood, rosemary

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Introduction

Putative effects of aromas on aspects of human behaviour can be traced back to ancient Greece, where the extracts of aromatic plants were used for cosmetic, religious and medical purposes. Today the popularity of aromas for pleasure, relaxation and in therapeutics is unabated and typified in the ever popular application of aromatherapy [Tisserand, 1993]. The essential oils used in aromatherapy are extracted from natural sources such as plant leaves, fruits, roots and barks. The unique relationships between plant essential oil aromas and any behavioural impact are potentially due to the complex molecular composition containing a range of alcohols, aldehydes, acids, phenols, esters, ketones and terpenes [Hopkins, 1996].

A small, but growing body of research has been carried out to investigate the possible influence of the aromas of essential oils on cognition and mood in the healthy population – see Herz for a review [Hertz, 2009]. Diego and colleagues found subjective mood and objective electroencephalogram (EEG) effects for lavender and rosemary as were predicted based on the aromas' reputed properties [Diego *et al*. 1998]. However, whilst both aromas improved the speed of maths computations, only lavender increased accuracy. Moss and colleagues reported differential effects of lavender and rosemary on aspects of cognition, particularly working memory, but also that rosemary aroma led to an improvement in long-term memory compared with controls [Moss *et al*. 2003]. The potential for equivalence of the impact of herbal supplementation and aroma exposure was investigated by Moss and colleagues [Moss *et al*. 2010]. The authors report largely consistent effects for *Salvia officinalis* aroma but not *Salvia lavandulaefolia* aroma compared with the effects of oral administration of extracts of these herbs as detailed by Scholey and colleagues [Scholey *et al*. 2008].

Consideration of the mechanisms by which essential oil aromas might impact upon behaviour provides a number of possibilities. Given that the properties of aromas are to a great extent defined by folk wisdom rather than scientific evaluation, expectancy might be a reasonable candidate or at least a confounding variable worthy of addressing. Indeed, Moss and colleagues found a complex pattern of relationships between induced expectancies and aroma effects when investigating the influence of chamomile aroma on cognition and mood [Moss *et al*. 2006]. Their findings support to some extent those previously identified elsewhere for the impact of expectancy on physiological measures [Campenni *et al*. 2004], and of priming on relaxation effects under aroma conditions [Howard and Hughes, 2008]. Indeed the latter argue that expectancies and not aroma is the major factor underpinning observed psychophysiological effects. However, Wartik used EEG recording and reported that jasmine produced increased alpha-power in the frontal cortices, indicative of increased arousal and unlikely to be as a result of expectancy [Wartik, 1995]. Furthermore peppermint aroma seems capable of reliably producing small EEG and electromyogram or muscular conductance fluctuations during rapid eye movement and nonrapid eye movement sleep [Badia *et al*. 1990]. The authors suggest that such findings rule out the possible effects of expectancy.

A second potential mode of influence of aromas is the hedonic valence mechanism that describes the relationship between the pleasantness of an aroma, the associated effect on mood and the consequential impact on behaviour/performance [Baron and Bronfen, 1994]. In support of the proposition, Degel and Köster discuss data that run counter to predictions based on received wisdom, namely, the authors report improved mathematical performance for exposure to the 'sedating' aroma of lavender compared with the 'stimulating' aroma of jasmine [Degel and Köster, 1999]. By considering participants' ratings of pleasantness for the two aromas, Degel and Köster identify that the more pleasant lavender was associated with better performance. However, the evidence in support of the hedonic valence mechanism can be difficult to disentangle from other possible explanations based on physiological processes. For example, Degel and Köster go on to consider how the improved performance could be

equally well explained by the sedating effect of lavender reducing arousal in a stressful environment, and so improving performance in accordance with the Yerkes–Dodson law.

The mechanism of interest in the current study, and potentially more valuable regarding the usefulness of aroma as an intervention is the pharmacological mechanism outlined by Jellinek [Jellinek, 1997]. This describes how constituents of the essential oils may influence behaviour through the central nervous or endocrine systems. Volatile compounds (e.g. terpenes) may enter the blood stream by way of the nasal or lung mucosa. Terpenes are small organic molecules which can easily cross the blood –brain barrier and therefore may have direct effects in the brain by acting on receptor sites or enzyme systems. Such compounds have been detected in the blood of rodents exposed to the vapours of essentials oils. Jirovetz and colleagues demonstrated that serum levels of linalool and linalyl acetate in rats following lavender exposure were related to observed sedation [Jirovetz *et al*. 1990]. Kovar and colleagues assayed serum levels of 1,8-cineole, and recorded locomotor activity when rosemary oil was administered by inhalation or orally [Kovar *et al*. 1987]. The data showed that both the inhalation and oral administration of rosemary oil stimulated locomotor activity and that this was related to serum 1,8-cineole concentration. These findings cannot be explained by expectancy theory and suggest that more than simple stimulation of the olfactory organ is involved, with a direct pharmacological action on the central nervous system being implicated.

In vitro neuropharmacological research has also provided some interesting data that are pertinent here. Orhan and colleagues reported that extracts of rosemary displayed significant inhibitory effects on both acetylcholinesterase (AChE) and butyrylcholinesterase enzymes [Orhan *et al*. 2008]. The authors go on to identify that the major active component of the essential oil is 1,8-cineole, a terpene that has previously been identified as possessing anti- AChE activity [Perry *et al*. 2000, 2003; Savelev *et al*. 2003]. Such activity is suggestive of potential cognitive impact and indeed underpins the pharmacological activity of a number of dementia treatments [Orhan *et al*. 2008]. However, it should be stressed that any activity might be a consequence of the synergistic combinations of components present rather than a single compound. The possibility of such a pharmacological mechanism for rosemary aroma would provide supporting evidence to the concept that each individual aroma of an essential oil has its own unique pattern of influence on both cognition and mood as a result of the unique composition of volatile aromatic compounds. To investigate this further, the current study assayed serum for 1,8-cineole and correlated this with cognitive performance under conditions of rosemary aroma exposure.

Method

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Design

The study used a correlational design to investigate a possible relationship between the plasma 1,8-cineole levels and cognitive performance and mood. Treatment was by way of exposure to the aroma of rosemary essential oil. Participants were randomly assigned to be exposed to the aroma in the cubicle for 4, 6, 8 or 10 min prior to completing the cognitive tests. This period was not kept constant so as to facilitate a range of levels of absorption of compounds to take place across the participant group. Total pre-test time was held constant across participants by asking them to wait outside the cubicle for 6, 4, 2 or 0 min dependent on their assigned exposure condition. The time spent completing the tasks did not differ between participants. All participants were blind as to the nature of the study, being informed that the study was investigating the relationship between mood and cognitive performance. On the occasions that participants commented on the aroma the experimenter explained that the aroma had ‘nothing to do with me’ and that it was ‘left over from a previous study’. When asked at the end of testing and prior to debriefing, none of the participants indicated that they felt that the aroma had affected them in any way. To assess any relationship between ‘pleasantness’ of the

aroma (i.e. hedonic valence) and performance measures, subjective ratings were obtained from each participant at the end of testing as in previous studies [e.g. Moss *et al*. 2010].

Participants

Twenty healthy volunteers [12 women, mean age 23.2 years, standard deviation (SD) 3.2 and 8 men, mean age 22.6 years, SD 2.9] took part in the study. All volunteers completed a health screening questionnaire prior to participation. None were excluded from the study.

Testing cubicles

The testing cubicle measured 2.4 m long × 1.8 m wide × 2.4 m high and was maintained at a temperature of between 18°C and 22°C throughout the testing sessions. The door was kept closed except for participant access.

Aromas

'Tisserand' pure essential oil (Tisserand Aromatherapy, Newtown Road, Hove, Sussex, UK) of rosemary was used to produce the ambient aroma. Four drops of the oil were applied to a diffuser pad for a 'Tisserand aroma stream'. The aroma stream was placed under the bench in the testing cubicle and was switched on for 5 min prior to the introduction of each participant.

Cognitive measures

Serial threes subtraction task A starting number between 800 and 999 was displayed on the computer screen. The participant was asked to subtract three from this number and enter their answer using the key pad; they were then required to subtract three from this answer and enter it likewise. They were instructed to continue in the same way until the programme stopped after 2 min.

Serial sevens subtraction task A starting number between 800 and 999 was displayed on the computer screen. The participant was asked to subtract seven from this number and enter their answer using the keypad, they were then required to subtract seven from this answer and enter it likewise. They were instructed to continue in the same way until the programme stopped after 2 min.

Rapid visual information processing task Participants were presented with a continuous series of digits in the centre of the screen and they were asked to detect sequences of any three consecutive odd digits or any three consecutive even digits by pressing the space bar. The task stopped automatically after 3 min.

All tasks were drawn from the Computerized Mental Performance Assessment System battery developed by the Brain Performance and Nutrition Research Centre at Northumbria University. The tasks were selected for a number of reasons. Firstly they tap into a number of cognitive domains: working memory, executive function and sustained attention, and represent functions involved in a wide range of real world activities [Ritter *et al*. 2007]. Secondly these tasks have previously been demonstrated to be sensitive to the impact of herbal and dietary based interventions [e.g. Reay *et al*. 2006; Scholey *et al*. 2010], and they have been identified as possessing different degrees of cognitive demand based on participants' self reports [Scholey *et al*. 2001; Scholey and Kennedy, 2002]. This was considered potentially important as some interventions have been shown to differentially affect task performance based on cognitive load [e.g. Kennedy *et al*. 2002].

Subjective mood measures

Mood was assessed using the Bond-Lader visual analogue scales [Bond and Lader, 1974]. The scales were developed for use in medical psychology and psychopharmacology research, with the original

paper cited in more than 1000 peer reviewed publications including clinical trials worldwide. A critical review has described them as ‘A simple technique for measuring subjective experience. They have been established as valid and reliable in a range of clinical and research applications’ [McCormack *et al*. 1988, p. 1007]. The Bond–Lader mood scales have also been used widely in aroma and other herbal intervention studies where they have been demonstrated to be sensitive to changes in subjective state [e.g. Moss *et al*. 2003, 2006, 2008, 2010; Scholey *et al*. 2010; Kennedy *et al*. 2002]. The 16 visual analogue scales of the Bond-Lader assessment were combined as recommended by the authors to form three mood factors: ‘alert’, ‘calm’ and ‘content’.

Procedure

Ethical clearance was granted by the School of Psychology and Sport Sciences Ethics Committee. Recruitment took place 1 week prior to testing and at this point participants were told the aims of the study and given a time to attend the laboratory. All testing took place in the same cubicle between 9:00 am and 12:00 noon. Participants were asked to read a brief for the study and instructions for task completion prior to providing informed consent. A pre-test mood scale was completed prior to entering the aroma-infused cubicle. The cognitive tasks were completed followed by a second (post-test) mood scale. Finally, a sample of venous blood was taken by a trained phlebotomist. The participants were then debriefed, thanked for their participation and any questions answered.

Blood sampling and analysis

One 5 ml blood sample was taken per participant, by venous puncture into a serum gel monovette. All samples were immediately centrifuged at 3000g for 10 min at 20°C using an Allegra X-22 centrifuge (Beckman Coulter Ltd, High Wycombe, UK). The serum was then decanted into a 1.5 ml microtube and stored in a freezer at −80°C until all the samples were ready for analysis.

Standards

Terpinolene was used as an internal standard in the serum samples; it was added at a concentration of 2 ppm to all samples. A calibration set was also made up using standard 1,8-cineole and terpinolene at similar concentrations.

Extraction

The sample (0.9 ml) containing 2 ppm internal standard was loaded onto a 2.5 ml C8 solid phase extraction cartridge that had been activated using methanol and subsequently washed with distilled water. The sample was allowed to flow through at a slow rate to maximize residence time. After all of the sample had passed through the column it was washed with distilled water. Elution of adsorbed substances was achieved by washing through with 1 ml methanol. The eluted sample was placed in a gas chromatography sample vial ready for analysis.

Analysis

Analysis was carried out using a Thermo gas chromatograph fitted with a DSQ mass spectral detector (Thermo Fisher Scientific, Austin,Texas, USA) operating in single ion monitoring mode with splitless injection to maximize sensitivity (the principal ions had been determined in full scan mode with the prepared standards). The initial column temperature was 40°C, rising at a ramp rate of 5°C per min to 110°C followed by a second ramp of 20°C per min to 300°C.

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Results

1,8-Cineole assay

Quantification was achieved by comparing the peak area for 1,8-cineole with that of the internal standard and relating this to the concentration of the internal standard after applying a correction factor for the relative response factors of 1,8-cineole and the internal standard. The retention time for 1,8-cineole was 9.66 min and that of the internal standard, terpinolene, was 11.13 min (Figure 1). The desired graduation in serum 1,8-cineole concentration was achieved across the participant sample as evidenced by the significant correlation between pre-test exposure time and serum 1,8-cineole levels, $r(18) = 0.727$, $p < 0.001$.

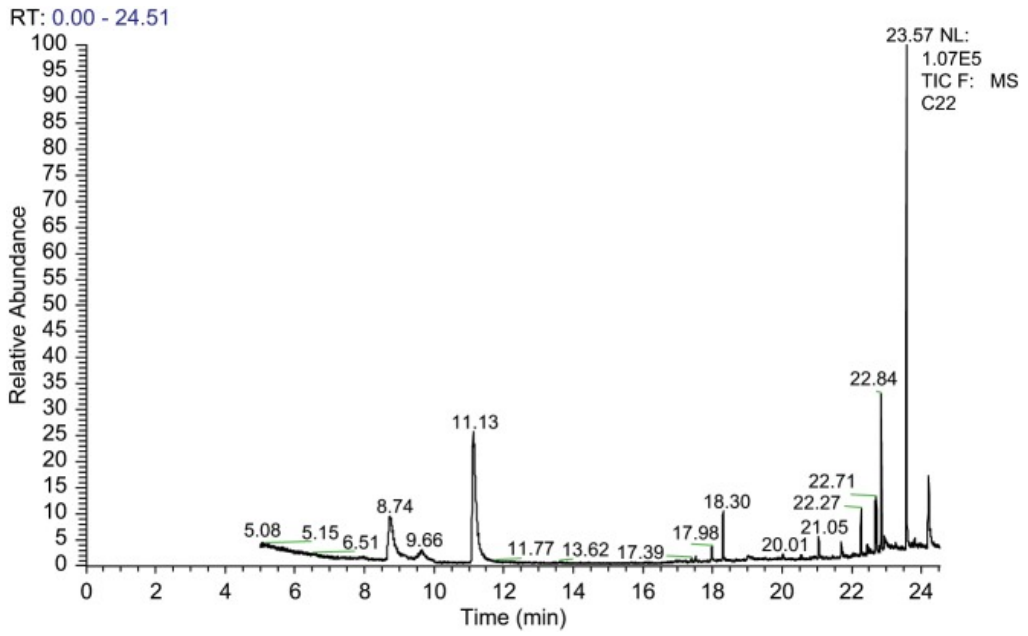

Figure 1.

Chromatogram for a 5 ppm concentration standard. 1,8-Cineole comes out at 9.62 min and the internal standard, terpinolene, at 11.12 min.

Data were analysed using SPSS V.16. Pearson correlations were performed to determine the degree of relationship between plasma 1,8-cineole and the behavioural variables (cognition and mood).

Cognitive performance measures

Serial threes subtraction task

A positive linear relationship was found between serum 1,8-cineole concentration and the number of correct answers on the serial threes subtraction task: $r(18) = 0.469$, $p = 0.037$; $r^2 = 0.22$.

A negative linear relationship was found between the reaction time of participants on the serial threes subtraction task and the serum 1,8-cineole concentration: $r(18) = -0.502$, $p = 0.024$; $r^2 = 0.25$.

Serial sevens subtraction task

A positive linear relationship that approached but did not quite reach statistical significance was found between the number of correct answers on the serial sevens subtraction task and serum 1,8-cineole concentration: $r(18) = 0.433$, $p = 0.056$; $r^2 = 0.19$.

A negative linear relationship was found between the reaction time of participants on the serial sevens subtraction task and serum 1,8-cineole concentration: $r(18) = -0.466$, $p = 0.038$; $r^2 = 0.22$.

Rapid visual information processing No relationship was found between the number of correct responses on the rapid visual information processing (RVIP) task and serum 1,8-cineole concentration: $r(18) = 0.117$, $p = 0.624$; $r^2 = 0.01$.

A negative linear relationship was found between reaction time on the RVIP task and serum concentration of 1,8-cineole: $r(18) = -0.446$, $p = 0.049$; $r^2 = 0.19$.

Mood measures

To calculate the change in the three mood variables, pre-test values were subtracted from post-test values.

Alertness No relationship was found between serum concentration of 1,8-cineole and pre to post change in alertness: $r(18) = 0.069$, $p = 0.773$; $r^2 = 0.01$.

Contentedness A negative linear relationship was found between 1,8-cineole concentration and the change in contentedness: $r(18) = -0.454$, $p = 0.044$; $r^2 = 0.21$.

Calmness No relationship was found between serum 1,8-cineole concentration and the change in calmness: $r(18) = -0.268$, $p = 0.253$; $r^2 = 0.07$.

Hedonic valence No relationship was found between self-reported pleasantness of the aroma and any of the performance variables, $p > 0.05$ in each case.

Discussion

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The results reported here support the proposal that 1,8-cineole would be detectable in the blood serum of healthy human volunteers following inhalation of the aroma of rosemary essential oil. Previously only demonstrated in animals [Jirovetz *et al*. 1990; Kovar *et al*. 1987], this study supports the suggestion that active compounds present in aromas may be absorbed through the nasal or lung mucosa and thus provide the potential for pharmacological activity as outlined by Jellinek [Jellinek, 1997]. The small size of these lipid soluble compounds facilitates passage across the blood–brain barrier [Boyle *et al*. 2005] and consequently they may produce effects at the neuronal level by either acting directly on receptor sites, or indirectly by impacting on enzyme activity. 1,8-Cineole is one of a number of volatile organic compounds present in the essential oil of rosemary. Typically between 35% and 45% by volume of rosemary essential oil, 1,8-cineole may possess direct pharmacological properties [Perry *et al*. 2000, 2003] or may serve as a suitable marker for the absorption of highly active compounds such as rosmarinic acid and ursolic acid that are present at much lower concentrations in rosemary essential oil. Orhan and colleagues [Orhan *et al*. 2008] reported powerful AChE and butyrylcholinesterase inhibition by rosmarinic acid extracted from rosemary essential oil, which as a whole was found to possess moderate inhibition of AChE in keeping with previous data [Perry *et al*. 1996]. Similarly ursolic acid is a potent inhibitor of AChE [Chung *et al*. 2001], which is found in rosemary essential oil [Huang *et al*. 1994]. Taken together these findings strongly support the contention that inhalation of rosemary essential oil would possess the potential to impact on cognitive performance in a positive manner by preventing the breakdown of acetylcholine and so perpetuating cholinergic stimulation. One further important consideration that should be taken into account here is the evidence provided by Savelev and colleagues that demonstrates the presence of synergistic and antagonistic relationships

between compounds with AChE properties extracted from *Salvia* species [Savelev *et al*. 2003]. It seems highly likely if not inevitable that such interactions would exist between compounds extracted from rosemary and consequently that composition–impact relationships are not easy to predict.

In addition to the aforementioned effects on the cholinergic system, Machado and colleagues used an animal model to demonstrate that a hydroalcoholic extract of rosemary interacts with the monaminergic system in a similar manner to antidepressant drugs [Machado *et al*. 2009]. Additionally the authors report significant effects on the noradrenergic and dopaminergic systems. Such interactions are potentially the cause of mood effects reported in humans following exposure to rosemary aroma. When considered in the round a reasonable conclusion is that each plant species (and by virtue their essential oils) may have a distinct influence on cognition and mood as a result of the unique composition of volatile compounds present. Furthermore given that the biochemical composition of plants is highly dependent upon geographical location, growing conditions and indeed time of day [Sousa *et al*. 2010] the determination of standardized extracts is one that presents serious challenges for the progress of phytomedicine.

With regard to the behavioural effects of exposure to rosemary essential oil aroma, the results reported here support previous work indicating that rosemary aroma can influence cognitive performance and mood [Moss *et al*. 2003]. Here, serum levels of 1,8-cineole were correlated with the performance outcomes (number of correct responses and reaction times) for each task. Although it is wise to exercise caution when dealing with small samples, effect sizes (r^2) of between 0.1 and 0.3 (medium to large effects by Cohen's definitions) [Cohen, 1992] were found for all cognitive task variables except the accuracy of the RVIP task for which no relationship was identified between 1,8-cineole and percentage correct hits. These effect sizes compare favourably to those found previously in our laboratory for the effect of rosemary aroma on cognition. Moss and colleagues found r^2 values of 0.06 and 0.07 for the aspects of long-term and working memory that were found to be enhanced by exposure to aroma compared with controls [Moss *et al*. 2003].

The tasks used in this study were selected to tap into different cognitive processes: the RVIP task provides an assessment of sustained attention and central executive function compared with the continuous working memory, arithmetic processing and central executive composition of the serial subtractions tasks. Furthermore, the opportunity was taken to observe effects on tasks with discreet differences in cognitive load; with the serial sevens task being considerably more demanding than the serial threes task [Scholey and Kennedy, 2002]. Comparison of the relative relationships between plasma 1,8-cineole and performance on the serial subtraction tasks indicates that 1,8-cineole (or other essential oil components as discussed above) had a greater impact on the task with a lower cognitive load. Analysis of the difference between the correlation coefficients shows a stronger relationship between 1,8-cineole for accuracy [$t(15) = -3.27$, $p = 0.002$] and speed [$t(15) = -6.06$, $p < 0.001$] for the serial threes task than for the serial sevens task. The finding that the strength of relationships differs between tasks implies that there may be an interaction with cognitive load, and that at some point task difficulty might be such that enhancement is not possible as a consequence of exposure to rosemary aroma. Alternatively, more of the active compounds might need to be absorbed to have an effect. Increased levels of absorption would also provide opportunity to observe the classic inverted U-shaped function that underpins pharmacological impacts on behavioural variables of all kinds [Yerks and Dodson, 1908]. No relationship was found for the RVIP task that has a low cognitive load. It would appear that a demand on memory processes is required for enhancement to be observed, and that attention-based resources of the kind involved in the RVIP task may not be available for improvement. This supports findings reported by Moss and colleagues when no effect was found for rosemary aroma on the accuracy of attention factor [Moss *et al*. 2003]. Importantly, consideration of the relationships between 1,8-cineole and reaction times on the subtraction tasks indicates that for both tasks the

improved accuracy is not the consequence of a speed–accuracy trade off. Clearly there is scope for further investigation of the precise cognitive components that might be available for modulation by essential oils, with previous research largely focusing on tasks such as mathematical computations [e.g. Ludvigson and Rottman, 1989; Diego *et al*. 1998; Degel and Köster, 1999] or more global cognitive factors [e.g. Moss *et al*. 2003, 2008, 2010]. Cognitive psychology has much to say about the fractionation of memory systems [Baddeley, 1996] and it would be valuable to use such arguments to inform future studies of the effects of aromas to better understand exactly how any effects are underpinned by the cognitive architecture.

Table 1.

Means and standard deviations for serum 1,8-cineole concentrations, cognitive task performance, baseline and change in mood (a positive value indicates an increase for that dimension over the experimental session).

	Mean	SD	Skewness	Kurtosis
1,8-Cineole (mg/ml)	0.105	0.024	−0.29	−0.48
Threes correct responses	23.4	11.8	0.55	−0.03
Threes RT (ms)	5593.9	2846.8	1.11	0.18
Sevens correct responses	12.4	9.2	1.60	0.96
Sevens RT (ms)	8657.3	3790.8	0.51	−0.67
RVIP correct responses	11.8	7.9	0.85	−0.23
RVIP RT (ms)	524.1	82.5	0.53	1.35
Alert baseline (mm)	47.6	15.4	−0.863	0.735
Content baseline (mm)	57.6	12.8	−0.533	−0.293
Calm baseline (mm)	48.6	19.0	−0.439	−0.967
Alert change (mm)	14.0	16.6	0.06	−0.18
Content change (mm)	13.1	15.8	1.39	1.52
Calm change (mm)	8.7	21.1	0.47	1.13

RVIP, Rapid visual information processing task; SD, standard deviation.

Table 2.

Pearson correlation coefficients between all variables.

	Threes correct	Threes RT	Sevens correct	Sevens RT	RVIP correct	RVIP RT	Alert change	Content change	Calm change
	0.469*	−0.502*	0.433	−0.466*	0.117	−0.446*	0.069	−0.454*	−0.268

	Threes correct	Threes RT	Sevens correct	Sevens RT	RVIP correct	RVIP RT	Alert change	Content change	Calm change
1,8 Cineole									
Threes correct	-	−0.864***	0.880***	−0.466*	0.327	−0.167	−0.323	−0.565**	−0.264
ThreesRT	-	-	−0.692***	−0.819***	−0.291	0.190	0.269	0.525*	0.299
Sevens correct	-	-	-	−0.801***	0.323	−0.093	−0.247	−0.482*	−0.303
Sevens RT	-	-	-	-	−0.106	−.070	0.145	0.353	0.512*
RVIP correct	-	-	-	-	-	−0.161	−0.043	−0.115	0.224
RVIP RT	-	-	-	-	-	-	0.192	0.315	−0.118
Alert change	-	-	-	-	-	-	-	0.449*	−0.009
Content change	-	-	-	-	-	-	-	-	0.295

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Significant correlations are marked with asterisks *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$.

RT, Reaction Time (milliseconds); RVIP, Rapid visual information processing task; SD, standard deviation.

The observation that speed for the RVIP task was also improved may indicate that 1,8-cineole has a general psychomotor effect that is independent of cognitive resources, or even an effect on the peripheral cholinergic system that controls movement in a manner similar to that of other cholinergic drugs [Salamone *et al*. 1986]. This suggestion of a general psychomotor effect links interestingly to findings reported by Schifferstein and colleagues [Schifferstein *et al*. 2011] who reported enhanced dancing activity for all odorants tested compared with controls, irrespective of purported properties. It may be parsimonious to suggest that the perception of smells produces a global psychomotor enhancement, but the evidence does suggest somewhat greater specificity. For example, Moss and colleagues report a slowing of response speed during exposure to ylang ylang, something not associated with peppermint in the same study, indicating differential effects of the two aromas [Moss *et al*. 2008]. An alternative possibility regarding the finding of improved reaction time on all three tasks might be that 1,8-cineole affects speed as a consequence of increased subjective alertness of participants. However, correlations performed here between 1,8-cineole and the subjective mood reports suggest otherwise, with no strong evidence present that change in subjective ratings of alertness bore any relationship to plasma 1,8-cineole levels. Only contentedness possessed a significant relationship with 1,8-cineole levels, and interestingly to some of the cognitive performance outcomes, leading to the intriguing proposal that positive mood can improve performance whereas aroused mood cannot. Previously, Moss and colleagues suggested that the impact of aromas on task performance was independent of subjective feelings [Moss *et al*. 2003]. Others [e.g. Warm *et al*. 1991; Itai *et al*. 2000] have also argued for the independence of effects of aromas on cognition and mood, proposing avenues

of influence which are not related to psychological beliefs and expectations. Such proposals sit well with the pharmacological mechanisms described above. Whether the effects on mood found here and elsewhere are a consequence of interactions of compounds with the monoaminergic system is an intriguing possibility worthy of further investigation. The relationship between the Bond-Lader mood scales used here, and those of another widely used scale in environmental research, the Pleasure, Arousal, Dominance (PAD) scale has yet to be established. The PAD scale [Mehrabian and Russell, 1974] was developed at the same time as Bond and Lader were working on their scale, and has been described as 'the premier measure in the area of environmental psychology for assessing the impact of the environment on people' [Machleit and Eroglu, 2000, p. 102]. Given the considerable value of the two scales for assessing mood states it is perhaps surprising that they have not previously been explored in tandem. It would be of considerable interest to incorporate both scales in future work to identify the relationship between the two scales components. This would permit greater comparisons between existing and future work to be made. A further consideration regarding the use of self-report scales generally is that the relationship between self–report measures and physiological correlates of arousal tends to be inconsistent [e.g. Mikalsen *et al*. 2001]. Indeed self-reports have long been seen to depend upon the cognitive explanations available to the individual to interpret perceived changes in their state of arousal [Schachter and Singer, 1962]. For example, where physiological changes are expected, these tend to be under reported as changes in subjective state due to cognitive preparedness. In contrast where physiological changes are unexpected, changes in subjective state tend to be over reported due to the salience of the change in physiological arousal. This can be of particular concern in blind designs, or studies with potentially or directly misleading instructions of the kind often used for aromas, when the causes of changes in arousal may be difficult for participants to attribute. Such difficulties can produce problems for self-report measures and caution is advised when interpreting results. It is very important to recognize that the self-report mood scales used in this study are not seen as substitutes for or estimates of measures of physiological arousal, and that subjective alertness might not be dependent on changes in such measures as heart rate and blood pressure, or indeed any other measure of physiological arousal. However, the impact of aromas on the more subtle aspects of psychological mood state are still of interest – even when they appear not to be related to physiology or performance measures as here.

To further our understanding of the effects and mechanisms underpinning the behavioural impact of rosemary aroma, combined *in vivo* and *in vitro* studies need to be carried out to assess both pharmacological and behavioural properties of a single source plant strain. As well as AChE inhibition, receptor-binding properties should be investigated as previous research has shown herbal extracts to exhibit acetylcholine receptor activity, including nicotinic [Perry *et al*. 1996; Wake *et al*. 2000] and muscarinic [Wake *et al*. 2000] binding properties in human cerebral cortex tissue. If these assays are made in tandem with cognitive and mood assessments it would help confirm that rosemary possesses cholinergic properties, and that such properties underpin the cognitive effects reported following inhalation of rosemary aroma.

Footnotes

Go to:

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There are no conflicts of interest (real or apparent) that may have a direct bearing on the subject matter of this article.

Contributor Information

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