

## Research Article



## A Study on Phytochemicals, Fatty acid Analysis and Antimicrobial Activity of *Abrus precatorius* Linn Seeds

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### ABSTRACT

*Abrus precatorius* Linn seeds called Gundumani in Tamil is a brightly colored seed, most seeds are black, red and has been used in Siddha medicine for centuries. It had been widely introduced by humans, and the brightly colored and hard-shelled seeds are spread by birds. By the end of the twentieth century, it had been announced as an invasive weed in many regions including Belize, Caribbean Islands, Hawaii, India, Polynesia and parts of the United States. In the present study, GC-MS was applied for the analysis of phytochemicals present in the ethanol extract of *Abrus precatorius* seeds. Likewise, fatty acids present were analyzed using hexane extract. Among the phytochemicals identified, lipid compound was found to be predominant in seeds whereas linoleic acid was predominant among fatty acids studied. It was found to be active against bacteria when compared to fungi.

**Keywords:** *Abrus precatorius*, Fatty acid methyl ester, GC-MS, Phytochemicals, Seeds.

### INTRODUCTION

**A***brus precatorius* Linn (Fabaceae) is a medicinal plant otherwise called as Indian Wild Liquorice, Jequirity, Crab's Eye and Precatory Bean in English, originates from Southeast Asia and now can be found in subtropical areas of the world. High-climbing, twining, or trailing woody vine with slender herbaceous branches. Leaves alternate, petioled, 5-13 cm (2-5 in) long, even-pinnately compound with 5-15 pairs of leaflets, these oval to oblong, up to 1.8 cm (< 1 in) long, with margins entire. Flowers shaped like pea flowers, white to pink or reddish, small, in short stalked dense clusters at leaf axils. Fruit are short, oblong pod, splitting before falling to reveal 3-8 shiny hard seeds, 6-7mm (<1 in) long, scarlet with black bases. The name *Abrus*, meaning beautiful or graceful, is used to describe the appearance of the seed. The seed is found in a variety of colors such as black, orange and most commonly, red with a glossy appearance with the black band at the end that attaches to the plant. Seeds contain abrin, a toxalbumin, indole derivatives, anthocyanins, sterols, terpenes. Abrin causes agglutination of erythrocytes, haemolysis and enlargement of lymph glands. Abrin isolated from the seeds of red variety, exhibited a noticeable increase in antibody-forming cells, bone marrow cellularity and alpha-esterase positive bone marrow cells. Oral administration of agglutinins, isolated from the seeds, is useful in the treatment of hepatitis and AIDS.<sup>1</sup> Seeds of *Abrus precatorius* are commonly used as purgative, emetic, aphrodisiac and for treating nervous disorder in traditional medicine.<sup>2</sup> This plant is also poisonous to horses.<sup>3</sup> Symptoms of poisoning include nausea, vomiting, convulsions, liver failure and death usually after several days. Ingesting a single seed can kill an adult human. The seeds have been used as beads in jewelry, which is

dangerous, inhaled dust is toxic and pinpricks can be fatal. The seeds are unfortunately attractive to children. An ethanolic extract of *Abrus precatorius* was found to have antioxidant, anti-inflammatory and analgesic potential in rodents.<sup>4</sup> A methanolic extract of *A. precatorius* seeds causes reversible alterations in the estrous cycle pattern and completely blocked ovulation in Sprague-Dawley rats.<sup>5</sup> The methanolic extract produces dose-dependent bronchodilator activity in a guinea pig model.<sup>6</sup> Hence, the present study was undertaken to study the phytochemicals, fatty acids present and also to find its antimicrobial activity. The phytochemicals as well as fatty acid profile was studied by means of GC-MS using ethanol and hexane extract.

### MATERIALS AND METHODS

#### Sample used for the study

The seeds were purchased from local market at Krishnagiri, Krishnagiri District, Tamil Nadu, India. The seeds purchased were cleaned, freed from debris and then grounded to obtain powder. The powdered seeds were transferred in to an air tight plastic container and used for analysis.

#### Extraction process

10gm of powdered samples were extracted with 30ml ethanol overnight and filtered. The extract was concentrated to 1ml by bubbling nitrogen in to the solution. 2µl of the ethanolic extract was employed for GC-MS analysis for the identification of phytochemical compounds.<sup>7</sup> Further, the given samples were extracted with hexane and methylated, analyzed through GC-MS for the identification of the fatty acid profile.



### GC-MS analysis

GC-MS analysis was carried out on a GC clarus 500 Perkin Elmer system and gas chromatograph interfaced to a mass spectrometer (GC-MS) instrument employing the following conditions: Column Elite-5MS fused capillary column (30 x 0.25mm x 0.25µm df) composed of 5% Diphenyl / 95% Dimethyl poly siloxane. Helium (99.999%) was used as carrier gas at a constant flow of 1ml per min and injection volume of 2µl was adopted (split ratio of 10:1) The injected sample was detected by Turbo mass gold detector (Perkin Elmer) with the aid of Turbomass 5.2 software. During the 36 minute GC extraction process, the oven temperature was programmed from of 110° C with an increase of 10° C/min up to 200° C, then 5° C/min upto 280° C (9 minutes hold). The injector temperature was set at 250° C (mass analyzer). Other parameters involved in the operation of Clarus 500MS, was also standardized (Inlet line temperature: 200° C; Source

temperature: 200°C). Mass spectra were taken at 70eV and fragments from 45-450 Da. The MS detection was completed in 36 minutes. The detection employed the NIST (National Institute of Standards and Technology) - Year 2005 library.

### Antimicrobial Assay

The antimicrobial activity was assessed by the method of Kirby-Bauer disk diffusion method.<sup>8</sup> Results of the zone of inhibition was observed and measured after 24hr period of incubation time.

### RESULTS AND DISCUSSION

Gas chromatography and mass spectrometry is an effective combination for chemical analysis. The results of various phytochemicals observed are depicted in Table 1 and the chromatogram is shown in Figure 1. Similarly, the fatty acid profile is depicted in Table 2 and Figure 2, While antimicrobial activity is shown in Table 3.

**Table 1:** Bioactive Components identified in *Abrus precatorius* Linn seeds [GC MS study]\*

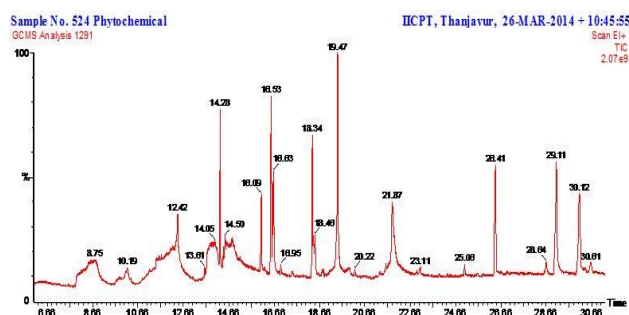
No.	RT	Name of the compound	Molecular Formula	MW	Peak Area %
1	8.75	α-D-Glucopyranose, 4-O-α-D-galactopyranosyl-	C <sub>12</sub> H <sub>22</sub> O <sub>11</sub>	342	12.57
2	10.19	α-D-Glucopyranoside, O-α-D-glucopyranosyl-(1.fwdarw.3)-α-D-fructofuranosyl	C <sub>18</sub> H <sub>32</sub> O <sub>16</sub>	504	2.68
3	12.42	n-Hexadecanoic acid	C <sub>16</sub> H <sub>32</sub> O <sub>2</sub>	256	9.78
4	14.05	d-Glycero-d-ido-heptose	C <sub>7</sub> H <sub>14</sub> O <sub>7</sub>	210	10.35
5	14.28	Palmidrol	C <sub>18</sub> H <sub>37</sub> NO <sub>2</sub>	299	4.26
6	14.50	9,12-Octadecadienoic acid (Z,Z)-	C <sub>18</sub> H <sub>32</sub> O <sub>2</sub>	280	1.42
7	16.09	Undecanal, 2-methyl-	C <sub>12</sub> H <sub>24</sub> O	184	2.63
8	16.53	Pyrrolidine, 1-(1-oxo-7,10-hexadecadienyl)-	C <sub>20</sub> H <sub>35</sub> NO	305	4.56
9	16.63	8,11,14-Eicosatrienoic acid, (Z,Z,Z)-	C <sub>20</sub> H <sub>34</sub> O <sub>2</sub>	306	2.88
10	16.95	7-Methyl-Z-tetradecen-1-ol acetate	C <sub>17</sub> H <sub>32</sub> O <sub>2</sub>	268	0.31
11	18.34	Cyclooctyl S-2-(dimethylamino)ethyl propylphosphonofluoridate	C <sub>15</sub> H <sub>32</sub> NO <sub>2</sub> PS	321	4.54
12	19.47	1,2-Benzenedicarboxylic acid, diisooctyl ester	C <sub>24</sub> H <sub>38</sub> O <sub>4</sub>	390	11.84
13	21.87	5,8-Octadecadienoic acid, methyl ester	C <sub>19</sub> H <sub>34</sub> O <sub>2</sub>	294	11.10
14	23.11	8,11,14-Eicosatrienoic acid, (Z,Z,Z)-	C <sub>20</sub> H <sub>34</sub> O <sub>2</sub>	306	0.41
15	25.06	Cucurbitacin b, 25-desacetoxy-	C <sub>30</sub> H <sub>44</sub> O <sub>6</sub>	500	0.53
16	26.41	γ-Tocopherol	C <sub>28</sub> H <sub>48</sub> O <sub>2</sub>	416	4.53
17	28.64	Cholestan-3-ol, 2-methylene-, (3á,5à)-	C <sub>28</sub> H <sub>48</sub> O	400	0.84
18	29.11	Stigmasterol	C <sub>29</sub> H <sub>48</sub> O	412	8.08
19	30.12	α-Sitosterol	C <sub>29</sub> H <sub>50</sub> O	414	6.02
20	30.61	Corymbolone	C <sub>15</sub> H <sub>24</sub> O <sub>2</sub>	236	0.68

\*Parameters tested are not covered under the scope of NABL accreditation

**Table 2:** Fatty acids identified in *Abrus precatorius* Linn seeds [GC MS study]

No.	RT	Name of the compound	Molecular Formula	MW	Peak Area %
1	13.94	9-Octadecenoic acid (Z)-, methyl ester	C <sub>19</sub> H <sub>36</sub> O <sub>2</sub>	296	38.54
2	14.70	9,12-Octadecadienoic acid (Z,Z)-	C <sub>18</sub> H <sub>32</sub> O <sub>2</sub>	280	61.46

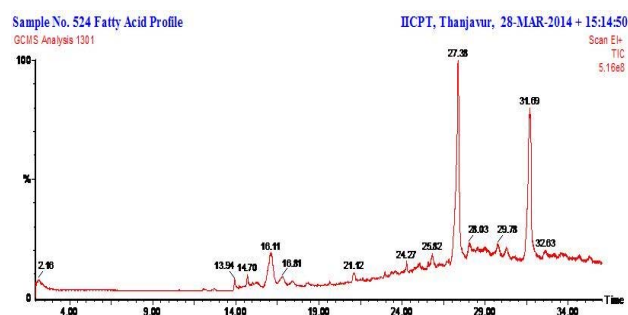




**Figure 1:** Chromatogram showing phytochemicals present in *Abrus precatorius* Linn seeds

### Phytochemicals

Totally 20 compounds were identified in *Abrus precatorius* Linn seeds. The compound 1 identified was  $\alpha$ -D-Glucopyranose, 4-O- $\alpha$ -D-galactopyranosyl- C<sub>12</sub>H<sub>22</sub>O<sub>11</sub>, having peak area percent as 12.57. Compound 2 identified was  $\alpha$ -D-Glucopyranoside, O- $\alpha$ -D-glucopyranosyl-(1.fwdarw.3)- $\alpha$ -D-fructofuranosyl, C<sub>18</sub>H<sub>32</sub>O<sub>16</sub>, showing 2.68 as peak area percent. Compound 3 identified was n-Hexadecanoic acid, C<sub>16</sub>H<sub>32</sub>O<sub>2</sub>, with peak area percent as 9.78. Compound 4 identified was d-Glycero-d-ido-heptose, C<sub>7</sub>H<sub>14</sub>O<sub>7</sub>, showing 10.35 as peak area percent. Compound 5 identified was palmidrol, C<sub>18</sub>H<sub>37</sub>NO<sub>2</sub>, having 4.26 as peak area percent. Compound 6 identified was 9, 12,-Octadecadienoic acid (Z, Z)-, C<sub>18</sub>H<sub>32</sub>O<sub>2</sub>, with peak area percent of 1.42. Compound 7 identified was Undecanal, 2- methyl-, C<sub>12</sub>H<sub>24</sub>O, showing 2.63 as peak area percent. Compound 8 identified was Pyrrolidine, 1-(-oxo-7,10-hexadecadienyl)- C<sub>20</sub>H<sub>35</sub>NO, having 4.56 as peak area percent. Compound 9 identified was 8, 11, 14- Eicosatrienoic acid, (Z, Z, Z)-C<sub>20</sub>H<sub>34</sub>O<sub>2</sub>, shows peak area percent of 2.88. Compound 10 identified was 7-Methyl-Z-tetradecen-1-ol acetate, C<sub>17</sub>H<sub>32</sub>O<sub>2</sub>, having peak area percent of 0.31. Compound 11 identified was Cyclooctyl S-2-(dimethylamino) ethylpropyl phosphonofluoridate, C<sub>15</sub>H<sub>32</sub>NO<sub>2</sub>PS, with peak area percent of 4.54. Compound 12 identified was 1, 2-Benzenedicarboxylic acid, diisooctyl ester, C<sub>24</sub>H<sub>38</sub>O<sub>4</sub>, showing 11.84 as peak area percent. Compound 13 identified was 5, 8-Octadecadienoic acid, methyl ester, C<sub>19</sub>H<sub>34</sub>O<sub>2</sub>, having 11.10 as peak area percent. Compound 14 identified was 8, 11, 14-Eicosatrienoic acid, (Z,Z,Z)-, C<sub>20</sub>H<sub>34</sub>O<sub>2</sub>, with peak area percent as 0.41. Compound 15 identified was Cucurbitacin b, 25-desacetoxy-C<sub>30</sub>H<sub>44</sub>O<sub>6</sub>, showing peak area percent as 0.53. Compound 16 identified as  $\zeta$ -Tocopherol, C<sub>28</sub>H<sub>48</sub>O<sub>2</sub>, having peak area percent as 4.53. Compound 17 identified as Cholestan-3-ol, 2-methylene-, (3 $\alpha$ ,5 $\alpha$ )-, C<sub>28</sub>H<sub>48</sub>O, showing peak area percent as 0.84. Compound 18 identified was Stigmasterol, C<sub>29</sub>H<sub>48</sub>O, shows peak area percent of 8.08. Compound 19 identified was  $\alpha$ -Sitosterol, C<sub>29</sub>H<sub>50</sub>O, having peak area of 6.02 percent. Compound 20 identified was Corymbolone, C<sub>15</sub>H<sub>24</sub>O<sub>2</sub>, with peak area of 0.68 percent. Lipid compounds were present as an important phytochemical in this seeds. The retention time starts at 8.75 and ends at 30.61. The molecular weights of the compound varied from 184 to 500.



**Figure 2:** Chromatogram showing fatty acid profile of *Abrus precatorius* Linn seeds

### Fatty acids

Fatty acids present are 9-Octadecenoic acid (Z)-, methyl ester, C<sub>19</sub>H<sub>36</sub>O<sub>2</sub>, showing peak area percent as 38.54 and 9, 12-Octadecadienoic acid (Z,Z)-, C<sub>18</sub>H<sub>32</sub>O<sub>2</sub>, with peak area percent as 61.46.

### Antimicrobial activity

**Table 3:** Antimicrobial activity Assay of *Abrus precatorius* Linn seeds\*

Microorganisms	Zone of Inhibition (mm)	
	50 $\mu$ g	100 $\mu$ g
<i>Aspergillus niger</i>	12	15
<i>Aspergillus flavus</i>	14	12
<i>Escherichia coli</i>	12	24
<i>Staphylococcus aureus</i>	20	22

\*Parameters tested are not covered under the scope of NABL accreditation

The antimicrobial activity of *Abrus precatorius* Linn seeds were tested against bacteria and fungi. Among the microbes tested, it was found to be active against *E.coli*, *Staphylococcus aureus* when compared to fungi.

### CONCLUSION

Presently phytomedicines are gaining recognition and about 80% of the world population depends on plant derived medicine for primary health care because it has minimum or no side effects. It is concluded from the study, that *Abrus precatorius* Linn seeds were found to be rich in linoleic acid which find its use in pharmacology. Even though, this seed is a poisonous one, it can be used for the cure of various ailments that affect animals, insects and humans. Further purification might give more valuable phytochemicals.

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