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Physiology Section

Effect of Herbal Perfumes and Synthetic Perfumes on Attention and Cognitive Functions in Young Adults: A Sequential Cross-sectional Study

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ABSTRACT

Introduction: Focused and sustained attention, executive function, working memory and task switching are important cognitive abilities. Efforts to enhance cognitive abilities, have engaged the scientific community since ages. Human beings are exposed to several fragrances and the perception of the sense of smell, plays an important role in the physiological effects of mood, stress, and working capacity. Perception of olfactory stimuli involves complex brain processing, which can be directly associated with cognition and emotion. Several Electroencephalogram (EEG) studies have revealed that, fragrances significantly modulate the activities of different brain waves and are responsible for various states of the brain.

Aim: To objectively demonstrate the effect of olfactory stimulation with herbal (rose) perfume on attention and executive functions.

Materials and Methods: This sequential cross-sectional study was conducted in the Department of Physiology at Osmania Medical College, Hyderabad, Telangana, India. The duration of the study was one month, from 16th August 2021 to 16th September 2021. Study included 40 healthy participants (15 males and 25 females), aged between 18-30 years to olfactory stimulation by herbal (rose) extract perfume and synthetic (rose) perfume, and the effects are assessed, using the psychophysiological tests viz., Rapid Serial Visual Presentation task (RSVP), Trail Making Tests (TMT) A&B, Ruff Figural Fluency Test (RFT), and compared with the baseline scores, which are then analysed using Paired t-test. The data was analysed by using Statistical Package for Social Sciences (SPSS) version 16.0.

Results: Olfactory stimulation with herbal rose perfume enhances the attention and speed of processing, as shown by the nearly significant improvement in the performance of the TMT-A (p-value=0.05433), with decreased time taken to perform the test, whereas, the synthetic rose perfume enhanced the attention, speed, switching between tasks, cognitive fluency, and flexibility. This is evident by the significant improvement in the performance of the TMT-A (p-value=0.00326) and TMT-B (p-value=0.0001693), with decreased time taken to perform the tests. The derived index TMT (B-A), which removes the speed component and is a purer index of executive functions, also showed significant results (p-value=0.002092).

Conclusion: The effects of herbal and synthetic rose olfactory stimulation have shown positive influence on the attention and cognitive functions and this finding can be applied to create student friendly learning environments, by providing pleasant stimuli through means of aromatic sprays delivering the odorant in a pulsatile manner at a frequency to overcome the ensuing olfactory adaptation.

Keywords: Cognition, Coronavirus disease-19, Executive function, Odorants, Psychophysiological tests

INTRODUCTION

Human beings are exposed to several fragrances in daily life, pleasant or unpleasant, which impacts behaviour and functioning. Smell effects emotions and is related to memory of events. In addition to perception of odour, olfaction influences emotions, memory, and sexual behaviour, through its connections with limbic and hypothalamic structures [1]. 'Aromachology' is the term, that was coined by the sense of smell Institute in 1982, which refers to the scientific analysis of olfactory effects on mood, physiology, and behaviour [2]. Enhancement of cognitive abilities by olfaction is an interesting physiological effect, which has been intriguing scientists for a significant period of time. Recent studies have shown the effect of aroma inhalation effecting cognition, mood, as well as, social behaviour. Olfactory stimulation has many psychophysiological effects [3-5].

As olfactory stimulation can affect attention and execution of tasks, the present study was conducted to understand the effect of odorant molecule on cognition, specifically of rose, which is commonly used but not well studied. Understanding the physiology and differential influences of different agents will help provide scientific basis to create student friendly learning atmospheres, and further, study the possible application and efficacy in cognitive disorders. Moreover, there is also a palpable lack of literature, on the comparison of effect of natural and synthetic odorant molecules [4]. The present study, therefore, aimed to objectively demonstrate the effect of olfactory stimulation with herbal perfume and synthetic perfume, on attention and executive functions, the two important components of cognitive functions.

MATERIALS AND METHODS

This is a sequential cross-sectional study was conducted in the Department of Physiology at Osmania Medical College, Hyderabad, Telangana, India. The duration of the study was one month {just after the wave of Coronavirus Disease-19 (COVID-19)}, from 16th August 2021 to 16th September 2021. For the study, ethical guidelines were followed, and approval bearing IEC number: (ECR/300/Inst/AP/2013/RR-19), was obtained before starting the study.

Sample size calculation: The sample size was calculated using the formula $n=\frac{Z^2 \times P(1-P)}{d^2}$.

Assuming a confidence level of 95% (Z=1.96), a margin of error of 10% (d=0.1), and an estimated prevalence of 50% (p=0.5), the sample size n=96.04 [6].

Inclusion criteria: Fifteen healthy male and 25 healthy female participants aged between 18-30 years, were included in the study.

Exclusion criteria: Participants with history of sinusitis, upper respiratory tract infections, Coronavirus Disease-19 (COVID-19), deviated nasal septum, anosmia, asthma and any allergies were excluded from the study.

Study Procedure

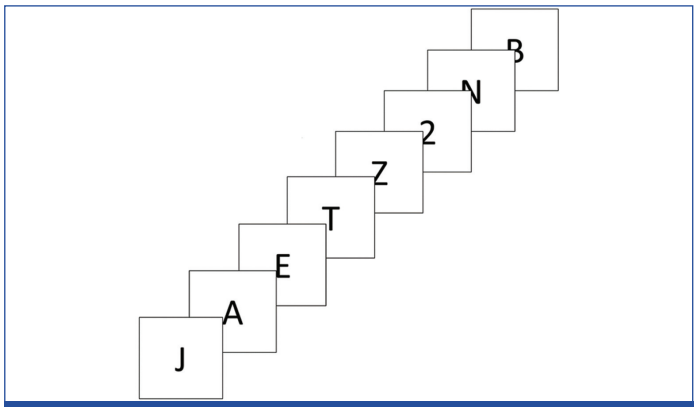
Rose herbal extract and synthetic rose perfumes, which are readily available in the market, were used and an undiluted solution was used for the study. Rose herbal extract was purchased from 'Bin Mahfooz Perfumers, Hyderabad' who prepared the herbal extract from rose plant after a series of distillations. The synthetic rose perfume brand was purchased from' Al-Haramain Perfumers, Hyderabad'. There is no conflict of interest pertaining to the dealers in perfumes. Ten drops of undiluted perfume were put on the filter paper which was placed at arm's length from the participant. A stopwatch was used to measure the time taken to complete each of the psychophysiological tests.

The psychophysiological tests were all free to use and derived from public access sources. The tests done included:

- **Rapid Serial Visual Presentation task (RSVP) [7,8]:** It is a task associated with alertness and vigilance. It is detection of a target digit amongst a series of distracter digits moving through the screen at the rate of 0.10 seconds [9,10]. The test included 50 slides with 10 target slides. Scoring was done based on the number of target letters identified.
- **Trail Making Tests (TMT) A and B [11,12]:** It provides measure for executive control processes. TMT- A requires the subject to draw lines and connect circled numbers in a numerical sequence from 1-25, whereas, TMT-B involves connecting circled numbers and letters in an alternating alpha-numeric sequence (1-A-2-B to 13-L). Score is time taken to complete the task (in seconds) including time for correction of errors.
- **Ruff Figural Fluency test (RFFT) [13-15]:** It assesses for non verbal fluency, an indirect measure of cognitive flexibility. RFFT consists of 40 boxes having five dots, each on a sheet of paper. The objective was to draw dissimilar or unique pattern in these boxes by joining the five dots. Scoring is done by providing five marks for each dissimilar pattern including unique rotations drawn. Number of repeated designs is counted as perseveration errors, and these yield a negative score. Further, the derived index i.e., the ratio score is calculated as: (perseverative errors/unique designs).

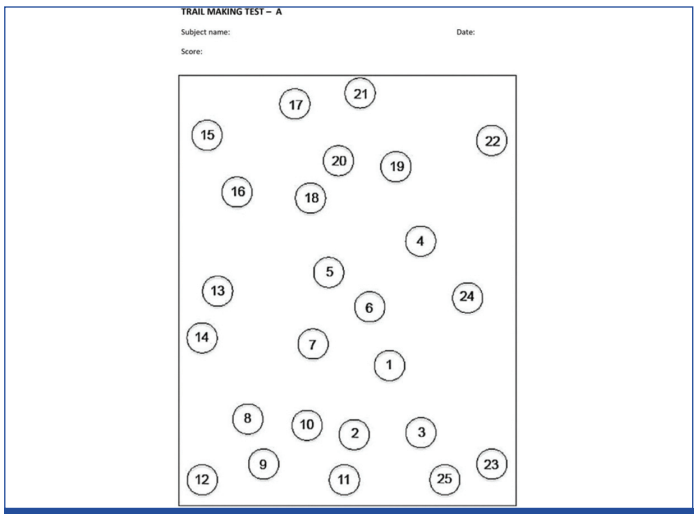
The tests were performed in a well-lit, closed room, during the daytime, in multiple batches maintaining social distancing measures. The subjects were advised to come after a good night sleep, without applying any perfumes or deodorants, to exclude the possibility of interference by other odours. In the first phase of tests, the baseline scores of psychophysiological tests were recorded, without any exposure to the rose fragrance. During the second phase, conducted after a duration of three days, the subjects were subjected to olfactory stimulation for five minutes before the beginning the test and maintaining exposure throughout, and with sniffs in between each test, using 10 drops of herbal extract on a filter paper placed at one arm distance in front of the subject. The psychophysiological tests were initiated after five minutes of exposure to the olfactory stimulation. The exposure was constant and continuous throughout the period of test, asking the subjects to give a strong sniff between each test, to maximise the olfactory stimulation.

Following a gap of three days after the second phase, the subjects were subjected to olfactory stimulation using 10 drops of synthetic rose perfume on a filter paper placed at one arm distance in front of the subject. The psychophysiological tests were initiated after five minutes of exposure to the olfactory stimulation. The exposure was constant and continuous throughout the period of test, asking the subjects to give a strong sniff between each test, to maximise the olfactory stimulation. The psychophysiological tests of each phase, were of a different pattern to increase the reliability of the test, despite decreasing the proposed duration between each phase due to the challenge of availability of subjects during the pandemic. The psychophysiological tests were chosen ensuring major components of cognitive functions can be calculated from them. The RSVP is a task associated with alertness and presentation task vigilance [Table/Fig-1] [10].

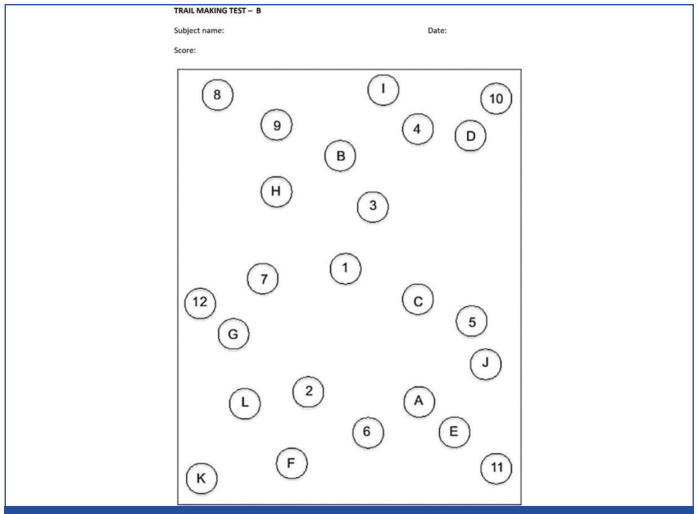


[Table/Fig-1]: A representation of the RSVP slides.

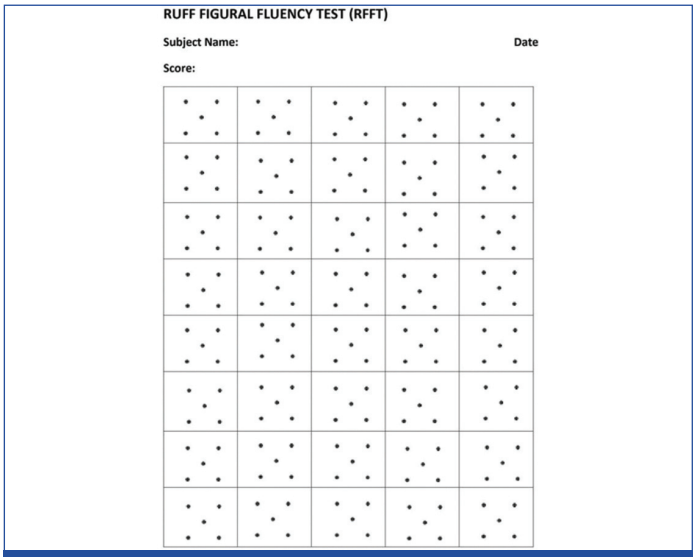
The TMT provides information on visual search, scanning, speed of processing, mental flexibility, and executive functions. Apart from the direct TMT scores, that is, time to complete parts A and B, derived scores are used as sensitive measures of cognitive inflexibility and executive dysfunction. These derived indices include the difference score (B-A), the ratio score (B/A), and the proportional score (B–A/A) [Table/Fig-2,3]. The (B-A) and (B/A) scores can elucidate the added task requirements of TMT-B and are considered as purer indices of the complex cognitive processes involved in part B. Specifically, the (B-A) difference score removes the speed component from TMT assessment and provides a purer indicator of executive control processes, by minimising visuoperceptual and motor demands. On the other hand, the (B/A) ratio score captures the essence of cognitive flexibility, by diminishing the influence of psychomotor demands and controlling for intrasubject variability factors. Finally, the (B–A/A) proportional score has been referred as a sensitive index of prefrontal cortex functioning. The direct TMT scores are mostly affected by age and education in different sociocultural environments [12].



[Table/Fig-2]: TMT-A: Sample figure from Hannah M. Lindsey. Neuropsychological features of civil litigators and criminal offenders: A comparative analysis of brain, behavior, and cognitive functioning 2012 [17].



[Table/Fig-3]: TMT-B: Sample figure from Hannah M. Lindsey. Neuropsychological features of civil litigators and criminal offenders: A comparative analysis of brain, behavior, and cognitive functioning. 2012 [17].



[Table/Fig-4]: Ruff Figural Fluency Test (RFFT).

The RFFT gives an insight about the non verbal capacity for initiation, planning, and divergent reasoning. RFFT was created by Ruff R to be a non verbal analogue of verbal fluency measures. Fluency itself is understood as the ability to apply some strategies to generate non verbal responses maximising response efficiency and simultaneously minimising response repetition [13]. RFFT is the first test to capture figural fluency in a reliable and valid manner, and also is a quick and simple test to administer. The main outcome of the RFFT is the total number of unique designs, which range from 0 points (worst score) to 175 points (best score) [15,16]. Thus, each unique design would be given five points. A total number of designs (or a total number of unique designs) drawn is the indicator of user performance in the RFFT, while the error ratio indicates correctness of performance taking into consideration a number of perseverative errors in relation to a total number of designs. Those two indices are used to assess an individual figural fluency, which refers to the cognitive ability to come up with unique patterns or designs or figures.

The battery of psychophysiological tests employed in the present study, thus, indicates alertness and vigilance, visual search, scanning, speed of processing, mental flexibility, and executive functions, and non verbal capacity for initiation, planning, and divergent reasoning.

Following are the parameters that were recorded in the psychophysiological studies:

- RSVP: Number of target letters identified.
- TMT A and B: Time taken to complete TMT-A and TMT-B separately, and the derived indices which include the difference score (B-A), the ratio score (B/A), and the proportional score (B-A/A).
- RFFT: Total number of unique designs drawn, total number of perseverative errors (repetitive designs) and the derived index i.e., the ratio score: (perseverative errors /unique designs).

The study was conducted in three phases: baseline without any olfactory stimulation, with olfactory stimulation using herbal and then synthetic perfume. Owing to the COVID-19 situation and difficulty to enrol more subjects, the duration between each phase was reduced to three days. The test-retest reliability, for psychophysiological tests in normal individuals. Especially, RFFT is adequate to high, over intervals of three weeks upto 12 months [Table/Fig-4] [13].

Despite this limitation, the study was conducted with reduced intervals but using different patterns for each of the psychophysiological test (changing of target and non target characters in RSVP; changing of the sequence but maintaining the same length of pen-drag in TMT A and B; using different arrangement of five dots in the RFFT), with the assumption that the change in patterns of the test, but sticking to its original format would overcome the reliability issue.

STATISTICAL ANALYSIS

The data was entered in Microsoft Excel 2007 and was analysed by SPSS version 16.0. Paired t-test was used to find the significance between mean values of the different psychophysiological tests in each phase. Data has been presented as mean, standard deviation or 95% confidence intervals. The p-value<0.05 was considered as statistically significant.

RESULTS

The statistical analysis of the baseline values, of the psychophysiological tests, which were done without any olfactory stimulation, and the values with olfactory stimulation using herbal rose perfume, shows that, there is nearly significant improvement in the performance of the TMT-A (p-value=0.05433), with decreased time taken to perform the test with herbal rose olfactory stimulation. There is also significant result seen in the RFFT ratio score (p-value= 0.001586). The other parameters and derivatives have not shown any significant results with using herbal rose olfactory stimulation [Table/Fig-5].

Test	Mean±SD (baseline)	Mean±SD (herbal)	p-value
TMT-A (seconds)	24.00375±6.016	19.69±4.401	**0.05433**
TMT-B (seconds)	45.6495±13.119	39.404 ±11.247	0.3403
TMT B-A	21.64575±11.118	19.714±10.219	0.6009
TMT B/A	1.95807308279357±0.541	2.038034±0.577	0.6891
TMT(B-A)/A	0.95807308279357±0.541	1.038034±0.577	0.6891
RFFT total number of unique designs	26.85±7.353	31.775±6.370	0.3741
RFFT perseverative errors	1.9±2.550	2.1±2.121	0.2555
RFFT ratio score	0.08288683234457 13±0.137	0.069719±0.081	**0.001586**

[Table/Fig-5]: Effect of herbal rose perfume in psychophysiological tests.
Paired t-test was done and p-value<0.05 was considered significant
TMT: Trail making test
RFFT: Ruff figural fluency test
SD: Standard deviation; Bold p-value: statistically significant

The statistical analysis of the baseline values, of the psychophysiological tests, which were done without any olfactory stimulation, and the values with olfactory stimulation using synthetic rose perfume, shows that, there is significant improvement in the performance of the TMT-A (p-value=0.00326) and TMT-B (p-value=0.0001693), with decreased time taken to perform the tests with synthetic rose olfactory stimulation. The derived index TMT (B-A), which removes the speed component and is a purer index of executive functions, showed significant results (p-value=0.002092) in this setting.

There are also significant results seen in the RFFT indices, number of unique designs (p-value=0.04026), Perseverative errors (nearly significant)(p-value=0.05625)andratioscore(p-value=0.000005856). The rest of the other parameters and derivatives have not shown any significant results with using synthetic rose olfactory stimulation [Table/Fig-6]. The RSVP test has not shown any variation in different phases. Each participant was able to find more than 50% of the target images during the baseline test, as well as, during the phases of olfactory stimulation. Therefore, this test did not yield any results to the present study.

Test	Mean±SD (baseline)	Mean±SD (synthetic)	p-value
TMT-A	24.00375±6.016	16.4705±3.711	0.00326
TMT-B	45.6495±13.119	34.0755±7.024	0.0001693
TMTB-A	21.64575±11.118	17.605±6.703	0.002092
TMTB/A	1.95807308279357±0.541	2.133863962±0.548	0.9339
TMT (B-A)/A	0.95807308279357±0.541	1.133863962±0.548	0.9339
RFFT total number of unique designs	26.85±7.353	33.7±5.268	0.04026
RFFT perseverative errors	1.9±2.550	1.7±1.870	0.05625
RFFT ratio score	0.0828868323445713±0.137	0.054480997±0.064	0.000005856

[Table/Fig-6]: Effect of synthetic rose perfume in psychophysiological tests.
Paired t-test was done and p-value<0.05 was considered significant.
TMT: Trail making test;
RFFT: Ruff figural fluency test; SD: Standard deviation

Comparison of the scores between herbal and synthetic rose olfactory stimulation, shows that, the herbal and synthetic rose perfumes have different effects on cognitive and executive functions. The herbal rose perfume has nearly significant influence on attention and speed of processing evident by TMT-A scores [Table/Fig-7] and significant effect on cognitive fluency evident by RFFT ratio scores [Table/Fig-8], whereas, the synthetic rose perfume positively influences the attention, speed, switching between tasks, cognitive fluency, and flexibility [Table/Fig-7,8] and this effect is better with synthetic rose perfume, when compared with herbal rose perfume.

TMT score	Parameters		
TMT-A	**Baseline**	**Herbal**	**Synthetic**
Mean	24.00375	19.69	16.4705
SD	6.016	4.401	3.711
p-value		0.05433	0.00326
TMT-B	**Baseline**	**Herbal**	**Synthetic**
Mean	45.6495	39.404	34.0755
SD	13.119	11.247	7.024
p-value		0.3403	0.0001693
TMT-(B-A)	**Baseline**	**Herbal**	**Synthetic**
Mean	21.64575	19.714	17.605
SD	11.118	10.219	6.703
p-value		0.6009	0.002092

[Table/Fig-7]: Comparison of TMT scores between herbal and synthetic rose olfactory stimulation.
Paired t-test was done and p-value<0.05 was considered significant.
TMT: Trail making test; SD: Standard deviation

RFFT score	Parameters		
RFFT total number of unique designs	**Baseline**	**Herbal**	**Synthetic**
Mean	26.85	31.775	33.7
SD	7.353	6.370	5.268
p-value		0.3741	0.04026
RFFT perseverative errors	**Baseline**	**Herbal**	**Synthetic**
Mean	1.9	2.1	1.7
SD	2.550	2.121	1.870
p-value		0.2555	0.05625
RFFT ratio score	**Baseline**	**Herbal**	**Synthetic**
Mean	0.082886832	0.069719	0.054481
SD	2.550	0.081	1.870
p-value		0.001586	0.000005856

[Table/Fig-8]: Comparison of RFFT scores between herbal and synthetic rose olfactory stimulation.
Paired t-test was done and p-value<0.05 was considered significant.
RFFT: Ruff figural fluency test; SD: Standard deviation

DISCUSSION

The sense of smell effects mood, stress and working capacity. Electrophysiological studies using EEG have shown that exposure to fragrances affect cognitive functions and spontaneous brain activity [3]. The present study compare the effects of olfactory stimulation with herbal and synthetic perfumes, and found that visual search, scanning and speed of processing improved with both herbal and synthetic rose olfactory stimulation. The improvement with synthetic rose was more than with herbal rose, along with improvement in mental flexibility, and executive functions. This is consistent with a study conducted by by Sowndhararajan K et al., which showed that, isomers produce differences in brain function due to differences in the area of brain affected [18].

Therefore, it is proposed that, compounds with different chemical structure but similar smell will have different effects on mood, behaviour, attention, and cognitive functions. The creation of a certain odour or aroma is accomplished by the activation of multiple receptors that lead to the formation of specific activity patterns in the olfactory bulb in which the tuning events occur [19]. The discriminatory capacity of the mammalian olfactory system is such that, thousands of volatile chemicals are perceived as having distinct odours. Malnic B et al., used a combination of calcium imaging and single cell RT-PCR to identify odorant receptors for odorants with related structures, but varied odours and found that, multiple Olfactory Receptos (ORs) recognise one odorant, and different combinations of ORs recognise different odorants, implying that, the olfactory system uses a combinatorial receptor coding scheme to encode odour identities. Their studies also indicate that slight alterations in an odorant, or a change in its concentration, can change its "code," potentially explaining how such changes can alter perceived odour quality [20].

There was a positive effect of synthetic rose on executive control processes, determined by TMT (B-A) which is a purer indicator of executive control, as it removes the speed component. The herbal rose showed significant results in the error ratio of RFFT which indicates correctness of performance whereas, the synthetic rose has shown to positively affect non verbal capacity for initiation, planning, and divergent reasoning and correctness of performance. Diego MA et al., studied the essential oils of lavender and rosemary and compared the mood arithmetic computation, and EEG. Pre vs post "Aromatherapy" comparison of mood, math computation and EEG, showed, both lavender and rosemary odours increased speed in math computations, increased in relaxation, and decreased in anxiety. However, EEG changes were consistent with the idea that, lavender is sedating and rosemary is alerting [21]. Moss M et al., compared objective cognition measures and subjective mood using the essential oils of lavender and rosemary, and found that, lavender caused decrease in working memory and impaired reaction time of memory and attention tasks compared to control (no odour). Rosemary enhanced performance for memory and secondary memory factors, but also showed impaired reaction

time compared to control. After testing, the rosemary group was more alert than the lavender, as well as, the control group and both aroma groups were more content than the control group [22]. A study conducted by Warm JS et al., showed that exposure to fragrances helped to increase stimulation during prolonged tasks which require close attention [23].

The effects of herbal and synthetic rose olfactory stimulation have shown positive influence on the attention and cognitive functions and this finding can be applied to create student friendly learning environments by providing pleasant stimuli, through means of aromatic sprays delivering the odorant in a pulsatile manner at a frequency to overcome the ensuing olfactory adaptation. Moreover, anxiety associated with examinations can be effectively reduced using aromatherapy [24].

Limitation(s)

The COVID-19 situation caused hindrance to get more subjects enrolled for the study. The results of the psychophysiological tests can provide more reliable results if, the sample size is considerably increased. The reliability of the psychophysiological tests can be increased if, the duration between each phase of the test is increased to atleast three weeks to 12 months.

CONCLUSION(S)

To summarise the wide range of olfactory receptors present in the human body and their ability to respond to a huge spectrum of different olfactory substances, through a variety of permutations and combinations between the olfactory substance and the olfactory receptors enlightens about the possibilities of effects of different compounds. With a change in the chemical structure of the chemical compound the effects of it in different physiological systems varies. The herbal and synthetic rose perfumes also, have different effects on the physiological systems, especially of different cognitive and executive functions. Comparison of the scores between herbal and synthetic rose olfactory stimulation, shows that, the herbal and synthetic rose perfumes have different effects on cognitive and executive functions. The herbal rose perfume has nearly significant influence on attention and speed of processing and significant effect on cognitive fluency, whereas, the synthetic rose perfume positively influences the attention, speed, switching between tasks, cognitive fluency, and flexibility, and this effect is better with synthetic rose perfume, when compared with herbal rose perfume. These findings should be further correlated with imaging techniques to objectively identify the specific areas of the orbitofrontal and piriform cortex and their degree of activation with different compounds. The effects of herbal and synthetic rose olfactory stimulation have shown positive influence on the attention and cognitive functions and this finding can be applied to create student friendly learning environments by providing pleasant stimuli, through means of aromatic sprays delivering the odorant in a pulsatile manner at a frequency to overcome the ensuing olfactory adaptation.

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PLAGIARISM CHECKING METHODS: [Jain H et al.]
- Plagiarism X-checker: Dec 17, 2022
- Manual Googling: Mar 22, 2023
- iThenticate Software: Apr 17, 2023 (10%)

ETYMOLOGY: Author Origin

EMENDATIONS: 6

AUTHOR DECLARATION:
- Financial or Other Competing Interests: None
- Was Ethics Committee Approval obtained for this study? Yes
- Was informed consent obtained from the subjects involved in the study? Yes
- For any images presented appropriate consent has been obtained from the subjects. NA

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