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Research Article

SCREENING OF VEGETABLE EXTRACTS FOR ANTIOXIDANT ACTIVITY USING REDOX REACTIONS – BY COLORIMETRY

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Abstract

Vegetables are well known for their antioxidant properties which can help prevent many diseases. Crude extracts of 15 vegetables were screened for their invitro antioxidant content. The total antioxidant content of the fresh vegetable extracts was determined colorimetrically. The antioxidant activity was expressed in terms of EC50 values which varied from 0.0192µg/ml to 0.00672µg/ml. The vegetables studied possess valuable antioxidant properties for culinary and nutritive use. The contribution of various vegetables to total antioxidant intake was the main objective of our study. Among all the vegetables studied, bottle gourd and coccinea were found to have high antioxidant potency as shown by EC 50 values.

Keywords: invitro, antioxidant properties, EC50, colorimetry.

1.Introduction

Vegetables are good source of biologically active natural products. The fresh extracts of vegetables rich in antioxidants are very important in food industry , since they retard the oxidative degradation of lipids, fight free radicals that damage healthy cells and improve the nutritional value of food (1).They do so by inhibiting the initiation and propagation of oxidative chain reactions (2) Free radicals are responsible for oxidative chain reactions. There are several enzyme systems in the body that can scavenge free radicals, but we can also gain these molecules from the food we eat. The phenolic components such as flavonoids (3) phenolic acids, phenolic diterpenes are responsible for antioxidative effect. Antioxidants inhibit cell proliferation and development of abnormal preneoplastic and neoplastic cells (4). Increased consumption of fruits and vegetables containing high levels of phytochemicals is recommended to prevent chronic diseases related to oxidative stress in human body. Functional foods that contain significant amounts of bioactive components provide desirable health benefits beyond basic nutrition and prevent chronic diseases (5).

Several accepted methods of evaluating antioxidant activity exists invitro(6) . In the present paper antioxidant activity was determined by colorimetry involving redox reactions. EC 50 values of each of the vegetable extract was determined. EC50 value is a measure of drug potency. EC50 is the half maximal effective concentration and refers to the concentration of a drug antibody or toxicant which induces response halfway between the baseline and maximum after a specified exposure time (7) .

2.Materials and Methods

The 15 vegetables studied as shown in table-1 were purchased from local market of Banjarahills, Hyderabad, India.

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2.1 Extraction:

Fresh vegetables were collected , cleaned and ground to a fine paste, squeezed with a muslin cloth and the extract collected was further filtered using whattman filter paper no: 42 to remove any suspended particles .1ml of the pure extract was diluted up to 100ml using distilled water.

2.2 Antioxidant activity (Potassium Permanganate free radical scavenging activity) Determination

The antioxidant activity of the vegetable extracts was examined on the basis of the scavenging effect on standard Potassium permanganate free radical activity (8) (Brace et.al. ,2002) . Potassium Permanganate was standardized using ferrous ammonium sulphate. The method is based on redox reactions between vegetable extracts and Potassium permanganate in sulphuric acid medium, leading to sample discoloration (9). Potassium Permanganate is a strong oxidizing agent which gets reduced from Mno_4^- (dark purple) to Mn^{2+} ions (colorless) on addition of the vegetable extracts. No additional indicators are needed and reduction of permanganate requires strong acidic conditions. Standard Potassium permanganate solution (0.00048M) was added to each of the above mentioned vegetable extracts (shown in Table-1) in the presence of sulphuric acid medium and optical density values were noted using colorimeter at 520nm .Selection of wavelength was done (10) . as shown in table 2. Distilled water was used to set the absorbance to zero and the instrument was calibrated.

Results and Discussion

In the Present study the free radical scavenging activities of vegetable extracts (Shown in Table -1) were evaluated colorimetrically using standard Potassium permanganate which is a powerful oxidizing agent. Redox reaction method is a sensitive way to determine the antioxidant activities of vegetable extracts (Kolear.et.al) . Standard Potassium permanganate solution (0.00048M) was found to give maximum absorbance at 520 nm as shown in Table-2. Hence all the observations were done at 520 nm .

Table – 1 Characteristics of the vegetables used

S.No	English name	Botanical name	Family name	Therapeutic uses
1	Cabbage	*Brassica oleracea var. capitata*	cruciferae	prevention of cancer , anti-inflammatory etc.
2	Bottle gourd	*Lagenaria siceraria*	cucurbitiacae	diuretic,sedative,antibilious,to treat urinary disorders,other nerveous disorders etc.
3	cuccumber	*Cucumis sativus*	cucurbitiacae	treat digestion problem , anti-bilious etc.
4	Green mango	*Mangifera indica*	Anacardiaceae	tenderising agent , to treat gastro intestinal disorders etc.
5	Beet root	*Beta vulgaris l*	chenopodiaceae	antitumour ,carminative,ammenagouge,and heostatic properties etc
6	Tomato	*Solanum lycopersicum*	solanceae	to treat high blood pressure, osteoarthritis and digestive disorders etc
7	Curry leaves	*Murraya koenigii*	Rutaceace	Prevents Anemia, Hair Treatments ,Digestion etc
8	Carrot	*Daucus carota subsp. sativus*	Apiaceae	Eyes health,inflammation ,digestion etc
9	Ivy gourd	*Coccinia grandis*	cucurbitaceae	Urinary tract infections,eczema,sprains etc
10	Mint	*Mentha Longigolia*	Lamiaceace	Irritable bowel Syndrome, stomach ache ,nausea etc
11	onion	*Allium.cepa*	Amaryllidaceae	Control sugar levels,blood pressure ,prevention of cancer etc
12	Coriander	*Corriandrum Sativum*	Apiaceae	leveling blood glucose level in diabetics, nausea ,vomiting,etc
13	spinach	*Spinacia oeracea*	amaranthaceae	Anaemia ,neurological disorders etc
14	Karela	*Momoridica Charantia*	Cucurbitacae	Antidaibetic,laxative,emetic,anthalmintic agent, lowering blood glucose level etc.
15	False Daisy	*Wedelia Calendulaceaae*	Asteraceae	Hair , Skin Treatments etc

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Table – 2 Selection of wavelength

S.no	***Wavelength***	***Optical Density/ Absorbance***
1.	450	0.19
2.	470	0.37
3.	510	0.49
4.	**520**	**0.66**
5.	540	0.59
6.	570	0.31
7.	600	0.10
8.	670	0.04

Table -3 concentration of $Kmno_4$ vs Optical density

S.No	**Concentration $Kmno_4$ (µg/ml)**	**Optical Density**
1.	0.0004	0.11
2.	0.0008	0.23
3.	0.00012	0.34
4.	0.00016	0.44
5.	0.00020	0.53
6.	0.00048	0.66

Table -4 EC50 values of Vegetable plant extracts

S.no	**English name of Medicinal Plants**	**Botanical name**	**EC50 VALUES (µg/ml)**
1.	Cabbage	*Brassica oleracea var. capitata*	0.00192
2.	Bottle gourd	*Lagenaria siceraria*	0.00144
3.	cuccumber	*Cucumis sativus*	0.0024
4.	Green mango	*Mangifera indica*	0.00192
5.	Beet root	*Beta vulgaris*	0.00192
6.	Tomato	*solanum lycopersicum*	0.00192
7.	Curry leaves	*Murraya koenigii*	0.0024
8.	Carrot	*Daucus carota subsp. sativus*	0.00192
9.	Ivy gourd	*Coccinia grandis*	0.00144
10	Mint	*Mentha Longigolia*	0.00672
11.	onion	*Allium.cepa*	0.00192
12.	Coriander	*Corriandrum Sativum*	0.00384
13.	spinach	*Spinacia oeracea*	0.00384
14.	Karela	*Momoridica Charantia*	0.00312
15	False Daisy	*Wedelia calendulaceaae*	0.00672

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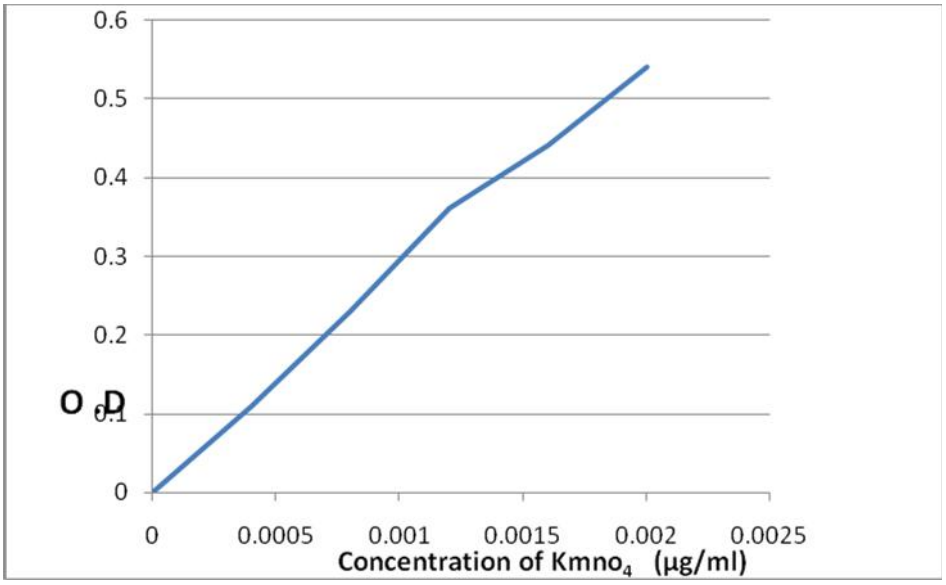
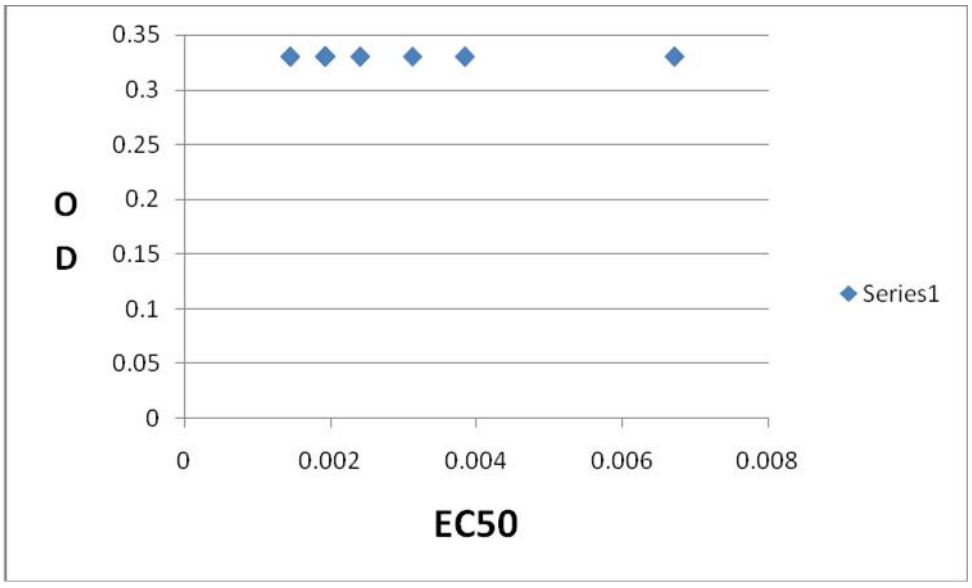
The optical density values of different concentrations of Potassium permanganate were noted at 520 nm and a plot of optical density Vs. concentration Potassium permanganate was drawn as shown in figure -1 . A straight line (calibration curve) passing through the origin was obtained .

Figure -1 Plots of Concentration $Kmno_4$ (µg/ml) vs optical density



The total antioxidant activity studies were done by Redox reactions colorimetrically and EC50 values for each plant extract were obtained from the optical density of potassium permanganate (0.66), concentration of potassium permanganate (0.00048M) and from the plots of OD vs. concentration of vegetable extracts as shown Figure-2

Figure-2 Plots optical density vs EC50 (µg/ml)



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Conclusion

The Free radical scavenging action of the vegetable extracts are in the order *Lagenaria siceraria , Coccinia grandis > Brassica oleracea , Mangifera indica , Beta vulgaris > Solanum lycopersicum , Daucus carota ,Allium cepa > Murraya koenigii , Cucumis sativus > Momoridica Charantia > Spinacia oeracea ,Corriandrum Sativum > Mentha Longigolia , Wedelia calendulaceaae . Lagenaria siceraria* and *Coccinia grandis* were found to be the most potent that can scavenge free radicals as shown by their lowest EC50 Values and *Mentha longigolia* and *Wedelia calendulaceaae* are the least potent as shown in Table-4 .

The study revealed that the vegetables studied had moderate to significant antioxidant activity .The increased antioxidant potency of *Lagenaria siceraria , Coccinia grandis* can be attributed to the presence of high polyphenolic contents (11) .

The above analysis was done colorimetrically and found to be much easier with reproducible results and less time consuming compared to other available procedures .

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