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# The Potency of Antioxidant Perfume of Essential Oils to Reduce Free Radical Content in Air

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**Abstract.** Free radical contamination is very dangerous for health. Perfume contains essential oils whose components are potential as antiradical. Antioxidant perfume is a perfume made from essential oil with a certain concentration which is very effective to reduce free radicals in the air. This study aims to determine the effectiveness of essential oils as antiradical compounds using laboratory experiments in the reduction of free radical concentration of DPPH with various concentrations of fruit extract which containing terpenoid component. DPPH concentration changes in the air followed by using electronic nose (E-Nose) equipped with multisensory gas. The experimental results showed that the critical concentration of fruit extract which effectively decreased free radical i.e. apple, orange, grape, melon, and lemon were 7.47%, 6.21%, 15.61%, 7.58%, and 6.22%. The greater concentration of these critical concentrations of fruit extracts is potentially as prooxidants.

## 1. Introduction

Free radicals are potentially damaging to the environment and human health. Because when free radicals enter the body, it will rapidly attract biological macromolecules electrons that surround proteins, nucleic acids, and deoxyribonucleic acids (DNA) which then interfere metabolic processes in the body [1]. It is now known that various types of volatile non-methane organic compounds from different vegetation have been emitted into the atmosphere every day. These organic compounds include isoprene, monoterpene $C_{10}H_{16}$, sesquiterpene $C_{15}H_{24}$, as well as the number of oxygenated compounds such as methanol, hexene, 2-methyl-3-buten-2-ol, and 6-methyl-5-hepten-2. In the troposphere layer, the organic compound with free radical is hydroxyl (OH), nitrate ($NO_3$) and ozone ($O_3$) which the reaction reacts to the lower layer of the troposphere [2]. Different half-life times, for example linalool a monoterpene often found in perfume products, has a half-life of 52 minutes with OH radicals, 55 minutes with ozone and 6 minutes with $NO_x$. Limonene has a citrus scent having a half-life of 49 minutes (OH), 2.0 hours ($O_3$) and 5 minutes ($NO_x$) [3].

Although terpenoids can react with these radicals to reduce the reactive effects of free radicals, but some researchers have found that their reaction products actually produce aerosol particles in the troposphere layer [4]. The termination reactions of these radicals produce stable products even in aerosol form. The reaction products include formaldehyde, methyl vinyl ketone, methacroline, organonitrate and others [5]. The products of perfume, air freshener or cosmetics usually contain terpenoid compounds as their aroma components. That causes when evaporated or sprayed into the air, the terpenoid

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components will help to reduce the presence of free radicals in the atmosphere with different half-lives depending on the type of terpenoid component [6].

Antioxidant perfume is a perfume made from essential oil with a certain concentration which is very effective to reduce the free radical in the air because there are antiradical compounds in essential oils. There are some natural ingredients that are potential as antioxidants and can be formulated as perfumes, including essential oils [7]. Some essential oils have antioxidant activity, such as phenol group compounds, such as eugenol. Eugenol can lower free radicals through the abstraction of hydrogen atoms [8], while terpenoids through a chain-based reaction mechanism carrying HOO radicals, which react rapidly with terpenoid radicals to halt chain reactions [9]. Measurement of antioxidants in the air requires instruments such as electronic nose (E-nose). E-nose is an instrument designed to mimic the mammalian olfactory system [10]. In this study, there will be held an experiment decrease free radical activity that comes from compound 2,2-diphenyl-1-picrylhydrazyl (DPPH) in the presence of perfume material derived from various extracts of aromatic fruits. The changes in free radical activity in the air are monitored using electronic nose (E-Nose).

## 2. Research Methods

### *2.1. Chemicals and Instrumentation*

Essential oils were obtained from extraction of fruit by maceration using propylene glycol solvent. DPPH or 1,1-diphenyl-2-picrihidazyl was used as free radical source.

The extract was characterized using gas chromatography - mass spectrometer (Shimadzu QP-2010S equipped with RTX-5MS capillary column (30 m x 0.25 mm)). The gas carrier was set up at 50ml/min, from 60-300ºC with 10º/min speed, injector temperature of 250 ºC, interface temperature of 250 ºC, and the spectrum library from Wiley 9. Electronic nose or e-nose instrumentation was prepared and assembled as shown in Figure 1. Inside the sensor room, there are 10 sensors (arrays) of various types and sensitivities as shown in Table 1. These sensors consist of semiconducting $SnO_2$ metal sensitive to particles in the air. If the air is clean, the sensor shows a low conductivity. It is expected that these sensors give respond to the component in the fruit extract. The sensor conductivity increases as the particle concentration in the air increases.

**Table 1**. Sensor Sensitivity

| Type of Sensor | Sensitive to gas |
|---|---|
| MQ-2 | LPG, iso-butane, propane, methane, alcohol, $H_2$, and cigarette smoke |
| MQ-3 | Benzene |
| MQ-4 | Methane and benzene |
| MQ-5 | LPG, natural gas and urban gas |
| MQ-6 | LPG, iso-butane, propane |
| MQ-7 | CO |
| MQ-8 | $H_2$ |
| MQ-131 | $CO_2$ |
| MQ-135 | Acid cigarettes, $CO_2$, $NH_3$, NOx and benzene |

### ***2.2.*** *Reaction Procedure*

The fruit samples used were oranges, grapes, lemons, melons, apples and each of them were destroyed and weighed until 100 g, and then soaked in propylene glycol by ratio of (1: 3) for 3 x 24 hours. The extract product was concentrated using a rotary evaporator vacuum. Next, a solution of fruit extract as a source of aroma with concentration of 5, 10, 15, 20, and 25% (v/v) in propylene glycol solvent was prepared. DPPH was dissolved in propylene glycol to obtain a 0.2 mMol liquid DPPH and stored in dark coloured bottles, before further use.

In the e-nose instrumentation, two hose were installed for the vapor of DPPH solution and a solution of fruit perfume extract. The first stage of the vapour was the DPPH flowing into the sensor room for data collection of signals and then blowing out, each of extraction took 30 seconds for a total 300 seconds. Then, the extract steam for each concentration and type of fruit extract was also performed in the same way. For each extract replacement, the instrument was flushed using propylene glycol vapor for 2 minutes. Observation of the effect of fruit extract on the change of DPPH free radical concentration was done by blowing the solution of DPPH and fruit extract simultaneously with the same treatment as described above for 300 seconds. Gas sensor response data was collected in the form file.xls which further processed to obtain the relationship of concentration with the magnitude of each sensor response.

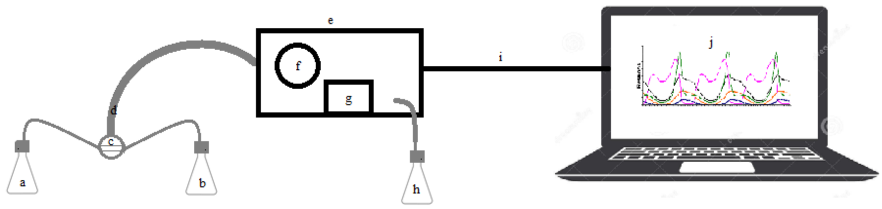



**Figure 1**. Trial series using e-nose. Description: (a) The sample bottle contains the DPPH, (b) the sample bottle containing the volatile, (c) valves, (d) the steam connecting hose, (e) E-nose instrumentation; (f) the sensor room; (g). power supply, (h) vapour trap bottle containing propylene glycol, (i) electrical cable, (j) computer.

## 3. Result and Discussion

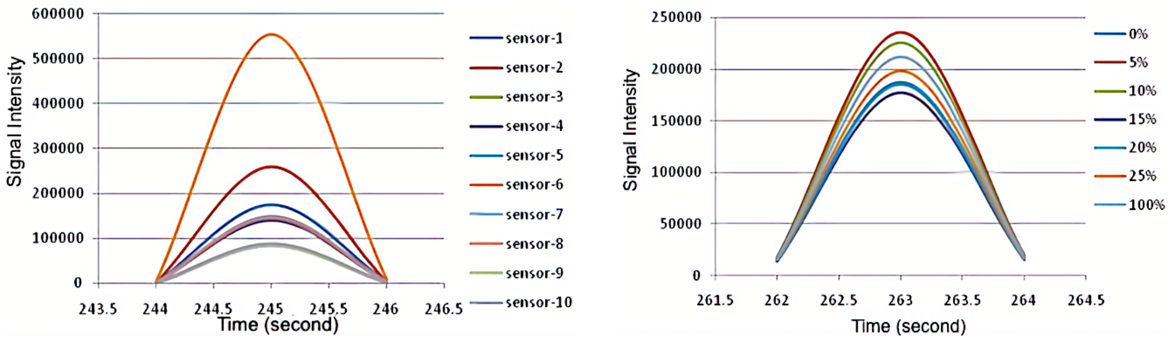
### *3.1. GC-MS Result*
Characterization of fruit extract as a source of perfume using gas chromatography-mass spectrometer provides information on the types of compounds contained in the extract, as shown in Table 2.

**Table 2.** Components in fruit extract GC-MS analysis result

| Fruit Extract | Major Components | Minor Components |
|---|---|---|
| Lime *(Citrus hystrix*) | linalool, citronellal; ß-phelandrene, isopulegol, ß-citronelol | sabinene; thujene; terpinen-4-ol; cephrol |
| Melon (*Cucumis melo*) | myrsenol; lomine; jasminaldehyde; amilcinnamaldehyde; amil cinnamic aldehyde; alpha hexyl cinnamic aldehyde | caproil alcohol; penticarbinol; gardeniol; acetic styralyl; aceticneryl; 2-decalactone |
| Lemon (*Citrus limon*) | d1-Limonene; cinene; nesol; 2-methyl-4-isoprophenilcyclohexa-1-ene | |
| Grapes (*Vitis vinifera L)* | d1-Limonene; sinene; nesol; 4-methyl-3,6,9-trioxadecan-1-ol | Ethyl butanoic; ethyl butyrate acid; linalool l; benzylcarbinol; geraniol; aceticneryl; acetic geranyl; methyl dihydrojasmonate; cis-2-hexenol, |
| Apeple (*Malus sylvestris)* | heptanoic allyl, citronellal | tert-Butyl acetoacetate, gamma-undecanalactone, trans-alfa ionone, Benzyldimethylcarbinol acetate, 2-tertiobutylcyclohexyl acetate, o-tert-Butylcyclohexanol |

### *3.2. E-nose Responses*
The molecules in the fruit extract do not contain small size compounds that can be addressed by all types of sensors in Table 1. However, Figure 2 shows that the presence of molecular parts similar to the molecule responded, causing the sensors to respond to the vapor extract of fruit and DPPH vapor with different sensitivity.

The e-nose detector response curve to the DPPH vapor of both single vapor and steam mixture with fruit extract shows significant signal intensity change. E-nose response to the highest DPPH radical vapor on sensor 6, then sensor 2 and sensor 1. The response of air sensor containing a mixture of DPPH radical vapours and fruit extracts showed a decrease in the response intensity to a minimum concentration of fruit extracts which tended to increase again (Figure 3). This proves that in the sensor space that is analogous to the air space, there has been an interaction between DPPH molecules and scented molecules of fruit extract. This interaction results in a decrease of sensor signals conductivity.




(a) (b)

**Figure 2.** E-nose responses to (a) steam of DPPH solution and (b) steam of 20% lime extract.

### 3.3. *Critical Concentration of Fruit Extract*

The free radical intensity of DPPH can be optimally optimized by the effect of a particular concentration of fruit extract [11]. E-nose instrumentation will detect using appropriate sensor to produce an average graph which is the representative image of the free radical response of DPPH + fruit extract. The valley peak is a critical point indicating the optimum concentration of fruit extracts that can be used as antioxidant free radical antidote. As depicted in Figure 3 and summarised in Table 3, if the concentration exceeds at the critical point, then the fruit extract will increase and strengthen the intensity of DPPH free radical which means to be pro-radical [12].

**Table 3.** Sensor sensitivity of DPPH vapour and fruit extracts

| No. | Extracts | Sensors | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
| 1 | Apple | - | - | v | v | - | - | - | v | v | - |
| 2 | Lime | - | - | - | v | v | - | v | v | v | v |
| 3 | Grape | - | - | v | v | - | - | - | - | - | - |
| 4 | Melon | - | - | v | v | v | v | v | v | v | v |
| 5 | Lemon | v | v | - | - | - | - | - | - | - | - |

The sensitivity of fruit extract and DPPH free radicals to the sensors in e-noses varies depending on the type of fruit extract. Regression analysis of the sensor response curve to the DPPH mixed vapor and fruit extract showed the quadratic equation where $R^2 \approx 1$. By solving the quadratic equation, each curve obtained the optimum average concentration of each fruit extract, referred as the critical concentration, as shown in Table 4. The decrease in intensity of the sensor response is predicted due to a decrease in sensor conductivity caused by its interaction with DPPH radicals. The DPPH vapor response numbers decrease as the concentration of extract increases, reaching a critical state after the critical value of the curve tends to increase again. This phenomenon showed that there has been an interaction between the DPPH radical and the chemical components of the fruit extract vapor inside the electronic nose. The reduction of the response can be identified as the curve of antioxidant activity of fruit extract, while the rebound curve can be said as an increasing of the radical concentration or can be called pro-oxidant [12].

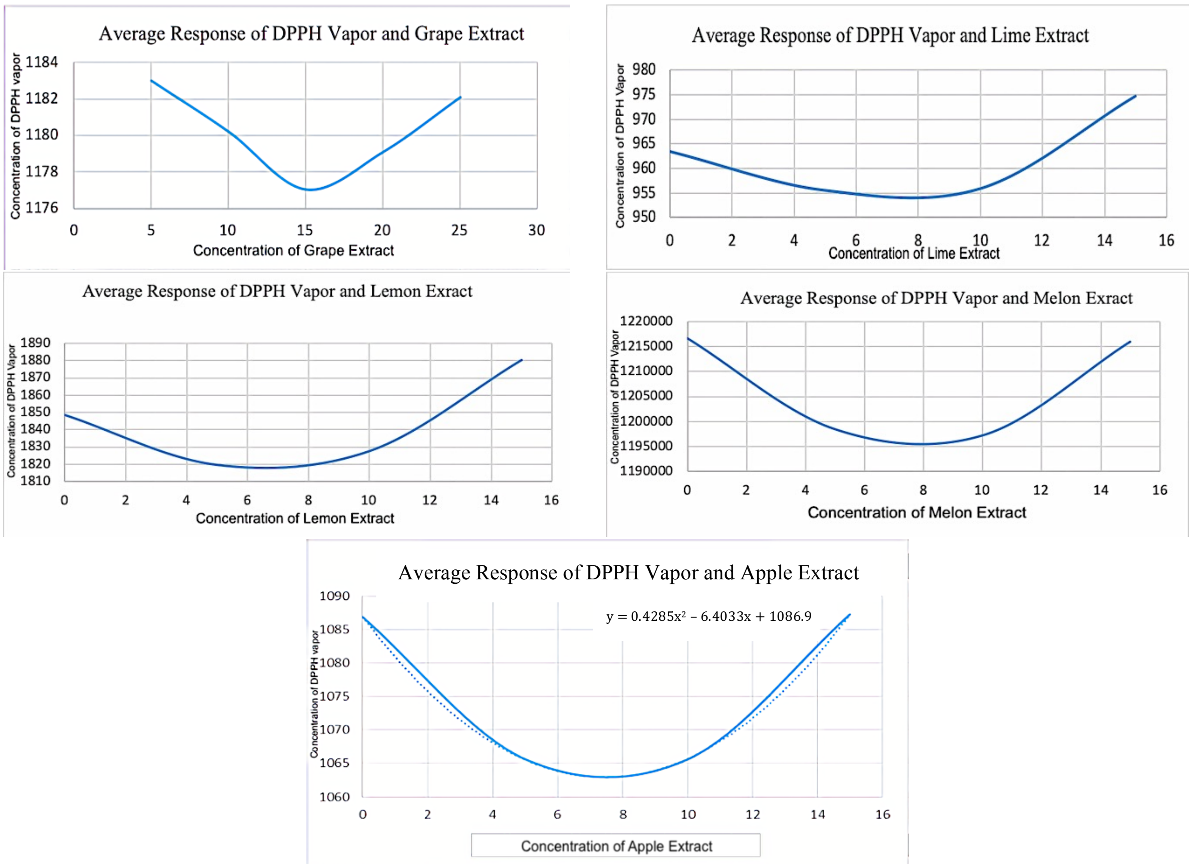



**Figure 3.** E-nose response to DPPH vapor in the presence of fruit extracts.

**Table 4**. The optimum concentration of each fruit extract in reducing DPPH free radical level

| No. | Extracts | Critical Concentration (%) |
|---|---|---|
| 1 | Apple | 7.47 ± 0.55 |
| 2 | Lime | 6.22 ± 0.07 |
| 3 | Grape | 15.61 ± 0.07 |
| 4 | Melon | 7.59 ± 0.04 |
| 5 | Lemon | 6.23 ± 0.81 |

It takes a relatively small concentration of fruit extract to be able optimally become antioxidant antidote to free radicals. Referring to Table 2 and Table 4, the main components of lime that have the greatest ability to reduce DPPH levels require 6.218% as the optimum concentration in antioxidant perfume formulations. This suggests that the main component of the orange linalool, which is a monoterpene derivative, can reduce radicals better than the minor components in other fruit extracts. The optimum concentration of limonene which is the main compound of lemon in antioxidant perfumes shows unrelated figures of 6.232%. Limonene is a monoterpene compound that has capability of reducing radical by conjugation system [13]. Components of apples serving as DPPH reducers are allyl hexanoic and citronellal. Melons have a major component of mirsenol, which is a terpenoid compound. This compound has a double bond conjugated, so it is strongly suspected to act as an antioxidant in melon extract. Although the wine-scented component is derived from a group of ester compounds, but the presence of a major component of limonene, and geraniol allows the grape extract to be antioxidant. Although it has the same main components, the wine has many minor components while the lemon does not have a minor compound. This suggests that in lemon there is more limonene than wine, so in compare with lemon, the more optimum concentration of wine is needed to reduce DPPH.

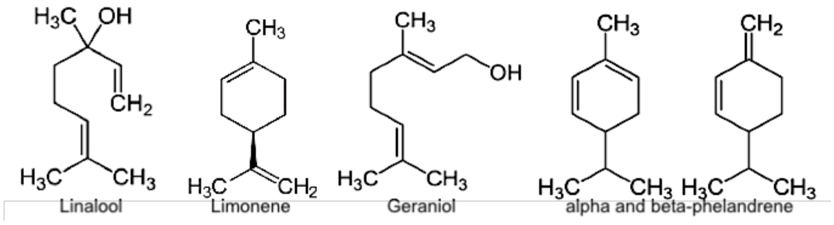



**Figure 4.** Terpenoid components in fruit extract that can be able to act as an antioxidant [9].

## 4. Conclusion

Experimental results of extra fruit effect on DPPH free radical decline showed that all fruits were able to decrease free radical of DPPH in the following order of abilities: lime> lemon> apple> melon> grape. Linalool in oranges is a monoterpene derivative that most potentially reduces free radicals and is followed by limonene in lemons. Diterpenoid compounds that are widely found in fruits have different ability to reduce radical. This confirms that the optimum concentration is important to know because the concentration of these compounds will become pro-radical. Fruit extracts that contain essential components potentially as perfume materials that can reduce free radicals in the air. Furthermore, E-nose can be used for tracing and simultaneously determining the activity of free radicals in the air in the gas phase.

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