ABSTRACT

Objectives: In this study, a preliminary phytochemical screening and lipid matter of Arenga engleri Becc. leaves (family Arecaceae) were studied for the first time. Methods: Gas chromatography coupled with mass spectroscopy (GC/MS) was used for the identification of compounds of saponifiable and unsaponifiable content. Results: The preliminary phytochemical screening showed the presence of saponins, tannins, flavonoids, cardioglycosides, carbohydrate and/or glycosides, unsaturated sterols and/or triterpenes and absence of anthraquinones, coumarins, volatiles and alkaloids or compound containing nitrogenous bases. GC/MS analysis revealed the higher percentage of unsaturated fatty acids (51.39%) than that of saturated ones (31.47%). The major unsaturated fatty acids present were linoleic acid (31.55%) and 7,10-hexadecadienoic acid (11.42%) while the major saturated one was palmitic acid (17.27%). The unsaponifiable matter was represented as hydrocarbons (41.19%), fatty alcohols (28.31%), terpenes (9.68%) and sterol (0.14%). 1-Octadecene (17.65%) and 1-hexadecene (12.41%) represented the major hydrocarbons while behenic alcohol (14.71%) was the major fatty alcohol, phytol (4.89%) was the major terpene and ethylisoallocholate (0.14%) was the only sterol identified.

Keywords: Arenga engleri; GC/MS; Lipoidal matter; Phytochemical screening.

INTRODUCTION

Arecaceae, previously called the Palmae family, comprises about 200 genera and 2600 species which are distributed throughout tropical and subtropical regions.1,2 Palms are called the “Trees of Life” as they have a potential role in people’s life supplying them with foods, fibers, shelter, fuels, oils, gums, waxes, poisons and medicines.2 Genus Arenga contains 22 species, distributed in South China, the Ryukyu Islands and Taiwan in the North to Christmas Island in the South and from India in the West to Queensland, Australia in the East.3 The genus is economically important as it is useful for sugar, starch, thatch production and potentially has an ornamental value.4 Previous reports showed the identification of squalene, lutein, β-sitosterol and stigmasterol from Arenga tremula5 and RP-HPLC analysis of Arenga wightii revealed the presence of caffeine and major phenolic compounds: gallic acid, ascorbic acid and chlorogenic acid6. Different species were reported to have hypocholesterolaemic7, antioxidant, anti-microbial8, anti-hypertensive, anti-inflammatory and analgesic activities9 and treating skin allergies10, headache, malaria and tuberculosis10. Arenga engleri Becc., commonly called the Formosa palm, Taiwan sugar palm or dwarf sugar palm, is an attractive medium-sized ornamental clustering

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palm which is native to Taiwan (Formosa) and Ryukyu Islands (South of Japan). The stems are covered with
delicate black fibers which have been used for a long
time to prepare coir raincoat, shoes, rope, fishing nets,
brushes and pads. The leaves are known to be useful
in thatching and wickerwork. The young ones and
apical buds are edible, the stems pith used to produce
starch while the sweet sap obtained from the
inflorescence stem used to make sugar and the flowers
have a very pleasant fragrance. The mesocarp of the
fruits are rich in irritant calcium oxalate raphides like
other most species of this genus and this make them
inedible and poisonous. Since there are no reported
studies about phytochemical constituents of
*Arenga engleri* Becc. leaves and its lipoidal profile, it
was found essential to carry out preliminary
phytochemical screening and investigation of lipoidal
matter of this palm.

**MATERIALS AND METHODS**

**Plant material**

Fresh leaves of *Arenga engleri* Becc. were
collected on October 2014 from Al Zohriya Garden,
Cairo, Egypt and kindly identified by Dr. Terase Labib,
Head of the Taxonomists at Orman Botanical Garden,
Giza, Egypt. A Voucher specimen (03Aen/2019) was
kept in the herbarium of the Department of
Pharmacognosy, Faculty of Pharmacy, Helwan
University, Egypt.

**Chemicals**

For phytochemical screening:
1 % Hydrochloric acid, 5 % alcoholic potassium
hydroxide, concentrated ammonium hydroxide solution,
10 % alcoholic solution of α- naphthol, sulfuric acid,
1 % aluminum chloride solution, magnesium turning,
concentrated hydrochloric acid, 0.1 % ferric chloride
solution, 1 % lead acetate solution, picric acid, sodium
hydroxide, acetic anhydride, glacial acetic acid,
chloroform, Fehling’s solution, Barfoed’s reagent,
Mayer’s reagent, Dragendorff’s reagent.

For lipoidal matter: n-Hexane, 10 % alcoholic
potassium hydroxide, ether, 10 % 2N hydrochloric acid,
sulphuric acid, methanol.

**Apparatus**

TRACE™ 1310 Gas Chromatograph produced by Thermo Scientific™ provided by FID (Flame
Ionization Detector), attached with ISQ LT single
quadrupole Mass Spectrometer (Regional center for
Mycology and Biotechnology, Al-Azhar University).

**Methods**

**Preliminary phytochemical screening**

Air dried powdered leaves of *A. engleri* were
screened for its constituents using standard protocols in
the mentioned references.

**Preparation of lipoidal matter**

The air-dried powder of *A. engleri* leaves (100 g)
were extracted by *n*-hexane. The solvent was
evaporated at 40°C under reduced pressure to give 7 g
residue of lipoidal matter.

**Fractionation of lipoidal matter**

Two gm of lipoidal matter were saponified by
refluxing with 50 ml of 10% alcoholic potassium
hydroxide solution for 2 hr followed by evaporating the
alcohol, diluting with distilled water and extracting with
ether exhaustively. The collected ethereal extract was
washed with distilled water till being free from alkalinity,
dried over anhydrous sodium sulphate, and then evaporated to give 1.16 g (58%) unsaponifiable
matter (USM) residue.

The remaining saponifiable aqueous layer left
after extraction with ether was acidified with 10% 2 N
hydrochloric acid and the liberated fatty acids were
extracted exhaustively with ether. The collected
ethereal extract was washed with distilled water until
neutralization, dried over anhydrous sodium sulfate, and
then evaporated to give 0.54 g (27%) total fatty acids
(TFA) residue.

**Preparation of fatty acid methyl esters**

The preparation of methyl esters of free fatty
acids (0.54 g) was carried out by refluxing with 100 ml
of absolute methanol and 5 ml sulphuric acid for 2 hr,
extracting with ether and drying the ethereal layer over
anhydrous sodium sulfate followed by evaporation of
ether to give residue of the fatty acid methyl esters
(FAME), kept for GC-MS analysis.

**GC-MS analysis of the FAME**

Fatty acid methyl esters were analyzed according to the following conditions: TR-FAME,
Thermo 260 M 142 P (30 m, 0.25 mm ID, 0.25 μm
film), 70% cyanopropyl- polysilphenylene siloxane
capillary column. The used gas: Helium (1.5 ml/ min).
Injector temperature: 200°C; temperature transfer line:
250°C; initial column temperature 80°C, programmed
by 3°C/ min up to the final temperature 230°C within 50
min and the ionization energy was 70 ev.

**GC-MS analysis of the unsaponifiable matter**

The analysis was carried out under the following conditions: DB-17 P/N 122-1751 (30 m, 0.25
mm ID, 1 μm film), 50% phenyl-methylpolysiloxane
capillary column. The used gases: H₂, N₂, air. Injector
temperature: 280°C; temperature transfer line: 300 °C;
initial column temperature: 100°C, programmed by
Table 1. GC-MS analysis of USM of A. engleri leaves

<table>
<thead>
<tr>
<th>Identified compounds</th>
<th>RRT* (min)</th>
<th>Area %</th>
</tr>
</thead>
<tbody>
<tr>
<td>3-Phenyl decane</td>
<td>0.809</td>
<td>0.07</td>
</tr>
<tr>
<td>1-Hexadecene</td>
<td>0.837</td>
<td>12.41</td>
</tr>
<tr>
<td>6-Phenyl undecane</td>
<td>0.866</td>
<td>0.57</td>
</tr>
<tr>
<td>5-Phenyl undecane</td>
<td>0.869</td>
<td>1.10</td>
</tr>
<tr>
<td>4-Phenyl undecane</td>
<td>0.878</td>
<td>0.74</td>
</tr>
<tr>
<td>3-Phenyl undecane</td>
<td>0.896</td>
<td>0.49</td>
</tr>
<tr>
<td>n-Dotriacontane</td>
<td>0.923</td>
<td>0.33</td>
</tr>
<tr>
<td>2-Phenyl undecane</td>
<td>0.928</td>
<td>0.40</td>
</tr>
<tr>
<td>6-Phenyl dodecane</td>
<td>0.945</td>
<td>1.37</td>
</tr>
<tr>
<td>5-Phenyl dodecane</td>
<td>0.948</td>
<td>1.26</td>
</tr>
<tr>
<td>4-Phenyl dodecane</td>
<td>0.959</td>
<td>0.81</td>
</tr>
<tr>
<td>3-Phenyl dodecane</td>
<td>0.977</td>
<td>0.77</td>
</tr>
<tr>
<td>1-Octadecene</td>
<td>1</td>
<td>17.65</td>
</tr>
<tr>
<td>2-Phenyl dodecane</td>
<td>1.01</td>
<td>0.24</td>
</tr>
<tr>
<td>6-Phenyl tridecane</td>
<td>1.02</td>
<td>1.05</td>
</tr>
<tr>
<td>5-Phenyl tridecane</td>
<td>1.024</td>
<td>0.83</td>
</tr>
<tr>
<td>3-Phenyl tridecane</td>
<td>1.05</td>
<td>0.27</td>
</tr>
<tr>
<td>2-Phenyl tridecane</td>
<td>1.08</td>
<td>0.34</td>
</tr>
<tr>
<td>17-Pentatriacontene</td>
<td>1.17</td>
<td>0.49</td>
</tr>
<tr>
<td>1-Hexadecanol</td>
<td>0.66</td>
<td>5.52</td>
</tr>
<tr>
<td>Behenic alcohol</td>
<td>1.14</td>
<td>14.71</td>
</tr>
<tr>
<td>n-Tetracosanol-1</td>
<td>1.26</td>
<td>8.08</td>
</tr>
<tr>
<td>Phytol</td>
<td>1.22</td>
<td>4.89</td>
</tr>
<tr>
<td>Squalene</td>
<td>1.67</td>
<td>4.79</td>
</tr>
<tr>
<td>Ethylisoallocholate</td>
<td>1.09</td>
<td>0.14</td>
</tr>
<tr>
<td><strong>Total hydrocarbons</strong></td>
<td><strong>41.19</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Fatty alcohols</strong></td>
<td><strong>28.31</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Total terpenes</strong></td>
<td><strong>9.68</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Total sterols</strong></td>
<td><strong>0.14</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Total identified compound</strong></td>
<td><strong>79.32</strong></td>
<td></td>
</tr>
<tr>
<td><strong>Unidentified compounds</strong></td>
<td><strong>20.68</strong></td>
<td></td>
</tr>
</tbody>
</table>

*RRT*: Relative retention time of 1-Octadecene with RT = 29.83 min.
Figure 1. GC-chromatogram of unsaponifiable matter of A. engleri leaves.

Figure 2. GC-chromatogram of fatty acids of A. engleri leaves.

10 °C/min up to the final temperature 270 °C within within 30 min and the ionization energy was 70 ev.

**Identification of USM and TFA**

After GC-MS analysis of USM and TFA, the compounds were identified by comparing their retention times and mass fragmentation patterns with those of the reference standard data of WILEY and NIST libraries. Quantitative determination was based on peak area integration.

**RESULTS**

**Preliminary phytochemical screening**

Preliminary phytochemical screening of *A. engleri* leaves showed the presence of saponins, tannins, flavonoids, cardiac glycosides, carbohydrate and/or glycosides, unsaturated sterols and/or triterpenes while anthraquinones, coumarins, volatiles and alkaloids or compound containing nitrogenous bases were absent.

**Investigation of lipoidal matter of A. engleri leaves**

Fractionation of lipoidal matter of *n*-hexane extract of *A. engleri* leaves yielded 58% unsaponifiable matter and 27% fatty acids. As shown in Table 1 and Figure 1, GC-MS results of unsaponifiable matter revealed that leaves of *A. engleri* contain 41.19% hydrocarbons, 28.31% fatty alcohols, 9.68% terpenes and 0.14% sterols. The most abundant compounds identified in hydrocarbons content were 1-octadecene (17.65%) and 1-hexadecene (12.41%). Behenic alcohol (14.71%) was the major fatty alcohol. Total terpenes identified as 4.89% phytol and 4.79% squalene. Ethylisoallocholate was the only sterol identified.

As shown in Table 2 and Figure 2, the percentage of unsaturated fatty acids (51.39 %) was higher than saturated ones (31.47 %). The major unsaturated fatty acids were linoleic acid (31.55 %) and 7,10-hexadecadienoic acid (11.42%) while the major saturated one was palmitic acid (17.27 %).

**DISCUSSION**

The results of preliminary phytochemical screening of *Arenga engleri* leaves showed the presence of sterols and triterpene that was confirmed by the GC-MS analysis of its lipoidal matter. As revealed in GC-MS analysis, the percentage of unsaponifiable matter was higher than saponifiable one and unsaturated fatty acids were more than saturated ones. Linoleic acid and palmitic acid represented the major identified unsaturated and saturated fatty acids, respectively. For unsaponifiable matter, hydrocarbons represented the major component then fatty alcohols and then terpenes while the only sterol identified was ethylisoallocholate. The most abundant compounds identified in hydrocarbons content were 1-octadecene and 1-hexadecene. Behenic alcohol was the major fatty alcohols and total terpenes identified were phytol and squalene.

Many biological activities have been reported for linoleic acid as antioxidant, anti-inflammatory by cyclooxygenase-I (COX-I) and cyclooxygenase-II (COX-II) inhibition and hypcholesterolemic activities. Studies revealed that elevated levels of linoleic acid in the plasma prevented and controlled hypertension. It helped in glycemic control and reduced the risk of Type 2 diabetes and also had anticancer activity. Palmitic acid showed antibacterial,
Table 2. GC-MS analysis of total fatty acids of *A. engleri* leaves

<table>
<thead>
<tr>
<th>Identified compounds</th>
<th>RRT* (min)</th>
<th>Area%</th>
</tr>
</thead>
<tbody>
<tr>
<td>4- o xo, Pentanoic acid (levulinic acid) C5:0</td>
<td>0.243</td>
<td>5.07</td>
</tr>
<tr>
<td>4-Hydroxy-4-methylhex-5-enoic acid C7:1</td>
<td>0.389</td>
<td>0.27</td>
</tr>
<tr>
<td>10-methyl Undecanoic acid (Isolauric acid) C12:0</td>
<td>0.641</td>
<td>0.97</td>
</tr>
<tr>
<td>Nonanedioic acid (Azelaic acid) C9:0</td>
<td>0.664</td>
<td>0.35</td>
</tr>
<tr>
<td>Octadeic-6,9-dien-12-ynoic acid C18:2</td>
<td>0.715</td>
<td>0.04</td>
</tr>
<tr>
<td>cis-5,8,11,14,17-Eicosapentaenoic acid (Timnodonic acid) C20:5</td>
<td>0.764</td>
<td>0.19</td>
</tr>
<tr>
<td>Tetradecanoic acid (Myristic acid) C14:0</td>
<td>0.776</td>
<td>2.31</td>
</tr>
<tr>
<td>10,13-Octadecadiynoic acid C18:0</td>
<td>0.804</td>
<td>0.09</td>
</tr>
<tr>
<td>Pentadecanoic acid C15:0</td>
<td>0.839</td>
<td>1.68</td>
</tr>
<tr>
<td>7,10-Hexadecadienoic acid C16:2</td>
<td>0.882</td>
<td>11.42</td>
</tr>
<tr>
<td>9-Hexadecenoic acid (Z) (Palmitoleic acid) C16:1</td>
<td>0.888</td>
<td>1.95</td>
</tr>
<tr>
<td>Hexadecanoic acid (Palmitic acid) C16:0</td>
<td>0.903</td>
<td>17.27</td>
</tr>
<tr>
<td>9,12-Octadecadienoic acid (Z,Z) (linoleic acid) C18:2</td>
<td>1</td>
<td>31.55</td>
</tr>
<tr>
<td>9,12,15-Octadeatrienoic acid (linolenic acid) C18:3</td>
<td>1.003</td>
<td>5.52</td>
</tr>
<tr>
<td>Octadecanoic acid (Stearic acid) C18:0</td>
<td>1.011</td>
<td>2.00</td>
</tr>
<tr>
<td>Hexanoic acid C6:0</td>
<td>1.034</td>
<td>1.71</td>
</tr>
<tr>
<td>2-Octyl-cyclopropaneoctanoic acid C19:0</td>
<td>1.056</td>
<td>0.06</td>
</tr>
<tr>
<td>6,9,12-Octadecatrienoic acid C18:3</td>
<td>1.097</td>
<td>0.36</td>
</tr>
<tr>
<td>Docosanoic acid (Behenic acid) C22:0</td>
<td>1.112</td>
<td>0.05</td>
</tr>
</tbody>
</table>

**Saturated fatty acids**  
**Unsaturated fatty acids**  
**Unidentified compounds**  

**RRT*:** Relative retention time of linoleic acid with RT = 22.85 min
antifungal, antioxidant, anti-inflammatory and hypocholesterolemic activities28.

1-Octadecene showed anticancer, antioxidant and antimicrobial activities20. Behenic alcohol was a fatty alcohol of antiviral activity29 while 1-hexadecane had antibacterial, antifungal and antioxidant activities20. Comparably to other species, fatty acids like 3-nonenolic acid, 13-docosenoic acid, butanoic acid, and 2-hydroxyisobutyric acid were identified in leaf and fruit of Arenga wightii Griff. by GC-MS analysis6.

CONCLUSION

From preliminary phytochemical screening, it is revealed that Arenga engleri leaves are rich in saponins, tannins, flavonoids, cardiac glycosides, carbohydrate and/or glycosides, unsaturated sterols and/or triterpenes and lacking antiaruniones, coumarins, volatiles and alkaloids or compound containing nitrogenous bases. Lipoidal matter investigation by GC/MS analysis showed that leaves contain valuable compounds that suggest the probability of using this palm medicinally and it is the first report for lipoidal investigation and phytochemical screening of Arenga engleri leaves growing in Egypt.

Conflict of Interest

The authors declare that they don’t have any kind of conflict of interest.

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