



In-vivo Pharmacological Evaluation of Cucurbita pepo and Benincasa hispida for Antioxidant activity

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Abstract: Analysis of the free radical scavenging activities of the selected *Cucurbita pepo* fruit and *Benincasa hispida* fruits extracts revealed a concentration dependent free radical scavenging activity resulting from reduction of DPPH, NO, Hydroxyl radical and superoxide radical radical to non-radical form. The scavenging activity of Ascorbic acid, a known antioxidant used as positive control, was however higher. DPPH radical is considered to be a model for a lipophilic radical. A chain in lipophilic radicals was initiated by the lipid autoxidation. DPPH is a stable free radical at room temperature and accepts an electron or hydrogen radical to become a stable diamagnetic molecule. The reduction capacity of DPPH was determined by the decrease in its absorbance at 517nm, which is induced by antioxidant. Positive DPPH test suggests that the samples were free radical scavengers. The scavenging effect of l-Ascorbic acid, and plant extracts increased gradually with increase in concentration. Nitric oxide plays an important role in various types of inflammatory processes in the body.

In the present study the fruit extracts of selected *Cucurbita pepo* and *Benincasa hispida* checked for its inhibitory effect on Nitric oxide production. Nitric oxide radical generated for sodium nitroprusside at physiological pH was found to be inhibited by the extracts. Results revealed that all the tested extracts showed the percentage of inhibition in a dose dependent manner. The extracts were exhibiting significant scavenging activity towards 1, 1-di phenyl picryl hydrazyl, Nitric oxide, Hydroxyl, Super oxide radicals. The activity

was found to be concentration dependent. In DPPH model the free radical scavenging capacity was found to be highly significant when compare other three models. In all the three selected plants Ethanol extract was found to have high scavenging activity than Ethyl acetate and petroleum ether extracts. Scavenging activity of ethanol extracts may be due to presence of the flavonoids and phenolic.

Introduction:

Green plants synthesize and preserve a variety of biochemical products, many of which are extractable and used as chemical feed stocks or as a raw materials for various scientific investigations. Many secondary metabolites of plant are commercially important and find use in a number of pharmaceutical compounds. [1]. There are decades of using specific plants or certain plants parts in the treatment of several health conditions throughout the world. [2].

Antioxidants may be defined as any substance, when present at low concentration compared with those of an oxidizable substrate, significantly delays or prevents oxidation of that substrate in a chain reaction [3]. Humans have evolved a highly complicated antioxidant protection system, which involves a variety of endogenous and exogenous compounds that are able to function interactively and synergistically to neutralize free radicals. These include antioxidant enzymes that catalyze free radical quenching reactions, metal binding proteins that sequester free iron and copper ions that are capable of catalyzing oxidative reactions, diet-derived antioxidants, and other low molecular weight compounds such as α -lipoic acid Antioxidants have become a popular research topic because they cannot be generated by the human body and hence have to be consumed in the diet [4]. Many fruit and vegetables have been found to be rich sources of antioxidants. Since a large portion of the human diet is based on fruit and vegetables, it is important to understand the biological and biochemical interactions between these dietary antioxidants and living systems [5]. The fruit of *Cucurbita pepo* Linn and *Benincasa hispida* are easily available in our country and being cheap can be widely used by the poor rural people and tribal. Considering the easy availability reduced cost and minimal side effects and information from the literature of discrete applications in digestive tract

of the fruits of the plant, the study has the following aims and objectives. So, the aim of our study is to elucidate the antioxidant role of extract of ripe fruit's pulp of *Cucurbita pepo* Linn. and *Benincasa hispida* for using different experimental model.

Material and Methods: The proposed work includes fruits of *Cucurbita pepo* Linn and *Benincasa hispida* are extract evaluated for antioxidant role using different experimental model.

Procurement and authentication of *Cucurbita pepo* and *Benincasa hispida* fruit: As the crude drugs form the basis for the manufacture of wide range of medicinal preparations needed by people, the development of pharmacognostical research has become indispensable for procuring therapeutically potent medicine prepared from genuine drug material. Fresh and fully grown fruits of *Cucurbita pepo* and *Benincasa hispida* were purchased from local market of Sagar India. The plants were authenticated Botanist of Department of Botany, Government College Khimlasa, Sagar. The fruits were washed properly with water. The seeds from fruits of *Cucurbita pepo* and *Benincasa hispida* were separated. The fruit were cut into small pieces, dried in sun and ground with the help of an electrical grinder to get powder, stored in airtight containers and used for phytochemical and pharmacological studies.

The macroscopical description fruits of *Cucurbita pepo* and *Benincasa hispida* include size, shape, nature of outer and inner surfaces, types of fracture, and organoleptic characters like color, odour, taste etc. were studied

Preliminary Phytoprofiles screening of *Cucurbita pepo* and *Benincasa hispida*: *Cucurbita pepo* and *Benincasa hispida* fruit powdered (100 g) was successively extracted with the following solvents of increasing polarity in a soxhlet apparatus. The dried powder was packed in Soxhlet apparatus extracted with 250 ml of petroleum ether for 72 h at 50°C. After the extraction, extract filtered and solvent was removed with the help of rotatory evaporator. The same process was carried out to get ethyl acetate and ethanol extracts. The total yield of the extracts obtained after removing the solvents was calculated. All the extracts were concentrated by distilling the solvents and the extracts were dried in an oven. Each time before extracting with the next solvent, the marc was

dried in an air. The consistency, color, appearance of the extracts and their percentage yield were noted.

Anti-oxidant Activity: Antioxidants may be defined as any substance, when present at low concentration compared with those of an oxidizable substrate, significantly delays or prevents oxidation of that substrate in a chain reaction. Antioxidants have become a popular research topic because they cannot be generated by the human body and hence have to be consumed in the diet. Many fruit and vegetables have been found to be rich sources of antioxidants. Since a large portion of the human diet is based on fruit and vegetables, it is important to understand the biological and biochemical interactions between these dietary antioxidants and living systems.

Chemicals and plant extract: 1,1-diphenyl-2-picryl hydrazyl (DPPH), DMSO, Ascorbic acid Sodium nitro prusside, Griess reagent (1% w/v sulphanilamide, 2% w/v H₃PO₄ and N-(1-naphthyl) ethylene diamine di hydrochloride), Hydrogen peroxide (H₂O₂) Thiobarbituric acid Sodium phosphate buffer, 1mM ferric chloride Nitro blue tetrazolium Phenazene methosulphate, Nicotinamide adenine phosphate di nucleotide

PECP: Petroleum ether extract of Cucurbita pepo (100-500µg/ml)

EACP: Ethyl acetate extract Cucurbita pepo (100-500µg/ml)

EOCP: Ethanol extract of Cucurbita pepo (100-500µg/ml)

PEBH: Petroleum ether extract of Benincasa hispida (100-500µg/ml)

EABH: Ethyl acetate extract of Benincasa hispida (100-500µg/ml)

EOBH: Ethanol extract of Benincasa hispida (100-500µg/ml)

Procedure: Antioxidant actions might be exerted by inhibiting generation of reactive oxygen species and reactive nitrogen species or by directly scavenging free radical or by raising the levels of endogenous antioxidant enzymes by up regulating expression of the genes encoding superoxide dismutase, catalase or glutathione peroxidase. For the assessment of free radical scavenging activity, the extracts of selected plants Petroleum ether, ethyl acetate and Ethanol extracts of Cucurbita pepo and Benincasa hispida fruits

were dissolved in 5% DMSO. DPPH, Nitric oxide, hydroxyl radical, superoxide radical methods were carried in the present study.

Determination of DPPH radical scavenging activity of Cucurbita pepo and Benincasa hispida fruits extracts: The free radical scavenging activity of the Petroleum ether, ethyl acetate and Ethanol extracts of Cucurbita pepo and Benincasa hispida fruits were evaluated using 1,1 diphenyl-2-picryl hydrazyl (DPPH) [6]. In its radical form, DPPH absorbs at 517nm, but upon reduction by an antioxidant or a radical species, the absorption decreases. 1ml of 0.25mM solution of DPPH in DMSO was added to the different 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 (shimadzu UV-Vis1800). All the test analysis were run in triplicate and averaged. Lower absorbance of reaction mixture indicates higher free radical scavenging activity. Ascorbic acid was used as a positive control. Ascorbic acid was used as a standard. The percentage DPPH decolorization of the sample was calculated by the equation:

$$\text{Percentage of DPPH scavenging} = [(A_0 - A_1) / A_0] \times 100,$$

A₀ = Absorbance of the control, and

A₁ = Absorbance of the extract/ standard.

Determination of Nitric Oxide (NO) radical scavenging activity of Cucurbita pepo and Benincasa hispida fruits extracts: Nitric oxide radical scavenging activity the method used was based on the standard methods with some modification [7]. Nitric oxide (NO) was generated from sodium nitro prusside (SNP) and was measured by the griess reagent (1% w/v sulfanilamide, 2% w/v H₃PO₃ and 0.1% w/v N-(1-Naphthyl) ethylene diamine dihydrochloride). SNP in aqueous solution at physiological PH spontaneously generates NO, which interacts with oxygen to produce nitrite ions that can be estimated by the use of griess reagent. Scavengers of NO compete with oxygen leading to reduced production of NO. SNP (1ml of mM) was mixed with 1ml of selected plants extracts in different concentrations (100µg/ml-500µg/ml) in (Di methyl sulphoxide) DMSO. The

mixture was incubated at 25°C for 180 minutes. To 1ml of the incubated solution, 1ml of Griess reagent was added. The absorbance of the chromophores formed during the diazotization of nitrite with sulphanilamide and subsequent coupling with N-(1-naphthyl) ethylene diamine dihydrochloride was read at 546nm by UV-Visible spectrophotometer (Shimadzu UV-Vis1800). All the test analysis were run in triplicate and averaged. Lower absorbance of reaction mixture indicates higher free radical scavenging activity. Ascorbic acid was used as a positive control. The percentage inhibition was calculated by

$$\text{Percentage of Nitric oxide scavenged} = [(A_0 - A_1)/A_0] \times 100,$$

Where A_0 = Absorbance of the control, and

A_1 = Absorbance of the extract/ standard.

Determination of hydroxyl radical scavenging activity of Cucurbita pepo and Benincasa hispida fruits extracts:

Hydroxyl radical scavenging activity was determined by the method based on the ability to compete with deoxyribose for hydroxyl radical [8]. Hydroxyl radical produced by the reduction of H_2O_2 by iron in the presence of ascorbic acid degrades deoxyribose to form products, which on heating with 2-thiobarbituric acid (TBA) form a pink coloured chromogen. The reaction mixture, of a final volume of 1ml, containing 0.4ml of 20mM sodium phosphate buffer (pH 7.4), 0.1ml of 100-500 $\mu\text{g/ml}$ of selected plant extracts in DMSO, 0.1ml of 60mM deoxyribose, 0.1 ml of 10mM H_2O_2 , 0.1 ml of 1mM ferric chloride, 0.1 ml of 1mM Ethylene diamine tetra acetic acid (EDTA) and 0.1ml of 2mM L-ascorbic acid, was incubated at 37°C for 1 h. The reaction was terminated by the addition of 1 ml of 17mM TBA and 1ml of 17mM trichloro acetic acid (TCA). The mixture was boiled for 15 min, cooled in ice and the absorbance was measured at 532nm by UV-Visible spectrophotometer (Shimadzu UV-Vis 1800). All the test analysis were run in triplicate and averaged. L-Ascorbic acid was used as a positive control. The hydroxyl radical scavenging of the extract was reported as the percentage of inhibition of the deoxyribose degradation and was calculated according to the following equation

Calculation of % Inhibition:

$$\% \text{ Inhibition} = [(A_0 - AT) / A_0] * 100$$

Where A₀ = Absorbance of the control, and

AT = Absorbance of the extract/ standard.

Determination of super oxide radical scavenging activity of Cucurbita pepo and Benincasa hispida fruits extracts: Super oxide anion derived from dissolved oxygen by a PMS-NADH coupling reaction reduces nitroblue tetrazolium(NBT), which forms a violet coloured complex [9]. A decrease in colour after addition of the antioxidant is a measure of its super oxide scavenging agent¹⁴. The experiment, the super oxide radicals were generated in 3 ml of Tris-HCL buffer(16Mm pH 8) containing 1 ml of NBT (50 μM), 1 ml NADH (78 μM) and test solution of selected plant extracts ,(100-500 μg/ml) to the above reaction mixture, 1 ml PMS solution(10 μ M) was added and incubated at 25°C for 5 min . The absorbance was read at 560nm (Shimadzu UV-Vis 1800) against blank sample. Decrease in absorbance of the reaction mixture incubated increases super oxide anion scavenging activity. All the test analysis were run in triplicate and averaged. L-ascorbic acid was used as a positive control.

The % inhibition of super oxide anion generated was calculated using the following equation

$$\% \text{ inhibition} = [(A_0 - AT) / A_0] * 100$$

Where A₀ = Absorbance of the control, and

AT = Absorbance of test extract/ standard.

PECP: Petroleum ether extract of Cucurbita pepo (100-500μg/ml)

EACP: Ethyl acetate extract Cucurbita pepo (100-500μg/ml)

EOCP: Ethanol extract of Cucurbita pepo (100-500μg/ml)

PEBH: Petroleum ether extract of Benincasa hispida (100-500μg/ml)

EABH: Ethyl acetate extract of Benincasa hispida (100-500μg/ml)

EOBH: Ethanol extract of *Benincasa hispida* (100-500µg/ml)

Result and Discussion:

Procurement and Authentication of Plant material (*Cucurbita pepo* and *Benincasa hispida*): Pharmacognostic study helps in confirmation and determination of identity, purity and quality of a crude drug. Morphological characterization of *Cucurbita pepo* reveals that it is a sprawling vine with yellow fruit-bearing flowers. The of *Cucurbita pepo* have a mild flavor. Size and weight of fruit may vary. *Benincasa hispida* is a large climbing or trailing herb with stout hispid stems. Fruits are 30 to 45 cm long broadly, cylindric, not ribbed hairy, ultimately covered with a waxy bloom. Fruit is covered in a fuzzy coating of fine hairs when young. The immature melon has thick white flesh that tastes sweet. By maturity, the fruit loses its hairs and develops a waxy coating, giving rise to the name wax gourd. The fruit may grow as large as 80 cm in length. It has yellow flowers and broad leaves.

Physicochemical parameter studies on selected plants: Total ash of *Cucurbita pepo* fruit was found 6.4%. Water soluble ash was found 2.91 % whereas 0.91% was acid insoluble ash. The ethanol soluble extractive values were found to be 9.6% and water-soluble extractive values were found to be 13.8 %. The moisture content of the powder estimated as percentage loss on drying (LOD) was found to be 29.2 % w/w. Total ash of *Benincasa hispida* fruit was found 5.7%. Water soluble ash was found 2.68 % whereas 0.98% was acid insoluble ash. The ethanol soluble extractive values were found to be 8.3 % and water-soluble extractive values were found to be 10.9% and loss on drying (LOD) was found to be 27.86% w/w.

Extraction of the Drugs: The coarse powder of the fruit of *Cucurbita pepo* and *Benincasa hispida* were subjected to successive solvent extraction using solvents of ascending polarity. After extraction the percentage yield of each extract was calculated with reference to the air dried drug used for the study. The percentage yield and other characteristic features of the extracts are tabulated below.

Preliminary Phytochemical Screening: The qualitative phytochemical screening of the

tubers for the presence of alkaloids, carbohydrate, reducing sugars, glycosides like anthraquinones, flavanoids, saponins, tannins, phenolic compounds, fixed oils, fats, proteins, amino acids and sterols in petroleum ether, ethyl acetate and ethanol extracts of the fruit of *Cucurbita pepo* and *Benincasa hispida* were carried out.

Analysis of the free radical scavenging activities of the selected *Cucurbita pepo* fruit and *Benincasa hispida* fruits extracts revealed a concentration dependent free radical scavenging activity resulting from reduction of DPPH, NO, Hydroxyl radical and superoxide radical to non-radical form. The scavenging activity of Ascorbic acid, a known antioxidant used as positive control, was however higher. DPPH radical is considered to be a model for a lipophilic radical. The scavenging effect of l-Ascorbic acid, and plant extracts increased gradually with increase in concentration. In case of *Cucurbita pepo* extracts the order of reduction potential was: Ascorbic acid > PECP > EACP > EOCP. Even in case of *Benincasa hispida* the reduction potential was in the order as: Ascorbic acid > PEBH > EABH > EOBH

Nitric oxide plays an important role in various types of inflammatory processes in the body. In the present study the fruit extracts of selected *Cucurbita pepo* and *Benincasa hispida* checked for its inhibitory effect on Nitric oxide production. Results revealed that all the tested extracts showed the percentage of inhibition in a dose dependent manner. The ethanol extract of *Benincasa hispida* at varied concentrations showed remarkable inhibitory effect of nitric oxide radical scavenging activity compared to other extract. The ethanol extract of *Cucurbita pepo* at varied concentrations showed remarkable inhibitory effect of nitric oxide radical scavenging activity compared to other extract. The ethanol extract of *Cucurbita pepo* showed more activity than ethanol extract of *Benincasa hispida*.

The hydroxyl radical is an extremely reactive free radical formed in biological systems and has been implicated as a highly damaging species in free radical pathology, capable of damaging almost every molecule found in living cells. The effect of the selected plant extracts were assessed by means of the iron (II)- dependent DNA damage assay. All the results showed hydroxyl radical scavenging activity in a dose dependent manner.

The ethanol extract of *Benincasa hispida* at varied concentrations showed remarkable

inhibitory effect of Hydroxyl radical scavenging activity compared to petroleum ether and ethyl acetate extract. The ethanol extract of cucurbita pepo at varied concentrations showed remarkable inhibitory effect of Hydroxyl radical scavenging activity compared to petroleum ether and ethyl acetate extract. The ethanol extract of cucurbita pepo showed more inhibitory effect of Hydroxyl radical scavenging activity compared ethanol extract of Benincasa hispida.

Superoxide is a reactive oxygen species, which can cause damage to the cells and DNA leading to various diseases. The ethanol extract of Benincasa hispida at varied concentrations showed remarkable inhibitory effect of superoxide radical activity scavenging compared to petroleum ether and ethyl acetate extract. The ethanol extract of cucurbita pepo at varied concentrations showed remarkable inhibitory effect of superoxide radical scavenging activity compared to petroleum ether and ethyl acetate extract. The ethanol extract of cucurbita pepo showed more inhibitory effect of superoxide radical scavenging activity compared ethanol extract of Benincasa hispida.

Summary and Conclusion

From this work we conclude that all the extracts were exhibiting significant scavenging activity towards 1, 1-di phenyl picryl hydrazyl, Nitric oxide, Hydroxyl, Super oxide radicals. The activity was found to be concentration dependent. In DPPH model the free radical scavenging capacity was found to be highly significant when compare other three models. In all the three selected plants Ethanol extract was found to have high scavenging activity than Ethyl acetate and petroleum ether extracts. Scavenging activity of ethanol extracts may be due to presence of the flavonoids and phenolic.

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Table 1: Organoleptic and Physicochemical parameters of *Cucurbita pepo* and *Benincasa hispida* fruit

S. No.	Parameters	<i>Cucurbita pepo</i>	<i>Benincasa hispida</i>
1	Shape and size	Fruits are ovoid ribbed size 13-45cm long covered with hard peel	Fruits are 30 to 50 cm long broadly, cylindric, hairy, covered with a waxy bloom

2	Colour	outer (greenish yellow) inner (yellow)	outer (greenish waxy) inner (white)
3	Odour	Characteristics	Characteristics
4	Taste	Sweet	Sweet
5	Total ash	6.4 % w/w	5.7 % w/w
6	Water soluble ash	2.91 % w/w	2.68 % w/w
7	Acid insoluble ash	0.91 % w/w	0.98 % w/w
8	Foreign organic matter determination	1.05 % w/w	0.98 % w/w
9	Ethanol soluble extractive	9.6 % w/w	8.3 % w/w
10	Water-soluble extractive	13.8 % w/w	10.9 % w/w
11	Loss on drying (%)	29.2 % w/w	27.86 % w/w

Table 2: Phytochemical analysis parameters of *Cucurbita pepo* and *Benincasa hispida* fruit

Tests of Phytoconstituents	<i>Cucurbita pepo</i> fruit extracts	<i>Benincasa hispida fruit extract</i>	<i>Cucurbita pepo</i> fruit extracts	<i>Benincasa hispida fruit extract</i>	<i>Cucurbita pepo</i> fruit extracts	<i>Benincasa hispida fruit extract</i>
	Petroleum ether extract		Ethyl acetate extract		Ethanol extract	
Extractive value	6.5	5.2	9.7	8.5	18.3	15.8
1. Alkaloids						
a) Mayer's reagent	-	-	-	-	+	+

b) Dragendorff's reagent	-	-	-	-	+	+
2. Flavonoids						
a) Shinoda test	-	-	+	+	+	+
3. Saponins						
a) Froth test	-	-	-	-	+	+
4. Carbohydrate						
a) Molisch's test	-	-	+	+	+	+
c) Test for gums	-	-	+	+	+	+
d) Test for mucilage	-	-	+	+	+	+
5. Phytosterols						
a) Libermann-	+	+	-	-	-	-

Burchard test						
c) Salkowski reaction:	+	+	-	-	-	-
6. Tannins and Phenolic						
a) With Lead acetate	-	-	+	+	+	+
7. Cardiac glycoside						
a) (a) Borntrager's test	-	-	+	+	+	+
b) Legal's test	-	-	+	+	+	+
8. Coumarins						
a) With ammonia	-	-			-	-
b) Hydroxylamine HCl	-	-			-	-
9. Proteins						
a) Biuret test	-	-	+	+	+	-
10. Triterpens						
a) Vanillin sulphuric acid	+	+			-	-

Anti-oxidant Activity:

Table 3: Cucurbita pepo and Benincasa hispida fruit on DPPH radical scavenging

Concentration	100µg/ml	200 µg/ml	300 µg/ml	400 µg/ml	500 µg/ml
Ascorbic acid	74.12	77.14	82.14	85.41	96.14
PECP	12.45	18.47	29.45	36.74	48.54
EACP	30.14	34.87	40.28	52.47	62.87

EOCP	64.87	74.87	78.25	80.98	84.65
PEBH	11.27	17.87	26.57	34.16	46.87
EABH	29.78	33.74	39.47	50.87	61.87
EOBH	62.47	71.14	75.87	78.14	82.59

Table 4: *Cucurbita pepo* and *Benincasa hispida* fruit on Nitric oxide radical scavenging

Concentration	100µg/ml	200 µg/ml	300 µg/ml	400 µg/ml	500 µg/ml
Ascorbic acid	76.24	80.14	84.98	90.25	97.54
PECP	12.54	25.64	38.74	41.54	54.87
EACP	52.68	55.74	58.74	65.45	71.05
EOCP	60.87	67.87	71.25	74.65	80.25
PEBH	9.87	21.54	35.41	37.84	51.42
EABH	50.75	53.12	56.18	62.18	68.36
EOBH	58.47	65.14	69.18	72.58	78.94

Table 5: *Cucurbita pepo* and *Benincasa hispida* fruit on Hydroxyl radical scavenging

Concentration	100µg/ml	200 µg/ml	300 µg/ml	400 µg/ml	500 µg/ml
Ascorbic acid	60.25	65.47	70.28	75.15	91.36
PECP	14.78	20.54	24.63	40.28	54.24
EACP	26.48	38.34	50.41	57.26	62.37
EOCP	54.38	60.26	65.38	69.24	75.26
PEBH	12.54	18.62	23.74	37.41	51.64

EABH	25.15	36.15	48.31	55.36	60.87
EOBH	50.27	57.15	61.84	65.63	71.24

Table 6: *Cucurbita pepo* and *Benincasa hispida* fruit on Super oxide radical scavenging activity

Concentration	100µg/ml	200 µg/ml	300 µg/ml	400 µg/ml	500 µg/ml
Ascorbic acid	35.25	45.35	54.17	62.87	70.84
PECP	20.68	24.74	39.54	50.47	55.31
EACP	18.98	32.54	35.45	49.75	60.38
EOCP	33.14	36.57	49.54	62.14	65.87
PEBH	18.31	22.23	36.41	47.24	52.63
EABH	16.26	30.31	32.45	47.51	58.24
EOBH	31.87	34.98	47.54	60.45	62.87

Table 7: *In vitro* 50% inhibition concentration (IC₅₀) of *Cucurbita pepo* fruit and *Benincasa hispida* fruit extracts on various models

Extract /compound	50% inhibition concentration (IC ₅₀) of DPPH model (µg/ml)	50% inhibition concentration (IC ₅₀) NO model (µg/ml)	50% inhibition concentration (IC ₅₀) Hydroxyl radical model (µg/ml)	50% inhibition concentration (IC ₅₀) superoxide model (µg/ml)
Ascorbic acid	66	67	82	160
PECP	500	492	503	499
EACP	398	100.2	398	509
EOCP	80.32	85	102	396

PEBH	508	495	508	504
EABH	406	102.54	403	513
EOBH	85.64	88	107	399



Figure 1: A. Cucurbita pepo

B. Benincasa hispida fruit

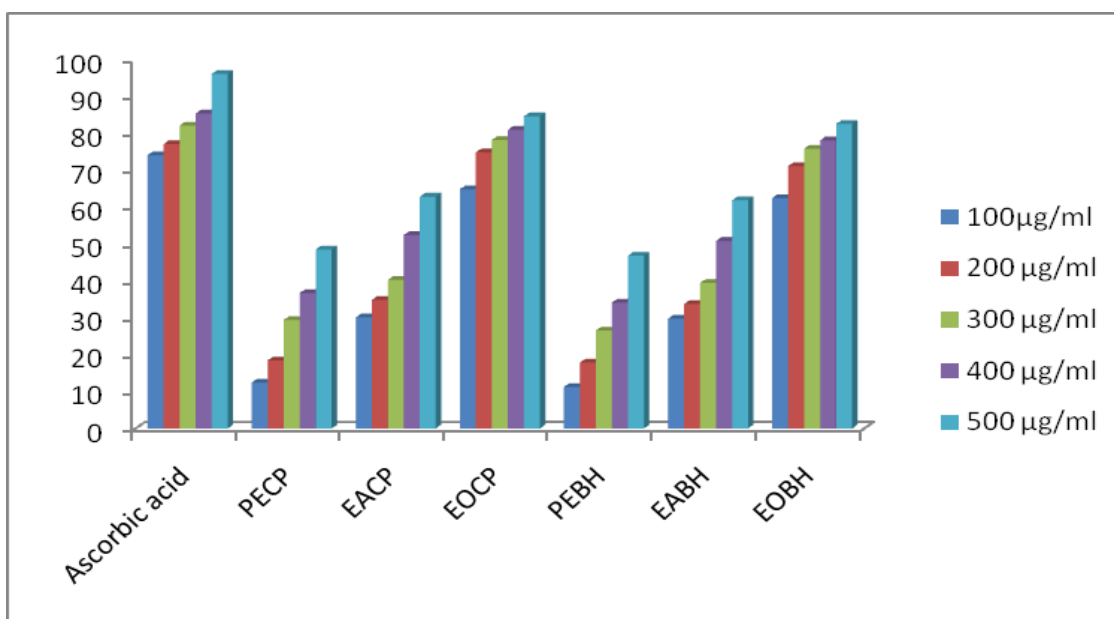


Figure 2: Graphical representation of Cucurbita pepo and Benincasa hispida fruit extracts on DPPH model

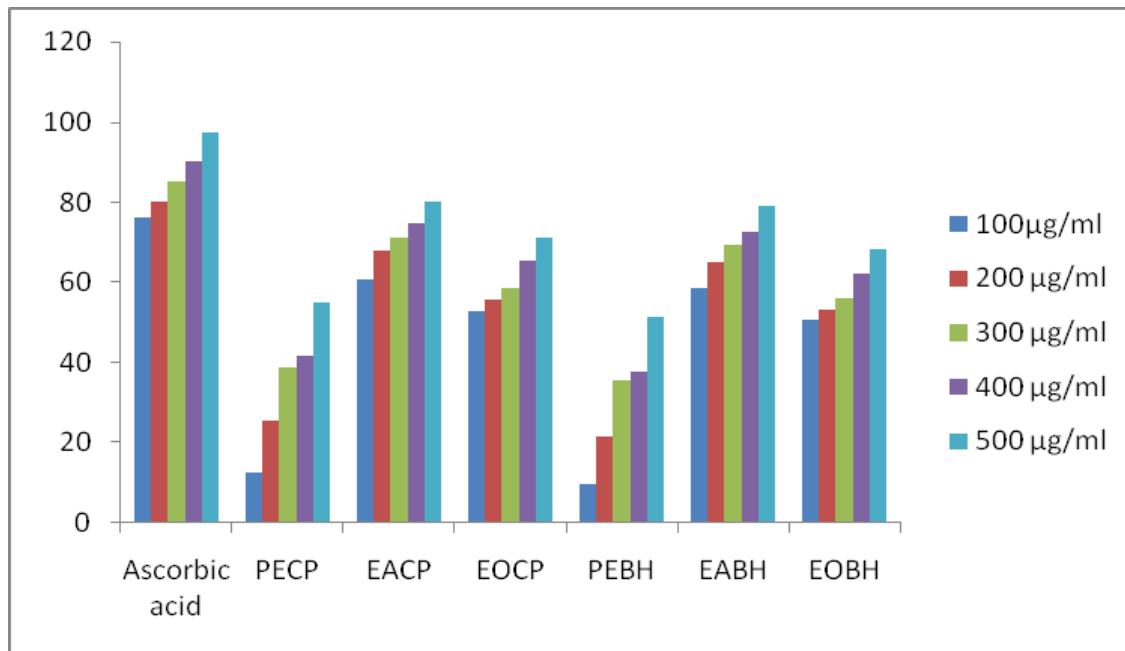


Figure 3: Graphical representation of Cucurbita pepo and Benincasa hispida fruit extracts on Nitric oxide radical scavenging model

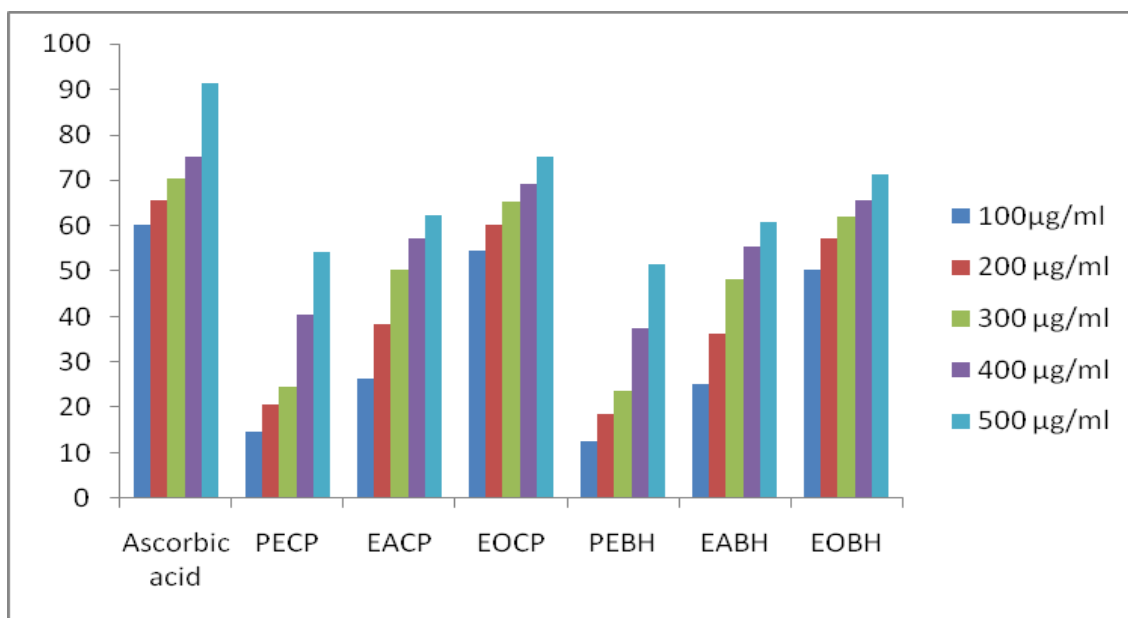


Figure 4: Graphical representation of Cucurbita pepo and Benincasa hispida fruit extracts on Hydroxyl radical scavenging model

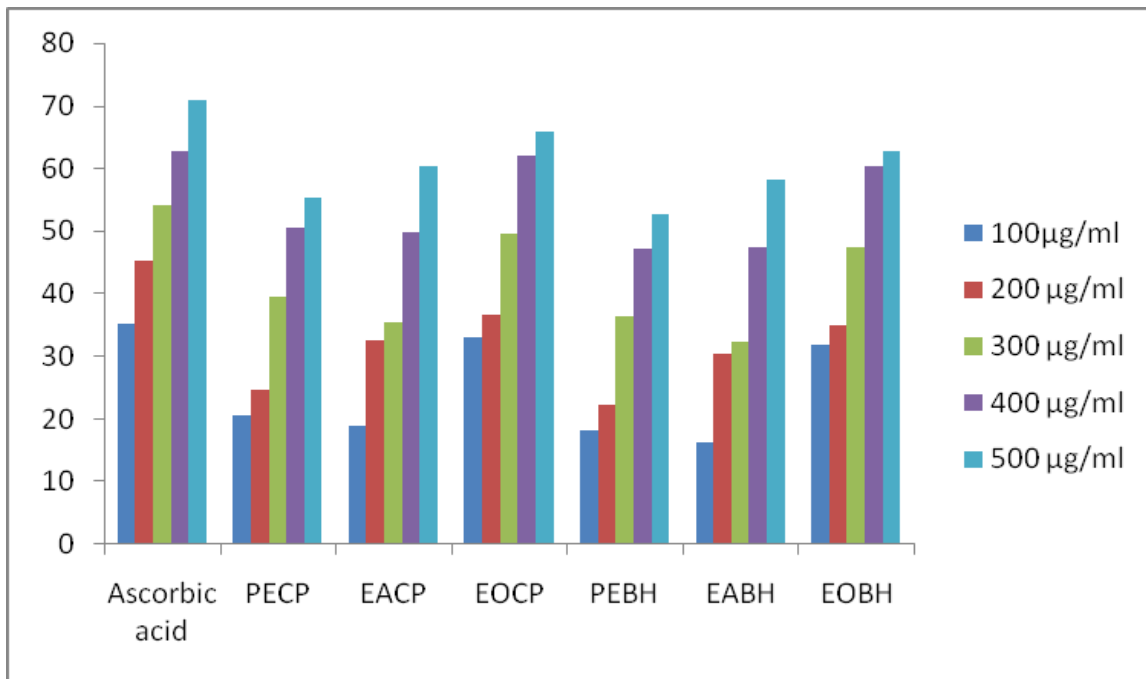


Figure 5: Graphical representation of Cucurbita pepo and Benincasa hispida fruit extracts on Super oxide radical scavenging model

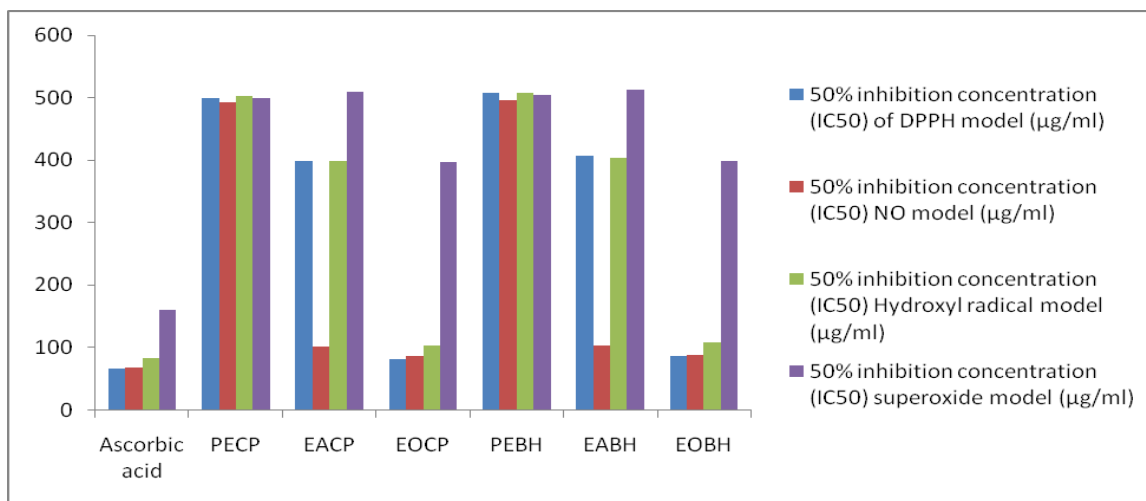


Figure 6: Graphical representation of 50%Inhibitory Concentration (IC50) Benincasa hispida fruits extracts and Ascorbic acid on various models