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Evaluation of Antioxidant activity of *Euphorbia hirta* by Hydrogen peroxide radical Scavenging activity

Shubhra Medhashri Mishra^{1*}, Anupam Kumar Pathak¹ and Pradeep Kumar Sharma²

¹Department of Pharmacy B.U Bhopal (M.P) ²Department of Applied Chemistry SATI Vidisha (M.P)

ABSTRACT

The present study was aimed to evaluate antioxidant property of entire herb of Euphorbia hirta by using Hydrogen peroxide radical scavenging method using ascorbic acid as standard. The ability of plant extracts to scavenge hydrogen peroxide is determined according to them. Hydrogen peroxide (H_2O_2) enters the human body through inhalation of vapours or mist and through eye or skin contact. In the body, H_2O_2 is rapidly decomposed into oxygen and water and this may produce hydroxyl radicals (OH) that can initiate lipid per oxidation and cause DNA damage. Absorbance of hydrogen peroxide at 230nm was measured. The Euphorbia hirta can be regarded as promising entrant for natural plant sources of antioxidants with high value.

Keywords: Antioxidants, Antioxidant activity, ascorbic acid, radical scavenging, Hydrogen peroxide.

INTRODUCTION

Majority of the diseases/disorders are generally associated to oxidative stress due to free radicals[1]. Free radicals are fundamental to many biochemical processes and represent an essential part of aerobic life and metabolism. [2]. Hydrogen peroxide occurs naturally at low concentration levels in the air, water, human body, plants, microorganisms, food and beverages. It is widely used as a bleaching agent in the textile, paper and pulp industries. Human beings exposed to H₂O₂ indirectly via the environment are estimated as 0.28 mg/kg/day with intake from leaf crops contributing most to this exposure. Hydrogen peroxide (H_2O_2) enters the human body through inhalation of vapour mist and through eye or skin contact. In the body H_2O_2 rapidly decomposed into oxygen and water and this may produce hydroxyl radicals (OH⁻) that can initiate lipid per oxidation and cause DNA damage [3] .The antioxidant phytochemicals such as polyphenols, flavonoids and associated compounds found in medicinal plants have received increasing attention for their potential role in prevention of human diseases [4]. According to WHO more than 21000 plants are used for medical treatment in all over the world [5]. Present study deals with the plant Euphorbia hirta (family Euphorbiaceae) synonym of E pilulifera and Chamaesyce which is commonly known as dudhi, Garden spurge, asthma weed etc .The plant is used for various actions likewise, sedative, antidiabetic analgesic, antipyretic, antimicrobial, anxiolytic, wound healing etc and antioxidant property [6] [7] [8]. Hence the objective of this investigation was to determine hydrogen peroxide radical scavenging activity in ethanolic extract of Euphorbia hirta.

MATERIALS AND METHODS

Plant material

The entire plant of *Euphorbia hirta* was collected from various localities of Bhopal (M.P) during the month of March .The plant was identified and authenticated by Dr. H.B Singh, raw material, Herbanium and Museum NISCAIR, New Delhi with Ref No. NISCAIR / RHMD /Consult /2013/2214/220.

Chemical: Ethanol, Methanol, Ascorbic acid, Hydrogen peroxide (H_2O_2) , Phosphate buffer saline, distilled water all the solvents and chemicals were of analytical grade.

Preparation of the Plant Extracts:

Entire herb of *Euphorbia hirta* was dried in shade. The shade dried plant material was crushed to get a coarse powder. About 250gm of dried powder was extracted with ethanol by soxhlet apparatus. Extract was concentrated till extract become only 15 to 20ml.

Resulting crude extracts was collected and kept in vacuum oven to remove final traces of solvent. The ethanolic extract on dry basis was found to be approx. 80gm.

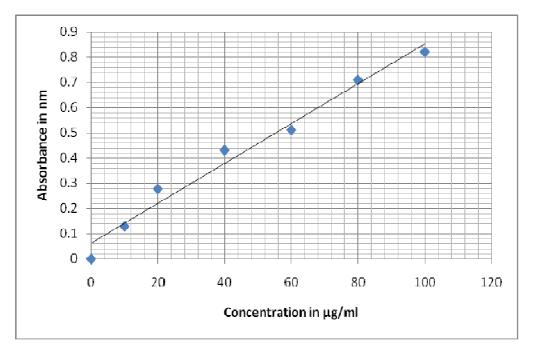
Determination of Hydrogen peroxide radical scavenging:

The ability of plant extracts to scavenge hydrogen peroxide is determined according to the method of Rachh et al (2009) [9]. A 20Mm solution of hydrogen peroxide was prepared in buffer saline (P_H 7.4).1ml of various concentrations of plant extract and standard ascorbic acid solution (10, 20,40,60,80 and 100 µg/ml in methanol) was added to 2ml of hydrogen peroxide solution. For making concentration of 10µg/ml, 1ml of this stock is added to 9ml of water respectively. Absorbance of hydrogen peroxide at 230nm was determined after 10minutes against a blank solution containing phosphate buffer without hydrogen peroxide.

20Mm solution of H_2O_2 in phosphate buffer saline (P_H 7.4), from this solution we will take 2ml solution of H_2O_2 and 1ml of extract solution. Whereas standard stock is prepared by adding 10mg ascorbic acid (0.01) in 100ml. From this stock and standard solution we take 1ml with 9ml of water ie; 10ml (conc.is $10\mu g/ml$) further dilution were made up to $80\mu g/ml$. From various dilutions we take 1ml solution with 2ml solution of H_2O_2 .

Concentration of various Dilution and its respective UV absorbance:

| a various Dilution a | iu its respective |
|----------------------|-------------------|
| Conc. in µg/ml | Absorbance |
| 10µg/ml | 0.1292 |
| 20µg/ml | 0.2781 |
| 40µg/ml | 0.432 |
| 60µg/ml | 0.512 |
| 80µg/ml | 0.7103 |
| $100 \mu g/ml$ | 0.822 |
| | |



The percentage inhibition was calculated using the following formula:

Percentage Inhibition = [(A_0 - A_t / A_0) ×100]

Where A_0 is the absorbance of the control and A_t is the absorbance of test extract. All the tests and analysis were run in triplicates and averaged.

 $\frac{0.2781 - 0.2608}{0.2781} \times 100 = 6.220\%$

Thus in total extract of 80gm, the hydrogen peroxide scavenging activity is found 6.220%.

RESULTS AND DISCUSSION

Ethanolic extract of *Euphorbia hirta* were prepared to examine the hydrogen peroxide radical scavenging and for antioxidant activity measurement the yield of extract obtained from 250g of dry plant material was found to be 80gm. The hydrogen peroxide radical scavenging in the examined extract using ascorbic acid as positive control ranged to 6.220%. The scavenging activity was quantified by measuring inhibition of the degradation of the deoxyribose by free

radicals. When extract and reference compound ascorbic acid was added to reaction mixture they removed hydroxyl radicals from the sugar and prevented their degradation.

CONCLUSION

Based on the results of our study we conclude that the significant importance of the species *Euphorbia hirta* for its therapeutic use. Based on this information, it could be concluded that this plant is natural sources of antioxidant substances of high importance in preventing or slowing oxidative stress related degenerative diseases. However further studies of this plant species should be directed to bring out in vivo studies of its medicinal active components in order to prepare natural pharmaceutical products of high value.

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