Comparative Analysis of Phytochemical compounds in Normal and root gall of Okra plant

Apexa Pareek¹, Payal Lodha²

¹(Department of Botany, University of Rajasthan, India)
²(Department of Botany, University of Rajasthan, India)

Abstract: Okra, Abelmoschus esculentus (L.) Monech belongs to family Malvaceae and is widely cultivated in Tropical and Subtropical countries. In India, okra has been ranked first in its consumption. A multitude of threats on stable and secure yields of this crop exists including losses caused by pathogens like bacteria, virus, fungi and nematode. The root knot nematode (Meloidogyne incognita) infects root part of okra plant which leads to gall formation on root. The plant – pathogen interaction leads to production of increased secondary metabolites owing to the stress conditions. The secondary metabolites are supposed to provide resistance against pathogen. GC-MS analysis of the normal and galled root of Abelmoschus esculentus (L.)Monech leads to the finding that under stressed conditions larger no. of secondary metabolites were produced. Further studies on the efficacy of these secondary metabolites can result into various findings and discovery of novel and useful secondary metabolites resulting in increased resistance against pathogen to host plant.

Keywords: Abelmoschus esculentus, GC-MS, Galled root, Host-pathogen interaction, Meloidogyne incognita.

I. Introduction

Okra, Abelmoschus esculentus (L.) Monech is annual member of the Malvaceae family. It is native plant to tropical Africa, Asia and northern Australia. Okra has high fiber vitamin C and mucilage content [1]. Okra is also known for being high in antioxidants [2, 3]. The fruit of okra is extensively used as vegetables in tropical and subtropical countries. Mucilage content is also found in root of plants which have a strongly demulcent action [4]. The infusion of root is used for treatment of syphilis. The juice of root is used externally to treat cut, wounds and boils. Mucilage found in okra, is responsible for washing away toxic substances and bad cholesterol which loads the liver. Mucilage is supposed to be replacement of plasma. Due to having many medicinal and nutrition quality okra is widely cultivated in tropical and subtropical countries [5, 6, 7]. This crop is also attacked by various pathogens like bacteria, virus and root knot nematode. These pathogens cause biotic stress to the plant. A stress can lead into various results. Stress can have a devastating impact on plant growth and yield [8] or can result into enhancement of production of secondary metabolites [9]. These secondary metabolites are capable of triggering changes into plants cell which helps to overcome the stress [10]. Present study reveals the comparative analysis of stress (Galled Root) and non-stressed (Normal Root) condition of Okra Plant.

II. Material And Methods

2.1 Dry powder preparation

Plants were collected from field area of greenhouse of Department of Botany. The roots were separated from plants and washed with tap water to remove soil particle followed by distilled water. Normal and infected roots were cut into small pieces and were shade dried separately. Dried roots were pulverized to powder using mechanical grinder.

2.2 Preparation of Extract

About 5 gm powder of normal and infected root was weighed and was extracted with methanol (70-80°C) by hot continuous percolation method in soxhlet apparatus for 24 hours. The extract was taken and filtered through whatmann filter paper. Then extract was concentrated by rotary evaporator to obtain extract.

2.3 GC-MS analysis

The GC-Ms analysis of methanolic extract of normal and galled root of Abelmoschus esculentus was carried out on Shimadzu QP-2010 plus with thermal desorption system TD 20. It includes auto sampler and a gas chromatograph which interfaced to a mass spectrophotometer. The column size of this system is 30m × 0.25mm i.d × 0.26µm with a film thickness of 0.26mm, composed of 5MS ( 5% diphenyl/ 95% dimethyl poly siloxane). Helium gas (99.999%) was used as carrier gas at constant flow rate of 1ml/min. The 2µl injection volume of sample was utilized with split ratio of 10:1. The injector temperature was programmed initially at 280 °C, the ion-source temperature was 200 °C, the oven temperature was programmed from 110 °C (for 4 min), with an increase of 10 °C/min to 200°C, then 5 °C/min to 280°C, ending with a 9 min isothermal at 280
2.4 Identification of phytochemical

Interpretation of phytochemical present in the sample was conducted using NIST, having more than 62,000 patterns and Wiley8 Library. The comparison of unknown spectrum with known spectrum of various components was done by stored spectrum of NIST library and Wiley8 Library. The name, molecular weight and structure of the components were ascertained.

III. Result

GC-MS is a combined technique of Gas Chromatography with Mass Spectrophotometry. MS is wide ranging analytical technique, which identify the charged species according to their mass to charge ratio (M/Z). GC-MS is one of the best techniques to identify the constituents of volatile compounds. The GC-MS analysis of normal and galled root (infected root) of Abelmoschus esculents showed the presence of eight (Fig 1) and thirty five (Fig 2) phytochemical compounds respectively. The identification of the phytochemical compounds was confirmed based on the peak area, retention time and molecular formula. The active principles with their retention time (RT), area %, compound name, of normal and galled root are presented in Table 1 and 2 respectively.

IV. Discussion

The GC-MS analysis showed that the methanolic extract of normal root had fewer compounds than the galled root extract of okra plant. Normal root extract showed the major compound present as 3-deoxy-D-mannoic acid with 66.91% peak area. The next highest found compound was Hydroxy methyl furfural (HMF), retention time is 6.156 with 22.78% peak area. This compound is derivative of furan, which has potential to be sustainable substitute for petroleum [11]. HMF is obtained from sugars (carbohydrates) so that carbohydrates can be transformed into HMF [12]. This compound can be converted into DMF (2, 5-dimethyl furan), a liquid that is potential biofuel with greater energy content bioethanol [13]. Whereas GC-MS analysis of the extract of galled root showed 35 compounds. The major compound was Octdec-9-enioic acid with 37.21 peak area. It is monounsaturated fatty acid which decreases LDL cholesterol and blood pressure [14]. Second major compound is hexadecanoic acid whose consumption increase the risk of cardiovascular disease. Other compound such as various fatty acids, thymol, and methyl stearate were also found.

V. Conclusion

This study showed that the Galled root (under Stressed condition) led to production of more phytochemical compounds than the normal root (under non-stressed condition) of Okra plant.

Acknowledgement

The author of this paper is thankful to AIRF, JNU, Delhi for helping and providing necessary research facilities for this work.

References

Comparative Analysis of Phytochemical compounds in Normal and root gall of Okra plant

[12]. Andreia A. Rosatella, Svilen P. Simeonov, Raquel F.M. Frade, Carlos A. M. Afonso, Hydroxy methyl furfural as a building block platform: Biological properties synthesis and synthetic applications, Green Chem. 13, 2011, 754-793


Figures And Tables

Figure 1:- Shows GC-MS Chromatogram of Normal Root Part of Abelmoschus esculentus

DOI: 10.9790/3008-10625963  www.iosrjournals.org  61 | Page
Comparative Analysis of Phytochemical compounds in Normal and root gall of Okra plant

**Figure 2:** Shows GC-MS Chromatogram of Galled Roots of *Abelmoschus esculentus*

<table>
<thead>
<tr>
<th>Peak</th>
<th>R.Time</th>
<th>Area</th>
<th>Area%</th>
<th>Name Of Compounds</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4.192</td>
<td>1810819</td>
<td>2.96</td>
<td>Cyclopentane, 1-Acetyl-1,2-Epoxyp-</td>
</tr>
<tr>
<td>2</td>
<td>5.092</td>
<td>256436</td>
<td>4.20</td>
<td>4H-Pyrane-4-One, 2,3-Dihydroxy-6-Methyl-</td>
</tr>
<tr>
<td>3</td>
<td>6.516</td>
<td>13913901</td>
<td>22.78</td>
<td>Hydroxy Methyl Furfural</td>
</tr>
<tr>
<td>4</td>
<td>10.502</td>
<td>1600026</td>
<td>2.62</td>
<td>D-Allose</td>
</tr>
<tr>
<td>5</td>
<td>11.146</td>
<td>47913</td>
<td>0.08</td>
<td>3-Hexadecene, (Z)-</td>
</tr>
<tr>
<td>6</td>
<td>13.749</td>
<td>40866860</td>
<td>66.91</td>
<td>3-Deoxy-D-Mannionic Acid</td>
</tr>
<tr>
<td>7</td>
<td>15.243</td>
<td>148648</td>
<td>0.24</td>
<td>9-Octadecenoic Acid (Z)-</td>
</tr>
<tr>
<td>8</td>
<td>16.969</td>
<td>121207</td>
<td>0.20</td>
<td>7-Tetradecenal</td>
</tr>
</tbody>
</table>

Table 1: Compounds identified from methanolic extract of Normal Root Part of *Abelmoschus esculentus* using GC-MS analysis.
Comparative Analysis of Phytochemical compounds in Normal and root gall of Okra plant

<table>
<thead>
<tr>
<th>Peak</th>
<th>R.Time</th>
<th>Area</th>
<th>Area%</th>
<th>Name Of Compounds</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4.489</td>
<td>1336610</td>
<td>0.91</td>
<td>Butanedioic Acid, Monomethyl Ester</td>
</tr>
<tr>
<td>2</td>
<td>5.099</td>
<td>2637618</td>
<td>1.56</td>
<td>4H-Pyrano-4-One, 2,3-Dihydro-3,5-Dihydroxy-6-Methyl-</td>
</tr>
<tr>
<td>3</td>
<td>5.792</td>
<td>374533</td>
<td>0.22</td>
<td>Pyridine, 1-Acetyl-1,2,3,4-Tetrahydro-</td>
</tr>
<tr>
<td>4</td>
<td>6.899</td>
<td>280440</td>
<td>0.17</td>
<td>Nonanoic Acid</td>
</tr>
<tr>
<td>5</td>
<td>7.258</td>
<td>3108191</td>
<td>1.84</td>
<td>Thymol</td>
</tr>
<tr>
<td>6</td>
<td>8.334</td>
<td>237029</td>
<td>0.14</td>
<td>Decanoic Acid</td>
</tr>
<tr>
<td>7</td>
<td>10.325</td>
<td>4555412</td>
<td>2.69</td>
<td>3-Propanediol</td>
</tr>
<tr>
<td>8</td>
<td>10.925</td>
<td>476945</td>
<td>0.28</td>
<td>N,N-Bis(2-Hydroxyethyl)Dodecanamide</td>
</tr>
<tr>
<td>9</td>
<td>12.716</td>
<td>1097046</td>
<td>0.65</td>
<td>Tetradecanoic Acid, Methyl Ester</td>
</tr>
<tr>
<td>10</td>
<td>13.269</td>
<td>4378070</td>
<td>2.59</td>
<td>Tetradecanoic Acid</td>
</tr>
<tr>
<td>11</td>
<td>13.410</td>
<td>785199</td>
<td>0.46</td>
<td>Pentadecanoic Acid, Methyl Ester</td>
</tr>
<tr>
<td>12</td>
<td>13.907</td>
<td>2246318</td>
<td>1.33</td>
<td>Pentadecanoic Acid</td>
</tr>
<tr>
<td>13</td>
<td>14.332</td>
<td>549732</td>
<td>0.32</td>
<td>8-Octadecanone</td>
</tr>
<tr>
<td>14</td>
<td>14.648</td>
<td>1372959</td>
<td>0.81</td>
<td>9-Hexadecenoic Acid, Methyl Ester, (Z)-</td>
</tr>
<tr>
<td>15</td>
<td>14.858</td>
<td>9052909</td>
<td>5.35</td>
<td>Hexadecanoic Acid, Methyl Ester</td>
</tr>
<tr>
<td>16</td>
<td>15.197</td>
<td>2402527</td>
<td>1.42</td>
<td>9-Octadecanoic Acid</td>
</tr>
<tr>
<td>17</td>
<td>15.439</td>
<td>24611629</td>
<td>14.54</td>
<td>Hexadecanoic Acid &lt;N-&gt;</td>
</tr>
<tr>
<td>18</td>
<td>15.822</td>
<td>245662</td>
<td>0.15</td>
<td>Octadecanoic Acid, Methyl Ester</td>
</tr>
<tr>
<td>19</td>
<td>15.926</td>
<td>1226226</td>
<td>0.72</td>
<td>Heptadecanoic Acid</td>
</tr>
<tr>
<td>20</td>
<td>16.086</td>
<td>297333</td>
<td>0.18</td>
<td>Hexade-&lt;9Z&gt;-Enal</td>
</tr>
<tr>
<td>21</td>
<td>16.515</td>
<td>2896832</td>
<td>1.71</td>
<td>9,12-Octadecadienoic Acid (Z,Z)-, Methyl Ester</td>
</tr>
<tr>
<td>22</td>
<td>16.648</td>
<td>24462940</td>
<td>14.45</td>
<td>11-Octadecenoic Acid</td>
</tr>
<tr>
<td>23</td>
<td>16.784</td>
<td>5264190</td>
<td>3.11</td>
<td>Methyl Stearate</td>
</tr>
<tr>
<td>24</td>
<td>17.264</td>
<td>62991483</td>
<td>37.21</td>
<td>Octadec-&lt;9-Enoic Acid</td>
</tr>
<tr>
<td>25</td>
<td>17.353</td>
<td>6745045</td>
<td>3.98</td>
<td>Stearic Acid</td>
</tr>
<tr>
<td>26</td>
<td>17.967</td>
<td>307133</td>
<td>0.18</td>
<td>Heptadecene-&lt;9&gt;-Carbonic Acid-(1)</td>
</tr>
<tr>
<td>27</td>
<td>18.092</td>
<td>652289</td>
<td>0.39</td>
<td>5,8,11,14,17-Eicosapentaenoic Acid, Methyl Ester</td>
</tr>
<tr>
<td>28</td>
<td>18.227</td>
<td>284393</td>
<td>0.17</td>
<td>2,4,4-Trimethyl-3-(3-Oxo-But-1-Enyl)-Cyclohex</td>
</tr>
<tr>
<td>29</td>
<td>18.317</td>
<td>157394</td>
<td>0.095</td>
<td>Hexadecenal</td>
</tr>
<tr>
<td>30</td>
<td>18.414</td>
<td>887002</td>
<td>0.52</td>
<td>13-Docosenoic Acid, Methyl Ester, (Z)-</td>
</tr>
<tr>
<td>31</td>
<td>18.579</td>
<td>1317555</td>
<td>0.78</td>
<td>Methyl 10,12-Pentacosadiynoate</td>
</tr>
<tr>
<td>32</td>
<td>18.860</td>
<td>948034</td>
<td>0.56</td>
<td>Cis-11-Eicosanoic Acid</td>
</tr>
<tr>
<td>33</td>
<td>19.006</td>
<td>393140</td>
<td>0.23</td>
<td>Eicosanoic Acid</td>
</tr>
<tr>
<td>34</td>
<td>20.839</td>
<td>253705</td>
<td>0.15</td>
<td>Heneicosanoic Acid, Methyl Ester</td>
</tr>
<tr>
<td>35</td>
<td>24.015</td>
<td>255667</td>
<td>0.15</td>
<td>Heneicosanoic Acid, Methyl Ester</td>
</tr>
<tr>
<td></td>
<td>169289188</td>
<td>100.00</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 2: Compounds identified from methanolic extract of Galled Root Part of *Abelmoschusesculentus* using GC-MS analysis