Antioxidant activity of different extracts of various parts (Leaves, Stem and Root) of Achyranthes Aspera

Vijay Kumar and Dr. Rakesh Kumar Jat

Abstract
Rural and marginal people of India depend upon the medicinal plants compare to synthesis drugs. Natural source of medicine have less cost and less side effect beside to synthesis drugs.so now day the herbal drugs are more popular in rural and city. Natural source are very important for Scientists towards to search of medication for various number of diseases Cost-effective, easy availability. The present study was aimed to investigation and designed to evaluate the antioxidant, activity of different plant parts of achyranthes aspera leaf, stem and root extracts in various solvent by using DPPH assay method for evaluation of radical scavenging activity. In this study different solvent such as hexane, benzene and ethyl acetate were subjected. Different extracts exhibited various types activities because it is depend upon the nature and properties of solvent. These activities are time and concentration depended. Ascorbic acid was used for the positive control. This research finding that ethanolic extracts of achyranthes aspera leaves showed the highest DPPH radical scavenging activity and hexane extracts of root of Achyranthes aspera exhibited the minimum antioxidant activity. Antioxidant activities of different extracts of achyranthes aspera were compared with each other. Ethanol extracts of leaves showed the highest DPPH radical scavenging activity at various concentrations 87.67 at 500 μg/ml, 81.21 at 400 μg/ml, 76.49 at 300 μg/ml, 68.86 at 200 μg/ml and 65.86 at 100 μg/ml. Dose depended antioxidant activity was observed in ethanolic extracts of leaves. Hexane extracts of root of Achyranthes aspera exhibited the minimum antioxidant activity. DPPH radical have scavenging activity so it is used in determine the antioxidant activity of different plant extracts of achyranthes aspera and compare to each other. Ascorbic acid was used as a standard and exhibited the maximum antioxidant activity. Different extracts of leaves of achyranthes aspera were compared with each other.

Keywords: Antioxidant, Achyranthes aspera, DPPH, ROS

Introduction
The plant territory has been the greatest and most important source of medicinal Preparations. Herbal medicine used to treat disease and promote health. There are many types of plant which are used from many years for curing the disease. Plant contains different types of substances like carbohydrates, lipids, proteins, glycosides, alkaloids, Tannins, flavonoids etc. responsible for theirs pharmacological activity. Plants have their different pharmacological and therapeutic importance. Different parts of plant which contain biologically active ingredients like root, bark, stem and leaf are used for treatment of acute and chronic ailments like Asthma, fever, hypertension, malaria, fungal, bacterial infection and heart disorder. These medicines are safe and effective to the proper treatment of different types of disease. According to WHO more than 20K species of traditional herbs of medicines are used form 80% of words people depend upon herbal medicines substance which are isolated form plants to be less expensive than synthetic drugs pharmaceutical products isolated from plants are at times found to be less costly than Synthetic drugs. Achyranthes aspera have rich source of bioactive and therapeutically active constituents and exhibiting the diversity and variation in bioactive components.

Achyranthes aspera is pungent, antiperiodic, purgative, diuretic and laxative and used in treatment of asthma, bleeding, in facilitating delivery, boils, bronchitis, cold, whooping cough, heart disease, dyspepsia, gonorrhea, colic, debility, pile, pneumonia, renal complications, scorpion bite, snake bite round consciousness and skin diseases etc. Molecules containing free radicals ROS are a family of active and involved in oxidative the modulation of biological cell functions. However, excessive ROS bring about stresses that cause injury to such as lipid, protein and DNA, various cellular constituents, senescence or apoptosis. Intracellular ROS can be finally resulting in growth arrest generated from aberrant mitochondria, which are well-known as sites of ROS generation and targets for ROS action. They cause cellular damage ROS are useful animal and plant host defense on the other hand, if produced in an uncontrolled
manner. As signaling molecules and in continued exposure to the harmful actions of ROS damages macromolecules and contributes significantly to the pathology of various human diseases. All aerobic cells generate ROS, including superoxide, $\text{H}_2\text{O}_2$ and hydroxyl radicals, enzymatically or non-enzymatically. The mitochondrial electron transport chain is the principal site of cellular production of ROS. ROS can also be generated during the process of phagocytosis, ischemia/reperfusion and metabolism of many drugs and other xenobiotic chemicals. Preservative antioxidant suppresses the free radicals formation. The accurate process of free radicals formation, mechanism of free radicals formation is not well explained right now. Hydroxide and hydrazide peroxides are the metal induced decomposition and capable to suppress such type reaction. Lipid hydro peroxidase like glutathione-s-transferase, phospholipid hydro-peroxidase glutathione peroxidase and glutathione can reduce lipid peroxides to water and hydro peroxides of phospholipids.

Material and Methods
The plant of Achyranthes aspera was collected from the Botanical garden of Shekhawati College of Pharmacy, Dundlod Rajasthan in the month of November 2016. The identity of the plant was confirmed at Department of Botany, University of Rajasthan Jaipur. After collection washed carefully and dried to constant weight at 45°C. The plants were stored in an air-tight container. All chemicals and solvents were of analytical grade purchased from local source. The selected plant extracts which were dissolved in DMSO (100-500μg/ml) were run in triplicate and averaged. Lower absorbance of 517nm by UV-Visible spectrophotometer. All the test analysis were received through same procedure.

Determination of ethanol-soluble extractives
All parts of plant achyranthes aspera were separated and converted in the coerce powder after drying. Then shaded the plant part through sieve no.40 and successively extracted with different solvent by soxhlet extraction. Around 20 g of crushed air dried powders were extracted with ethanol, hexane, chloroform and ethyl acetate solution in a soxhlet apparatus for 72 hrs. The obtained solutions were filtered and evaporated through rotary flash evaporator and kept in a refrigerator. All extracts were received through same procedure.

Determination of DPPH radical scavenging activity on selected plant extracts
The free radical scavenging activity of the Hexane extract, Chloroform, Ethyl acetate extract and ethanolic extract of selected plants were evaluated using 1,1 diphenyl-2-picyr hidrazyl (DPPH) assay method. In its radical shape DPPH absorbs at 517nm however reduction in the absorbance at 517 nm was determined.1ml of 0.25 mm solution of DPPH in DMSO was delivered to the exclusive concentrations of selected plant extracts which were dissolved in DMSO (100-500μg/ml). After 30 min, the absorbance was measured at 517nm by UV-Visible spectrophotometer. All the test analysis were run in triplicate and averaged. Lower absorbance of reaction mixture indicates higher free radical scavenging activity. Ascorbic acid used as a positive control and standard compound in this antioxidant activity.

Table 1: Achyranthes aspera root extracts on DPPH radical Scavenging model

<table>
<thead>
<tr>
<th>Concentration (μg/ml)</th>
<th>100</th>
<th>200</th>
<th>300</th>
<th>400</th>
<th>500</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ascorbic acid</td>
<td>75.23±0.22</td>
<td>78.33±0.18</td>
<td>84.64±0.17</td>
<td>87.69±0.65</td>
<td>95.66±0.07</td>
</tr>
<tr>
<td>A.S.H.E</td>
<td>04.85±0.00</td>
<td>05.98±0.33</td>
<td>07.54±0.30</td>
<td>12.81±0.35</td>
<td>19.72±0.49</td>
</tr>
<tr>
<td>A.S.E.A.E</td>
<td>34.68±0.56</td>
<td>52.55±0.90</td>
<td>57.45±0.98</td>
<td>59.64±0.23</td>
<td>62.95±0.39</td>
</tr>
<tr>
<td>A.S.E.E</td>
<td>42.05±0.90</td>
<td>43.22±0.77</td>
<td>44.95±0.90</td>
<td>46.82±0.76</td>
<td>52.04±0.54</td>
</tr>
<tr>
<td>A.S.C.E</td>
<td>45.34±0.67</td>
<td>58.15±0.09</td>
<td>64.15±0.23</td>
<td>67.68±0.56</td>
<td>68.99±2.89</td>
</tr>
</tbody>
</table>

Fig 1: Result for DPPH radical Scavenging model of Achyranthes aspera root extracts

DPPH free radicals are decolorize by the reaction of antioxidant compound and they further reacts with DPPH radical produce the purple colour and its convert in to 1-1-diphenyl-2-picyr hydrazine have the colorless property, and mainly measured at 517 nm. This is used in vitro antioxidant activity to determine the DPPH radical scavenging activity of crude plant extracts. Achyranthes aspera root extracts has been showed the potent scavenging activity at 500μg/ml in ethanol, Ethyl acetate, Hexane and chloroform which is 52.04±0.54, 62.95±0.39,19.72±0.49, 68.99±2.89 as compared to the positive control ascorbic acid . These activities are lower than ascorbic acid. Ascorbic acid showed Highest DPPH scavenging activity compare to other. The DPPH scavenging activities of Ascorbic acid was 75.23±0.22, 78.33±0.18, 84.64±0.17, 87.69±0.65 and 95.66±0.07 at different concentration of 100μg/ml, 200 μg/ml, 300 μg/ml, 400 μg/ml and 500 μg/ml. The results are presented in FG. DPPH radical scavenging activities are in dose dependent manner when the concentration increase extract inhibition of DPPH radical is also increased. Antioxidant plays a powerful role in food as well as biological system that evaluation the antioxidant activity.

The antioxidant activities of selected different extracts of achyranthes aspera were in the following order- A.S.B.E> A.S.E.A.E> A.S.E.E > A.S.H.E.
Table 2: *Achyranthes aspera* stem extracts on DPPH radical Scavenging mode

<table>
<thead>
<tr>
<th>Concentration</th>
<th>100 μg/mL</th>
<th>200 μg/ml</th>
<th>300 μg/ml</th>
<th>400 μg/ml</th>
<th>500 μg/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ascorbic acid</td>
<td>75.23±0.22</td>
<td>78.33±0.18</td>
<td>84.64±0.17</td>
<td>87.69±0.65</td>
<td>95.66±0.07</td>
</tr>
<tr>
<td>A.S.H.E</td>
<td>15.86±0.43</td>
<td>15.86±0.99</td>
<td>26.49±0.77</td>
<td>34.21±0.08</td>
<td>37.67±0.62</td>
</tr>
<tr>
<td>A.S.E.A.E</td>
<td>22.29±0.32</td>
<td>26.49±0.32</td>
<td>36.69±0.90</td>
<td>40.18±0.90</td>
<td>43.77±0.32</td>
</tr>
<tr>
<td>A.S.E.E</td>
<td>45.83±0.55</td>
<td>58.65±0.32</td>
<td>72.83±1.45</td>
<td>85.13±0.56</td>
<td>86.13±0.33</td>
</tr>
<tr>
<td>A.S.C.E</td>
<td>48.42±0.56</td>
<td>61.13±0.65</td>
<td>73.04±0.34</td>
<td>78.40±0.23</td>
<td>80.77±1.78</td>
</tr>
</tbody>
</table>

The outcomes of DPPH radical scavenging assay of *Achyranthes aspera* stem is showed and summarized in Table. Different extracts stem of *Achyranthes aspera* has been showed the robust scavenging activity at 500 μg/ml in ethanol, Ethyl acetate, Hexane and chloroform which is 86.13±0.33, 43.77±0.32, 37.67±0.62 and 80.77±1.78 as compared to the positive control ascorbic acid. These activities are lower than ascorbic acid. Ascorbic acid showed Maximum DPPH scavenging activity compare to other Different extracts of *Achyranthes aspera* exhibited the antioxidant activities in the following order- A.S.C.E > A.S.E.A.E > A.S.E.E > A.S.H.E.

Table 3: *Achyranthes aspera* leaves extracts on DPPH radical Scavenging model

<table>
<thead>
<tr>
<th>Concentration</th>
<th>100 μg/mL</th>
<th>200 μg/ml</th>
<th>300 μg/ml</th>
<th>400 μg/ml</th>
<th>500 μg/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ascorbic acid</td>
<td>28.42±0.43</td>
<td>31.08±0.33</td>
<td>36.51±0.99</td>
<td>37.33±0.44</td>
<td>41.24±0.51</td>
</tr>
<tr>
<td>A.S.C.E</td>
<td>47.03±0.7</td>
<td>49.04±0.32</td>
<td>60.65±0.67</td>
<td>65.02±0.32</td>
<td>72.76±0.12</td>
</tr>
<tr>
<td>A.S.H.E</td>
<td>24.95±0.78</td>
<td>28.58±0.12</td>
<td>41.50±0.98</td>
<td>42.50±0.43</td>
<td>45.58±0.89</td>
</tr>
<tr>
<td>A.S.E.E</td>
<td>65.86</td>
<td>68.86</td>
<td>76.49</td>
<td>81.21</td>
<td>87.67</td>
</tr>
</tbody>
</table>

Different extracts of leaves of *Achyranthes aspera*. Extracts exhibited the potent sifting activity at 500 μg/ml in ethanol, ethyl acetate, and hexane and chloroform solution. Which were 72.76±0.12, 45.58±0.89, 41.24±0.51 and 45.58±0.89 as compared to the positive control ascorbic acid used for positive control these activities are less potent when compare with ascorbic acid. Antioxidant activities of a substance are widely uses for evaluating the antioxidant activity of a substance in both food and biological systems hence it may be a powerful antioxidant.

**Conclusion**

In this study the different extract of the various plant parts *Achyranthes aspera* used for the antioxidant activity. *Achyranthes aspera* root, leaves and stem were found to be effective in DPPH radical scavenging activity in different solvent plant extracts. Ethanolic extract of selected *Achyranthes aspera* is the authentic choice to protect from oxidation.

So present study supports the traditional usage as antioxidant and exhibited the interesting inhibitory antioxidant activity against the reactive oxygen species this study will be helpful in the treatment of free radicals scavenging disease and based on the present study further works will be carried out in future.

**Acknowledgement**

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**Reference**