Parameter study, antioxidant activities, morphological and functional characteristics in microwave extraction of medicinal oleoresins from black and white pepper

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ABSTRACT
The study investigated the effects of extraction parameters on the yield, antioxidant activities, morphological and functional group characteristics of black and white pepper oleoresin extracts. Optimized oleoresin yields of 5.64 and 8.72 w/w% were obtained as black and white pepper extracts, respectively. Moreover, from the antioxidant assay, the concentration of black and white pepper extracts required to scavenge half of stable DPPH radicals were 94.92 and 107.57 μg/ml, respectively. From the complementary antioxidant assay; the concentration of black and white pepper extracts required to scavenge half of the stable ABTS free radicals were 82.36 and 94.71 μg/ml, respectively. This indicated that extracts from white pepper exhibited higher antioxidant capacity than the black pepper extracts. The results from the untargeted compositional GC-MS analysis identified a total of 23 and 31 bioactive compounds in black and white oleoresin extracts, respectively. This study, therefore, revealed the potential of microwave extraction in obtaining high-quality extracts.

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1. Introduction
The major cause of many terminal diseases such as cancer, diabetes, the cardiovascular and neurological disorder is attributed to the formation of free radicals [1,2]. Siqueira et al. [3] listed factors such as environmental pollution, stress, radiation and exposure to exogenous industrial chemicals as the main source of free radical-related diseases. The free radicals can, however, be neutralized through the action of a substance known as antioxidants [4,5]. Antioxidants are synthetic or natural products that inhibit the damage of metabolic cells. Although the synthetic form of antioxidants such as butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT) have shown great potentials in neutralizing free radical effects on the human body; they have however shown some carcinogenic and toxic effects [6]. Natural antioxidants are found in functional foods such as fruits, vegetables and seeds with immense nutritional and therapeutical properties [7]. The natural form of antioxidant is, therefore, a safe alternative in the prevention and treatment of free radical-related diseases when compared with the synthetic ones.

Black and white peppers are important functional commodity crops of the Piperaceae family with economical, nutritional and health benefits. It has application in the food industry as spices, traditional medicine as anti-rheumatic, chemical industries as insecticides and cosmetics [8,9]. The difference between the two species is in their processing time which is expected to have a great effect on their respective bioactive composition. Black pepper is produced from immature green berries to form dark brownish reticulated peppercorns [10]. However, white peppers are processed from a fully matures flowering green berries and allowed to mollified to form dried creamy coloured seeds [6].

Several conventional methods had been used in the extraction of bioactive oleoresins from black and white peppers. These include cold percolation, maceration, soxhlet extraction, hydrotrropic solubilization, and hydro-distillation [11,12]. However, there are several demerits abound when using these conventional techniques, which include cold percolation, maceration, soxhlet extraction, hydrotrropic solubilization, and hydro-distillation [11,12]. However, there are several demerits abound when using these conventional techniques, which include high solvent consumption, low extraction yield, solvent contamination, higher extraction cost and longer time [13]. These disadvantages necessitated a research into finding a better method with lower solvent consumption, environmentally friendly, higher yield, shorter irradiation time, higher selectivity, and higher extract quality [14]. The microwave reflux technology provided the new approach to the extraction of medicinal oleoresin from natural products. The ionic conduction and dipole interaction are the two basic phenomena that best explain the mechanism of microwave extraction. These involve an electromagnetic interaction between the electric...
and magnetic field [15]. The ionic conduction offers a stiff resistance to the movement of ions which in turn triggers heating effect and thereby resulting in the exudation of bioactive components from the pepper matrix. The need then arises to find operating conditions for optimum extraction of bioactive oleoresin. This research, therefore, focused on the optimization of microwave extraction process, identification of bioactive compounds and structural elucidation of the extracted oleoresins.

2. Material and method

2.1. Materials

A standard grade black and white pepper samples were purchased from the Malaysian Pepper Board (MPB) located in Sarawak. An analytical-grade ethanol (99.9% purity), acetone (97% purity), and distilled water were obtained from the Chemical Laboratory, Universiti Malaysia Pahang, Kuantan. DPPH (2,2′-diphenyl-1-picrylhydrazyl) and ABTS (2,2′-azinobis (2-ethyl benzothiazoline-6-sulfonate) reagents were procured from Sigma Aldrich Chemical Co. The two pepper samples were pulverized into a finely defined powder using Eppendorf 200- model grinder. The powdered samples were clarified into five different sizes of 0.105 mm, 0.154 mm, 0.30 mm, 0.45 mm and 0.90 mm. The samples were stored in an air-tight container.

2.2. Microwave reflux extraction

25 g of the clarified pepper powder was mixed with an appropriate volume of distilled water, inside a round bottom flask. This was placed inside the microwave cavity and refluxation performed in accordance with the designed parameters. The extracts were unloaded, filtered and concentrated using a rotary evaporator (BUCHI, R-200 model, Germany). The extraction yield was calculated on a dry weight basis of the fresh pepper powder and stored at 4°C in a dark vial prior for further component and physicochemical analyses. The percentage yield of extracts was determined based on the dry weight (d.w.) using the Eq. (1).

\[
\text{Extraction yield} = \frac{\text{Weight of oleoresin extracts}}{\text{Weight of dried sample}} \times 100\%
\]

2.3. Optimization of microwave reflux extraction

The optimal extraction conditions was determined using a robust L9—orthogonal design matrix called Taguchi methodology [14]. This was designed and analyzed using Minitab 17™ software to determine a combination of extraction parameters that jointly optimize the extraction yield. This is a multi-step optimization approach which provides a tolerance level in minimizing response deviation. In the process of searching for an optimal condition a “higher-the-better” transfer function was used as illu., stated in Eq. (2).

\[
\text{SNR} = -10 \log_{10} \left( \sum \frac{1^2}{y} / n \right)
\]

where, \(y\) is the response value, and \(n\) is the number of observation.

2.4. GC-MS analysis

Gas chromatography-mass spectrometry (Agilent 5973-model) was used to determine the chemical composition of the oleoresin extracts. C-18 capillary column with 30 m length, 0.25 mm diameter and 0.25 μm film thickness was used. The spectrometry analysis was carried out by diluting a micro-filtered extracts (1 μL) with an analytical standard grade acetone extract:10. This was injected into the GC-MS for components identification. The oven temperature was set from 50°C (kept constant for 5 min) at a rate of 3°C per minutes, next to 125°C at a rate of 3°C per minute (kept constant for 5 min) and finally at a rate of 3°C per minute to 290°C (kept constant for 5 min) with a total run time of 69 min. Helium was used as carrier gas with 0.7 mL/min flow rate; 1:50 split ratio, with an ionization mode of 70 eV. The bioactive components were identified in relation to the peak area fragmentation fingerprints matched with the NIST05a.Library database.

2.5. Scanning electron microscopy (SEM)

The morphological characteristics of the raw sample before extraction and the residue obtained after extraction were examined using the scanning electronic microscope (Phillips XL30, Holland). The SEM-images of fresh pepper samples and the one obtained from optimal extraction condition were properly evaluated to the he determine the structural transformation that has taken place. The test samples were mounted on an adhesive plate, air-dried and coated with thin layer gold before analyzing. The coating of the sputter was carried out to get rid of any electrical discharge and this was then analyzed using a voltage and magnification of 15 kV and (40–300)× respectively.

2.6. Fourier transforms infrared spectroscopy (FTIR)

IR-Spectra analysis was carried out on oleoresin extracts to determine the functional groups present and any chemical transformation that has taken place in the bonding structure. This was conducted using the FTIR spectrometer (Thermo-Nicolet iS5 iD7 ATR, Germany) equipped with an OMNIC software. The KBr standard
procedure was employed with a scanning wave number ranging between 4000 and 500 cm\(^{-1}\).

2.7. DPPH free radical scavenging

The choice of DPPH reagent is due to its high sensitivity in detecting a small variation in the antioxidant activities with the minimal use of test samples. DPPH is a stable free radical that can accept an electron or hydrogen radical to become a stable diamagnetic molecule. The method by Zhang and Xu [16] for DPPH free radical scavenging was used for this study. The negative control \((A_0)\) was prepared by mixing 0.5 ml of ethanol with 2.5 ml of the DPPH solution and absorbance was measured after 30 min of incubation at room temperature. Five different extracts concentrations of 50, 100, 150, 200 and 250 μg/ml were prepared. Absorbance \((A_1)\) was measured for a mixture of different concentration (0.5 ml) of extract consecutively taken and mixed with 2.5 ml of DPPH. However, in order to eliminate colour effect from the extract, an absorbance denoted by \(A_2\) was taken which comprises of oleoresin extracts (different concentration) and 2.5 ml ethanol. The percentage inhibition was calculated using Eq.(3).

\[
\text{Inhibition} \% = \left(1 - \frac{A_1 - A_2}{A_0}\right) \times 100 \quad (3)
\]

The percentage inhibition was plotted against the five concentrations (50–250 μg/ml) of the extract to obtain the inhibition curves. A straight-line equation was generated from the linear regression analysis. The IC\(_{50}\) value was thereafter estimated to determine the concentration of extract required to scavenge half (i.e. 50%) of the DPPH radicals.

2.8. Complementary assay: ABTS free radical scavenging

The ABTS method is based on spectrophotometric monitoring of the decay of the radical-cation \(\text{ABTS}^{\cdot+}\) produced by the oxidation of 2,2’-azinobis(3-ethylbenzothiazoline-6-sulfonate) (ABTS) caused by the addition of PH [17]. This assay was used to estimate the degree of inhibition of oleoresin extracts to the ABTS free radicals. The ABTS free radicals were synthesized by dissolving potassium peroxydisulfate in deionized water to make up a 7.2 mM stock solution. A 2.47 mM of the potassium peroxydisulfate was mixed with the stock solution to make up a final solution. This was allowed to stand for 16 h at room temperature before using 3.8 ml containing a mixture of ABTS and ethanol was added to a 100 μl of the spice extracts \((A_1)\). Absorbance was taken at 732 nm after 6 min of incubation with ethanol as the blank solution \((A_0)\) in each assay. Percentage inhibition was calculated using the Eq. (4) below.

\[
\text{Scavenging Activity(ABTS}^{\cdot+}) = \left(\frac{A_0 - A_1}{A_0}\right) \times 100\%
\]

The effective IC\(_{50}\) value estimated from the curve of inhibition per cent against extracts concentration. This is the concentration required to scavenge half of the ABTS free radicals.

3. Results and discussion

3.1. Parameter study

The effect of extraction time variation on the extraction yield was determined by keeping the microwave power (200 W), particle size (0.105 mm), and solvent volume (60 ml) as shown in Figure 1. The results obtained from time variation (30, 60, 90, 120 and 150 min) revealed an initial increase in the oleoresin yield as time increases steadily from 30 min to 90 min. Highest yield (5.80 w/w%) was obtained from white pepper as compared with 4.94 w/w% from black pepper which indicated a higher oleoresin yield from white pepper at the same extraction conditions. The extractable oleoresin dropped after 90 min of extraction and this suggests that a further increment in irradiation time will have no significant contribution to the extraction yield [18]. At the degradation region the energy absorption capacity of the pepper sample plummeted and hence the extraction yield at this point is relatively low.

Moreover, the effect of microwave power variation on the extraction yield was investigated at constant particle size (0.105 mm), and solvent volume (60 ml) using maximum extraction time (90 min). The microwave power variation was varied as 200, 250, 300, 350 and 400 W (Figure 2). The oleoresin yield increased as the power was varied from 200 to 300 W. This indicated an initial induction effect which later resulted in the migration of ions resulting from the dipole rotation in the electromagnetic field as described by Olalere et al. [11]. The optimal extraction yield was attained

Figure 1. Effects of irradiation time on oleoresin yield.
300 W beyond which volatile most of the bioactive compounds would have degraded. This is a marked melioration and improvement over the study carried out by Desai & Parikh [19] who arrived at maximum yield at 1700 W and 35 min of extraction time. With such high power level, most of the thermo-labile constituents of the pepper matrix would have denatured. Therefore, there is the need for a trade-off between the microwave power and the radiation time in order to preserve some of the heat-sensitive compounds.

Figure 3 illustrated effects of particle size on oleoresin yield at constant solvent volume (60 ml), extraction time (90 min), and microwave power level (300 W) from the previous variation were used as constant parameters. The results from this variation placed the highest oleoresin yield at a particle size of 0.105 mm. This is an indication of higher solvent penetration due to the larger surface area [20]. However, a granular pepper matrix produces lower energy absorption due to low dielectric heating [20]. Finely divided powder size has higher energy absorption capacity than coarse particles. Hence, a higher microwave energy is required for a more coarse particle as reported by Jouyban et al. [21].

Figure 4 illustrated the effect of solvent volume on the oleoresin yield at constant extraction time (90 min), and microwave power level (300 W). The optimal extraction parameters from previous variations were kept constant to investigate the effect of solvent volume on the extractable oleoresins from black and white pepper. The results from volume variation (60, 80, 100, 120, and 140 ml) gave maximum extraction yield with 120 ml solvent volume. Beyond this point, the oleoresin yield reduced which inadvertently resulted in the reduction of power dissipation per volume. This suggested that with larger solvent volume the energy requirement for vaporization increases and thus a lesser sensible heat for actual extraction reduces correspondingly [22].

3.2. Determination of factor range

From the results of parameter variation, a three-point operating levels were selected from each extraction variables to design a robust orthogonal matrix called Taguchi array [23]. The summary of the operational levels for each extraction variables is presented in Table 1.

3.3. Determination of optimized extraction conditions

The optimum extraction condition was attained at 120 min of extraction time, 350 W of microwave power, 0.105 mm of particle size and 120 ml of solvent volume during black pepper microwave refluxation (Table 2). However, in white pepper microwave refluxation, the optimum extraction condition was attained at 120 min of extraction time, 300 W of microwave power, 0.105 mm of particle size and 120 ml of solvent volume. Under the optimum condition, the optimal yield of 5.64 and 8.72 w/w% were obtained. The signal–noise ratio (SNR) measured the deviation of mean yield from the optimum setting. The experimental run with the largest SN-ratio is, therefore, the optimal condition and this forms the basis for determining the optimum condition for a set of independent extraction parameters.
3.4. Bioactive compounds and proximate composition

A total of 23 components were extracted from black pepper, comprising 43.42% alkaloids, 13.33% monoterpenes, and 16.24% sesquiterpenes. However, in white pepper extracts, a total of 31 bioactive components comprising 55.76% alkaloids, 20.16% monoterpenes, and 21.78% sesquiterpenes were identified as presented in Table 3. The higher number of a bioactive compound identified from both extracts is a remarkable improvement over the results of the investigation made from other studies [24, 25].

3.5. Functional group analysis

The absorption spectra of different functional groups contained black and white pepper extracts are illustrated in Figure 5. The absorption band of 3271 and 3293 cm\(^{-1}\) for black and white extracts respectively

### Table 1. Extraction factors and levels.

| Control factors | Symbols | Units | Black pepper Operating levels | White pepper Operating levels |
|-----------------|---------|-------|-------------------------------|-------------------------------|
|                 |         |       | (−1) | (0) | (+1) | (−1) | (0) | (+1) |
| Extraction time | \(x_1\) | min   | 60   | 90  | 120  | 60   | 90  | 120  |
| Microwave power | \(x_2\) | W     | 300  | 350 | 400  | 250  | 300 | 350  |
| Particle size   | \(x_3\) | mm    | 0.105| 0.154| 0.300| 0.105| 0.154| 0.300|
| Solvent volume  | \(x_4\) | mL    | 80   | 100 | 120  | 80   | 100 | 120  |

### Table 2. L\(_9\) (3\(^4\)) Orthogonal experimental design and response values.

| No | \(x_1\) | \(x_2\) | \(x_3\) | \(x_4\) | Black pepper extract (w/w %)* | White pepper extract (w/w %)* | SN-ratios | SN-ratios |
|----|---------|---------|---------|---------|-------------------------------|-------------------------------|-----------|-----------|
|    |         |         |         |         | \(y_{black}\)               | \(y_{white}\)               | \((SNR)_{y_{black}}\) | \((SNR)_{y_{white}}\) |
| 1  | −1      | −1      | −1      | −1      | 1.40 ± 0.42                  | 3.38 ± 0.02                  | 2.92      | 10.58     |
| 2  | −1      | 0       | 0       | 0       | 0.69 ± 0.38                  | 2.59 ± 0.08                  | −3.22     | 8.27      |
| 3  | −1      | +1      | +1      | +1      | 1.31 ± 0.15                  | 2.42 ± 0.07                  | 2.33      | 7.68      |
| 4  | 0       | −1      | 0       | +1      | 0.36 ± 0.11                  | 3.96 ± 0.08                  | −8.87     | 11.95     |
| 5  | 0       | 0       | +1      | −1      | 1.11 ± 0.03                  | 2.28 ± 0.01                  | 0.91      | 7.16      |
| 6  | 0       | +1      | −1      | 0       | 2.02 ± 0.09                  | 2.12 ± 0.06                  | 6.11      | 6.53      |
| 7  | +1      | −1      | +1      | 0       | 4.92 ± 0.25                  | 3.56 ± 0.04                  | 13.84     | 11.03     |
| 8  | +1      | 0       | −1      | +1      | 5.64 ± 0.01                  | 8.72 ± 0.03                  | 15.03     | 18.81     |
| 9  | +1      | +1      | 0       | −1      | 2.59 ± 0.05                  | 2.05 ± 0.02                  | 8.27      | 6.24      |

*Oleoresin yield were presented as means of three independent replicates ± SD.

### Table 3. GC-MS analysis of various components inside the oleoresins.

| No | Component          | Formula   | Black Composition (% mass) | White Composition (% mass) | Identification |
|----|--------------------|-----------|----------------------------|----------------------------|----------------|
| 1  | \(\alpha\)-Thujene  | \(C_{10}H_{16}\) | –                          | 0.24                       | MS Rl Rl      |
| 2  | \(\alpha\)-Pinene   | \(C_{10}H_{16}\) | 1.82                       | 2.25                       | MS Rl Rl      |
| 3  | Sabinene           | \(C_{10}H_{16}\) | 5.91                       | 8.37                       | MS Rl Rl      |
| 4  | Limonene           | \(C_{10}H_{16}\) | 4.35                       | 6.32                       | MS Rl Rl      |
| 5  | Isotujhol          | \(C_{10}H_{18}O\) | –                          | 0.16                       | MS Rl Rl      |
| 6  | Linalool           | \(C_{10}H_{18}O\) | 1.20                       | 1.37                       | MS Rl Rl      |
| 7  | Terpinen-4-ol      | \(C_{10}H_{18}O\) | 0.50                       | 1.02                       | MS Rl Rl      |
| 8  | \(\alpha\)-Terpinol | \(C_{10}H_{18}O\) | 0.65                       | 0.95                       | MS Rl Rl      |
| 9  | Thymol             | \(C_{10}H_{18}O\) | –                          | 0.20                       | MS Rl Rl      |
| 10 | Borneol            | \(C_{10}H_{18}O\) | –                          | 0.35                       | MS Rl Rl      |
| 11 | Thymoquinol        | \(C_{10}H_{18}O\) | –                          | 0.32                       | MS Rl Rl      |
| 12 | Carvacrol          | \(C_{10}H_{18}O\) | –                          | 0.42                       | MS Rl Rl      |
| 13 | \(\alpha\)-Cubebene| \(C_{15}H_{24}\) | 0.10                       | 0.04                       | MS Rl Rl      |
| 14 | \(\beta\)-Elemene  | \(C_{15}H_{24}\) | 0.31                       | 0.52                       | MS Rl Rl      |
| 15 | Copaene            | \(C_{15}H_{24}\) | 0.29                       | 0.43                       | MS Rl Rl      |
| 16 | \(\gamma\)-Elemene | \(C_{15}H_{24}\) | 0.76                       | 0.87                       | MS Rl Rl      |
| 17 | Caryophyllene      | \(C_{15}H_{24}\) | 9.99                       | 11.21                      | MS Rl Rl      |
| 18 | \(\gamma\)-Murolene| \(C_{15}H_{24}\) | 2.02                       | 1.10                       | MS Rl Rl      |
| 19 | \(\alpha\)-Selinene| \(C_{15}H_{24}\) | 0.10                       | 0.22                       | MS Rl Rl      |
| 20 | \(\beta\)-Bisabolene| \(C_{15}H_{24}\) | 2.38                       | 3.10                       | MS Rl Rl      |
| 21 | Caryophyene oxide  | \(C_{15}H_{24}O\) | 1.00                       | 1.05                       | MS Rl Rl      |
| 22 | \(\beta\)-Sitosterin| \(C_{20}H_{30}O\) | 1.11                       | 1.43                       | MS Rl Rl      |
| 23 | 1-Cinnamoyl piperidine | \(C_{15}H_{22}N_{2}O_{3}\) | –                          | 0.03                       | MS Rl Rl      |
| 24 | Piperlongumimine   | \(C_{15}H_{22}N_{2}O_{3}\) | 0.03                       | 0.14                       | MS Rl Rl      |
| 25 | Piperine           | \(C_{15}H_{22}N_{2}O_{3}\) | 44.81                      | 54.67                      | MS Rl Rl      |
| 26 | Piperanine         | \(C_{15}H_{22}N_{2}O_{3}\) | 0.30                       | 0.45                       | MS Rl Rl      |
| 27 | Piperamide         | \(C_{15}H_{22}N_{2}O_{3}\) | 1.02                       | 0.98                       | MS Rl Rl      |
| 28 | Piperolein         | \(C_{15}H_{22}N_{2}O_{3}\) | 0.17                       | 0.22                       | MS Rl Rl      |
| 29 | Piperetine         | \(C_{15}H_{22}N_{2}O_{3}\) | –                          | 0.06                       | MS Rl Rl      |
| 30 | Guineensine        | \(C_{24}H_{31}NO_{3}\) | 0.22                       | 0.21                       | MS Rl Rl      |
| 31 | Guineensine        | \(C_{24}H_{31}NO_{3}\) | 0.31                       | –                          | MS Rl Rl      |
Figure 5. FT-IR analysis of the black and white oleoresin extracts.

represented the hydroxyl group band with intermolecular H-bonding. A moderately sharped asymmetric CH$_2$ stretching of 2933 cm$^{-1}$ and a weaker 2923 cm$^{-1}$ were observed for white and black extracts, respectively. 1621 and 1632 cm$^{-1}$ are the absorption bands of –CO-N group for white and black extracts, respectively. 1446 cm$^{-1}$ which correspond to the CH$_2$ bending were observed in white pepper extract. An asymmetric stretching of = C-O-C and in-plane bending of the phenyl CH gave an observed peak of 1260 and 1150 cm$^{-1}$ respectively in white and black extracts, respectively. 998 cm$^{-1}$ is a representation of vibrational CH-bending for trans –CH = CH- present in both extracts. The peaks at 929 and 849 cm$^{-1}$ was only present in white pepper extract. C-O-stretching (929 cm$^{-1}$) and out-of-plane C–H-bending (849 cm$^{-1}$) was found only in the white extract. Aromatic stretching of the phenyl ring was observed at 1370 cm$^{-1}$ and this was present only white pepper extract. Generally, the observed amide and phenyl group validated the antioxidant capacity of both extracts and this shared high resemblance with the IR-spectra obtained by Gupta et al. [25]. The antioxidant activities of natural products mainly attributed to the presence of hydroxyl groups attached to aromatic rings, such as phenolic compounds. These compounds are not detected when GC-MS Analysis is used, because they are subjected to degradation due to high temperatures. On the other hand, as the piperine (as an amide) is the major

Figure 6. SEM-monograph (a) black pepper pre-extraction (b) black pepper post-extraction (c) white pepper pre-extraction (d) white pepper post-extraction.
compound in both extracts, so it could be responsible for this biological activity [26].

3.6. Morphological characterization

Figure 6 illustrated the monographs from white and black pepper before and after extraction. The most identifying features of black and white pepper’s anatomy are the spread of outer peripheral bundles and inner medullary bundles [27]. The two bundles are made up of elongated and stone-like parenchymatous cells as shown in Figure 3(a) and 3(c). The induction of the cellulosic cell wall was the first step of the extraction process. The outer vascular bundles swell, paving way for solvents penetration into the cell wall. This results in the disorganization of membrane structure which later ruptures the cell wall. The large and elongated parenchymatous and stone-like cells become giving space for a reduced surface area as shown in Figure 6(b) and 6(d).

3.7. Antioxidant activities of oleoresin extracts

The result obtained from the antioxidant assay revealed that 94.92 and 107.57 μg/ml of white and black pepper extracts are respectively required to scavenge half of the stable DPPH radicals. However, 82.36 and 94.71 μg/ml of white and black pepper extracts are required to scavenge half of the stable ABTS free radicals. Extracts from white pepper, therefore, exhibited higher inhibition towards the stable DPPH and ABTS free radicals than black pepper extract. The lower IC50 for white pepper extract, therefore, suggests that it has higher antioxidant capacity than the black pepper as reported by Zhang and Xu [16]. The result from inhibitory plots indicated that the concentration-dependent DPPH anti-radical assay presented a more sensitive and efficient method than the ABTS assay.

4. Conclusion

The results obtained from black and white pepper microwave refluxation placed the optimal oleoresin yield at 5.64 and 8.72 w/w%, respectively. From the results obtained from radicals scavenging activity, it was concluded that white oleoresins extract were much higher in inhibitory properties than black oleoresin extract. The morphological and functional group analysis further confirmed structural changes during the process of extracting the bioactive oleoresins from the black and white pepper.

Conflict of interest

The authors declare no conflict of interest.

Disclosure statement

No potential conflict of interest was reported by the authors.

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