IJPPR
INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals
ISSN 2349-7203
HUMAN

Human Journals
**Research Article**
June 2021 Vol.:21, Issue:3
© All rights are reserved by Basavaraj Chivde et al.

# Comparative Study of Total Phenolic and Flavonoid Content and Antioxidant Potential of Various Parts *Benincasa hispida*

IJPPR
INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals
ISSN 2349-7203
HUMAN

**Basavaraj Chivde*[1], Prasanna Sagar[2], S. B. Puranik[3]**

[1]*Research scholar OPJS University, Churu, Rajasthan, India*

[2]*Research Guide OPJS University, Churu, Rajasthan, India*

[3]*Drishti Institute of distance learning, Bangalore, India*

**Submitted:** 22 May 2021
**Accepted:** 29 May 2021
**Published:** 30 June 2021

HUMAN JOURNALS
www.ijppr.humanjournals.com

**Keywords:** Phenolic, flavonoid, antioxidant, *Benincasa hispida*, seed

**ABSTRACT**

In the present investigation, the total phenolic, flavonoid content and antioxidant potential of various part of *Benincasa hispida* were evaluated. The antioxidant potential of different extracts (ethyl acetate, ethanol and aqueous) of leaves, fruit and seeds were measured by three different scavenging assays such as DPPH, Superoxide, Nitric oxide, hydrogen peroxide and Hydroxyl radical. For total phenolic content, Folin-Ciocalteau assay was performed and determination of flavonoids was performed according to the colorimetric assay. Among all parts studied, the seed extract of *Benincasa hispida* reported the highest antioxidant capacity for scavenging activity, and also exhibited highest total phenolic and flavonoid content as compared to leaves and fruit. Hence definite and positive correlations were exhibited for parts (leaves, fruit and seed) between total phenolic and flavonoid content along with radical scavenging activity. It can be concluded that different parts of *Benincasa hispida* possess good antioxidant activity which can be utilized for the treatment of various oxidative stress disorders.

## INTRODUCTION

*Benincasa hispida* (Wax gourd) is an important medicinal plant known for its medicinal and nutritional properties. It is a tendril climber cultivated throughout India. This plant grows annually. It has been used as a food and medicine for thousands of years in East Asia. All parts of the plant are used medicinally. This plant is creeping with branched tendrils that can climb over with the help of some support, cover fences or sprawl along the ground. Stems are thick, hairy, grooved conspicuously and lined with sharp bristles. Leaves are round, kidney-shaped with upper rough surface. These have beautiful flower of golden yellow color. Fruits contain numerous white-colored embedded seeds[1]. Fruits are traditionally used to treat Cardiotonic, renal diseases, epilepsy, jaundice, dyspepsia, fever and menstrual disorders.

In ayurvedic medicines, seeds are used in the treatment of antihelmintic, anti-inflammatory demulcent, diuretic, expectorant, laxative and tonic. Fruit are diuretic, laxative, tonic. It is used in ayurvedic medicine in the treatment of epilepsy, lung disease, asthma and cough. Fruit juice is used in the treatment of insanity, epilepsy and another nervous disease. This plant has been used in the form of taila, rasayana in the siddha system of medicines. Phytochemical constituents of this plant are mannitol, triterpenoids, cucurbitin, β-sitosterin, flavonoids, glycosides, carotenes, vitamins and uronic acid[2].

Antioxidants are an important factor to maintain optimal cellular and human body health. Antioxidant compounds are gaining in importance due to their dual role in food and pharmaceutical industries as lipid stabilizers[3].

The importance of the reactive oxygen species (ROS) has attracted increasing attention over the last decade. ROS includes free radicals such as superoxide anion radicals ($O_2^-$), hydroxyl radicals ($OH^-$), hydrogen peroxide ($H_2O_2$) and singlet oxygen (O2) along with various forms of activated oxygen[4-5]. They are involved in various physicochemical processes and diseases such as aging[6], cancer[7], atherosclerosis[8] etc. Many plants contain substantial amounts of antioxidants including vitamin C and E, carotenoids, flavonoids, polyphenols, tannins and thus can be utilized to scavenge the excess of free radicals from the human body.

Hence, owing to the strong potential of the plant and no reported data on a comparative study of different parts of *Benincasa hispida* for its antioxidant potential, the present study was performed to estimate the total phenols and flavonoids contents and antioxidant potential of various parts of *Benincasa hispida.*

## MATERIALS AND METHODS

### Collection of plant materials

Plant specimen for the present study was collected from a medicinal plant vendor. Care was taken to select healthy plant materials (Leaves, fruit and seeds). The fruit was peeled off and seeds were removed. The fruit were taken and cut into pieces, dried and then ground to powder form. Seeds were separated, raised using tap water and dried in oven at $38^0$C for 24 h. The prepared seeds were stored in a dark place at ambient temperature. The seed powder was produced using grinder mill.

### Extraction

The 100 g of each powdered material was extracted with ethyl acetate, ethanol and water using soxhlet apparatus. The extract was stored in a glass bottle in refrigerated condition throughout the experiment.

### Phytochemical analysis (qualitative)

Phytochemical analysis ethyl acetate, ethanol and aqueous extract was performed to identify the constituents using standard procedures as described by Sofowara, Trease and Harborne[9-11].

### Estimation of Total phenol content

Total phenolic content was estimated using the Folin-Ciocalteu method of Yu et al.[12]. Extract (100µL) was mixed thoroughly with 2 ml of 2% Na2CO3. After 2 minutes 100µl of Folin-Ciocalteu reagent was added to the mixture. The resulting mixture was allowed to stand at room temperature for 30 min and the absorbance was measured at 743 nm against the blank. A calibration curve was established using varying concentrations of gallic acid. The values were expressed in mg/g of sample.

### Estimation of Total Flavonoid content

The determination of flavonoids was performed according to the colorimetric assay of Chang et al.[13]**.** To 1ml of extract, 3 ml of methanol, 0.2 ml of 1 M potassium acetate, 0.2 ml of 10% aluminium chloride and 5.6 ml of distilled water was added and left at room temperature for 30 minutes. The absorbance of the mixture was read at 415 nm using UV spectrophotometer. The calibration curve was prepared using quercetin as standard.

**Determination of Total Antioxidant Capacity**

The assay is based on the reduction of molybdenum (VI) to molybdenum (V) by the extract and the subsequent formation of a green phosphate Mo (V) complex at acid pH Preito et al.[14]. An aliquot of sample solution (100 µg/ml was combined with reagent solution (0.6 M Sulfuric acid, 28mM Sodium Phosphate and 4mM Ammonium molybdate). The tubes were capped and incubated in a boiling water bath at 950C for 60-90 min. The samples were then cooled at room temperature and the absorbance was measured at 695 nm against the blank in a spectrophotometer. The values were expressed as equivalents of BHT.

**Evaluation of Anti-oxidant activity by radical scavenging assay**

**DPPH radical scavenging activity**

Free radical scavenging capacity of various parts of *Benincasa hispida* was determined using DPPH[15]. DPPH radical scavenging activity was done by serial dilution by taking diluted methanol (1:20) as standard. 10 ml of various diluted extracts of various concentrations (100, 200 and 400 µg/ml) were added to 1 ml DPPH solution (0.004%) and incubated for 10 min at room temperature. The absorbance of test and reference standard, ascorbic acid was measured at 517 nm. The amount of DPPH scavenging was calculated by using the formula:

% DPPH radical scavenging = [(Absorbance of control – Absorbance of test sample)/ (Absorbance of control)] × 100

***Superoxide radical scavenging activity***

The superoxide radical scavenging activity was measured by the following described method[16-18]. 1 ml of each extract of various parts of *Benincasa hispida* at various concentrations (100, 200 and 400 µg/ml) were mixed with 1 ml of nitro blue tetrazolium (NBT) solution (156 mM NBT in phosphate buffer of pH 7.4) and 1 ml NADH in phosphate buffer of pH 7.4. The reaction was initiated by adding 100 ml of phenazine methosulfate (PMS) solution (60 mM PMS in phosphate buffer, pH 7.4) to the mixture. The reaction mixture was incubated at $25^0$C for 5 min and the absorbance was measured at 560 nm against blank sample and compared with reference standard ascorbic acid. Decreased absorbance of reaction mixture indicated increased superoxide anion scavenging activity. The percentage of inhibition of superoxide anion generation was calculated using the following formula:

% inhibition = [(Absorbance of control – Absorbance of test sample)/ (Absorbance of control)] × 100

***Nitric oxide scavenging activity***

Nitric oxide scavenging activity was determined by the following method described[19]. Briefly, 5 mM sodium nitroprusside was prepared in phosphate-buffered saline and mixed with different concentrations of extracts of various parts of *Benincasa hispida* at (100, 200 and 400 µg/ml) followed by incubation at $25^0$C for 30 min. A control without the extracts but with equivalent amounts of methanol was taken. After 30 min of incubation, 1.5 ml of solution was pipetted out and diluted with 1.5 ml of Griess reagent. Absorbance of chromophore formed during diazotization of the nitrite with sulphanilamide and subsequent coupling with N-1- naphthyl ethylenediamine dihydrochloride was measured at 546 nm and percentage scavenging activity was measured with reference standard ascorbic acid.

***Scavenging of hydrogen peroxide***

Scavenging of hydrogen peroxide was measured by the following described method[20]. A solution of hydrogen peroxide (40 mM) was prepared in phosphate buffer (pH 7.4). 1 ml of each extract of various parts of *Benincasa hispida* at different concentrations (100, 200 and 400 µg/ml) were added to 0.6 ml of 40 mM hydrogen peroxide solution. Absorbance of hydrogen peroxide at 230 nm was determined after 10 min against a blank containing phosphate buffer without hydrogen peroxide. The percentage scavenging of hydrogen peroxide of plant extract and reference standard ascorbic acid was calculated using the following formula:

% scavenged [$H_2O_2$] = [(Absorbance of control – Absorbance of test sample)/ (Absorbance ofcontrol)] × 100

***Hydroxyl radical activity***

Hydroxyl radical activity was measured by the following described method[21]. 1 ml of each extract of various parts of *Benincasa hispida* at various concentrations (100, 200 and 400 µg/ml) were placed in tubes and evaporated to dryness. 1 ml of ferrous- EDTA (0.13% ferrous ammonium sulfate and 0.26% EDTA), 0.5ml of 0.018% EDTA, 1 ml of DMSO (0.85% v/v in 0.1 M phosphate buffer, pH 7.4) and 0.5 ml of freshly prepared 0.22% ascorbic acid were added to each tube. The tubes were capped tightly and heated in a water bath at 80 -

$90^0$C for 15 min. The reaction was terminated by addition of 1 ml ice cold TCA (17.5% w/v) and followed by addition of 3 ml of Nash reagent and left at room temperature for 15 min. The intensity of color formed was measured at 412 nm against the reagent blank. The percentage inhibition was compared with a reference standard ascorbic acid and test compounds.

## STATISTICAL ANALYSIS

The data from the experiments were presented as mean ± S.E.M (n=3). Student's *t*-test was used for statistical analysis. Values were considered statistically significant when P<0.5.

## RESULTS AND DISCUSSION

Reactive oxygen species [ROS] have been implicated in several diseases like cancer, diabetes, atherosclerosis and cardiovascular disease. Synthetic antioxidants such as butylated hydroxytoluene (BHT) and buytlated hydroxyanisole (BHA) which are commonly used in processed foods, possessed a number of side effects which makes limited their use as antioxidant agent. In the present study the antioxidant potential of various parts of *Benincasa hispida* was performed in search of potential antioxidant agent from natural source[22-23].

### Phytochemical analysis (qualitative)

Phytochemical analysis of *Benincasa hispida* shows the presence of flavonoids, Saponins, terpenoids, phenolic compounds and alkaloids (Table No 1).

### Total Phenol Content

The Phenolic contents present in extracts of various parts of *Benincasa hispida* were presented in Table 2. Seed shows promising results wherein the ethyl acetate shows 28.34±0.67mg/g, ethanol has 96.40±0.96 mg/g and aqueous extract shows 81.48±0.94 mg/g of phenolic content in seeds. The ethanolic extracts of leaves, fruit and seed have noticed higher percentage of total phenolic compounds at all tested concentrations.

### Total Flavonoid Content

The Flavonoids contents present in extracts of various parts of *Benincasa hispida* were presented in Table 2. Similarly, as like phenol content, seed also shows higher concentration of flavonoid content. Ethyl acetate extract of seeds shows 17.76±1.28 mg/g, Ethanol extract shows 73.81±0.67 mg/g and aqueous extract shows 68.65±0.47 mg/g.

**The Total Antioxidant Capacity**

The total antioxidant capacity was presented in Table 2. The total antioxidant capacity was measured as equivalent of BHT (mg/g of extract). The Ethyl acetate extract of seed has 124±1.68, Ethanol extract 144.16±1.27 mg/g and aqueous extract 115.78±1.63 mg/g.

**DPPH Radical Scavenging Activity**

Among the extracts tested the highest antioxidant activity was exhibited by fruit and seed extracts. In the present investigation, the ethanolic extracts of fruit and seed extract were notably, exhibited scavenging activity compared to other extracts. The values were obtained with fruit and seed are 0.40±0.11 and 0.45±0.15 respectively which were significant to $p<0.5$ and are noticed at 400 µg/ml concentration of extract (Table No 3).

**Super Oxide Radical Scavenging Activity**

The ethanolic fruit and seed extracts reported significant scavenging activity than other extracts. The values of fruit and seed extracts which are obtained at 400 µg/ml are 0.47±0.26 and .51±0.28 respectively, which were comparable to the reference standard (Table.3).

**Nitric Oxide Scavenging Activity**

Among the extracts tested fruit and seed extracts produced satisfactory results. The values of fruit and seed extracts which were obtained at 400 µg/ml concentration are 0.28±0.16 and 0.54±0.29 respectively (Table.3).

**Scavenging of Hydrogen Peroxide**

Interestingly, all the plants notably exhibited scavenging activity. However, seed is proved to possess the highest scavenging activity than others. The value of seed extract obtained at 400 µg/ml is 0.43±0.21 (Table No 3).

**Hydroxyl Radical Activity**

The ethanolic extracts of seed, hold more scavenging activity compared to other extracts. The values which were noticed at 400 µg/ml concentration of fruit and seed extracts are 0.52±0.31 and 0.55±0.30, respectively (Table No 3).

## CONCLUSION

In the present investigation, the Phytochemical analysis of *Benincasa hispida* shows the presence of flavonoids, Saponins, terpenoids, phenolic compounds and alkaloids which may act as better antioxidants and have beneficial effects on many pathological conditions. It was observed that all the extracts showed concentration-dependent scavenging activity. The ethanolic seed extract of *Benincasa hispida* was reported to possess significant antioxidant activity and higher content of phenolic and flavonoids. From these findings, it can be suggested that the seeds of this plant is having an immense source of natural antioxidants and could play an important role as therapeutic agents to prevent oxidative stress and related disorders.

## REFERENCES

1. Washimkar VB, Shende MB. Kushmanda with brief uses and its Ingredients. Eur J Pharm Med Res. 2016; 3(9):158-162.
2. Ghosh K, Baghel MS. Pharmacognostical and Physicochemical Study of *Benincasa hispida* with Ayurvedic Review. Int J Res Ayur & Pharm. 2011;2(6):1664-1668.
3. Ramadan MF, Morsel JT. Screening of the antiradical action of vegetable oils. J Food Compos Anal. 2006; 19: 838-842.
4. Rangari VD. Pharmacognosy and Phytochemistry. Part-II. 1st Ed. Career Publications:Nashik. 2003.
5. Blois MS. Antioxidant determinations by the use of a stable free radical. Nature. 1998; 29:1199-1200.
6. Finkol T, Holbnok NJ. Oxidant, oxidative stress and the biology of aging. Nature. 2000; 239-247.
7. Senthil K, Aranganathur S, Nalio N. Evidence of oxidative stress in the circulation of ovarian cancer patients. Clin Chim Acta. 2004; 3(39): 27-32.
8. Upston JM, Kritharides L, Stocker R. The role of vitamin-E in atherosclerosis. Prog Lipid. Res. 2003; 42:405-422.
9. Sofowara A. Medicinal plants and Tradional medicine in Africa spectrum Books Ltd. 1993.
10. Trease GE, Evans WC. Pharmacognosy. 11th Ed. Brailliae Tiridal Can. Macmillan Publisher's: 1989.
11. Harborn JB. Phytochemical methods: A guide to modern techniques of plant analysis. 1973.
12. Yu L, Haley S, Perret J, Harris M, Wilson J and Qian M. Free radical scavenging properties of wheat extracts. J Agri Food Chem. 2002;50:1619-1624.
13. Chang C, Yang M, Wen HJ. Estimation of total flavonoid content in propolis by two complementary colorimetric methods. Food Drug Analysis. 2002; 10:178-182.
14. Preito P, Pinedo M and Aguilar M. Spectrophotometric quantitation of antioxidant capacity through the formation of a phosphomolybdenum complex: specific application to the determination of Vitamin E. Anal Biochem. 1999; 269:337-341.
15. Naznin A, Hasan N. *In vitro* Antioxidant Activity of Methanolic Leaves and fowers Extracts of *Lippia Alba*. Res J Medand Medical Sci. 2009;: 4(1): 107-110.
16. Nishikimi M, Rao NA, Yagi K. The occurrence of superoxide anion in the reaction of reduced phenazine methosulfate and molecular oxygen. Biochem Biophys Res Commun. 1972; 46(2):849-854.
17. Yen GC, Duh PD. Scavenging effect of methanolic extracts of Peanut hulls on free radical and Active-oxygen species. J Agric. Food Chem. 1994; 42(3): 629–632.
18. Murali KT, Vadluri R, Manoj Kumar E. *In Vitro* Determination of Antioxidant Activity of *Physalis angulata* Lnn. Int J Pharm Bio Sci. 2013; 4(3): 541-549.
19. Govindarajan R, Rastogi S, Vijaykumar M. Studies on antioxidant activities of *Desmodium gangeticum*. Pharm. Bull. 2003; 26(10): 1424-1427.

20. Ilhami G, Haci AA, Mehmet C, Determination of *in vitro* antioxidant and radical scavenging activities of propofol. Chem Pharm Bull. 2005; 53(3): 281-285.
21. Klein SM, Cohen G, Cederbaumm AI. Production of formaldehyde during metabolism of dimethyl sulfoxide by hydroxyl radical generating systems. Biochemistry, 1981; 20(21): 6006-6012.
22. Siddiqua A, Premakumari KB, Sultana R, Vithya and Savitha. Antioxidant Activity and Estimation of Total Phenolic Content of *Muntingia calabura* by Colorimetry. Int.J. Chem Tech Res. 2010; 2(1): 205-208.
23. Chavan C, Mulik S, Chavan M, Adnaik RS. Screening of Antioxidant Activity and Phenolic Content of Whole Plant of *Barleria prionitis* Linn. Int J Res Ayur & Pharm. 2011; 4: 1313-1319.

**Table No. 1: Qualitative Phytochemical examination of various parts of *Benincasa hispida***

| S. No | Compounds | LEAVES | | | FRUIT | | | SEED | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | Ethyl acetate extract | Ethanolic extract | Aqueous extract | Ethyl acetate extract | Ethanolic extract | Aqueous extract | Ethyl acetate extract | Ethanolic extract | Aqueous extract |
| 1. | Flavonoids | + | +++ | ++ | + | +++ | ++ | + | +++ | +++ |
| 2. | Terpenoids | + | +++ | ++ | + | ++ | +++ | + | +++ | ++ |
| 3. | Tannins | - | ++ | + | - | ++ | - | - | ++ | + |
| 4. | Phenols | ++ | +++ | ++ | ++ | +++ | ++ + | ++ | +++ | ++ + |
| 5. | Cardiac glycosides | - | + | + | - | + | + | - | + | + |
| 6. | Amino acids | ++ | +++ | + | ++ | +++ | + | ++ | +++ | + |
| 7. | Reducing sugar | + | +++ | ++ | + | +++ | ++ | + | +++ | ++ |
| 8. | Sterols | ++ | +++ | +++ | ++ | +++ | +++ | ++ | +++ | +++ |
| 9. | Phytosterols | + | +++ | +++ | + | +++ | +++ | + | +++ | +++ |
| 10. | Saponin | ++ | +++ | +++ | ++ | +++ | +++ | ++ | +++ | +++ |
| 11. | Carbohydrate | +++ | ++ | ++ | ++ | +++ | + | + | ++ | + |
| 12. | Coumarin | - | ++ | +++ | - | ++ | +++ | - | ++ | +++ |
| 13. | Alkaloids | + | ++ | ++ | + | ++ | ++ | + | ++ | ++ |

**Legends: +++ -Highly present; ++ -Moderately present; + -Present; --Absent**

**Table No. 2: Total phenol, Flavonoid and Antioxidant level of *Benincasa hispida***

| S. No. | Parameter | Concentration mg/g of extract | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | LEAVES | | | FRUIT | | | SEED | | |
| | | Ethyl acetate | Ethanol | Aqueous | Ethyl acetate | Ethanol | Aqueous | Ethyl acetate | Ethanol | Aqueous |
| 1 | Total Phenol (gallic acid/gm of extract) | 21.66±1.79 | 89.78±0.91 | 81.68±0.94 | 23.78±1.66 | 92.75±0.74 | 80.14±0.24 | 28.34±0.67 | 96.40±0.96 | 81.48±0.94 |
| 2 | Total Flavonoid (Quercetin/ gm of extract) | 14.27±1.38 | 70.67±0.88 | 68.34±0.92 | 16.34±1.28 | 71.37±0.28 | 64.65±0.34 | 17.76±1.28 | 73.81±0.67 | 68.65±0.47 |
| 3 | Total Antioxidant (BHT/gm of extract) | 121±1.28 | 142.16±1.27 | 114.33±1.62 | 122±1.78 | 142.78±0.47 | 115.28±1.28 | 124±1.68 | 144.16±1.27 | 115.78±1.63 |

**Table No. 3** ***Antioxidant activity of various parts of Benincasa hispida***

| | DPPH | | | Super Oxide | | | Nitric Oxide | | | H2O2 | | | Hydroxyl Radical | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | *100* | *200* | *400* | *100* | *200* | *400* | *100* | *200* | *400* | *100* | *200* | *400* | *100* | *200* | *400* |
| | ***Ethyl acetate extract*** | | | | | | | | | | | | | | |
| ***Leaves*** | ***0.12***±0.14 | ***0.20***± 0.16 | ***0.25***± 0.21* | 0.14 ± 0.21* | 0.22 ± 0.14* | 0.24 ± 0.11* | 0.11 ± 0.1* | 0.18 ± 0.05* | 0.22 ± 0.12 | 0.11 ± 0.05* | 0.18 ± 0.11 * | 0.20 ± 0.18* | 0.05 ± 0.13* | 0.14 ± 0.14 | 0.23 ± 0.16* |
| ***Fruits*** | ***0.20***±0.21 | ***0.25***± 0.24 | ***0.32***± 0.22* | 0.22 ± 0.16* | 0.26 ± 0.14* | 0.34 ± 0.22* | 0.22 ± 0.12* | 0.26 ± 0.18* | 0.29 ± 0.21* | 0.14 ± 0.06* | 0.17 ± 0.10 * | 0.26 ± 0.12* | 0.11 ± 0.12* | 0.21 ± 0.2* | 0.27 ± 0.20* |
| ***Seeds*** | ***0.24***±0.11 | ***0.26***± 0.14 | ***0.30***± 0.21* | 0.28 ± 0.22 | 0.30 ± 0.27* | 0.32 ± 0.29* | 0.14 ± 0.15* | 0.23 ± 0.11* | 0.28 ± 0.16* | 0.21 ± 0.16* | 0.26 ± 0.17 * | 0.28 ± 0.25* | 0.16 ± 0.21* | 0.28 ± 0.22* | 0.31 ± 0.27* |
| | ***Ethanol extract*** | | | | | | | | | | | | | | |
| ***Leaves*** | ***0.15***±0.23 | ***0.24***± 0.15 | ***0.32***± 0.23* | 0.20 ± 0.21* | 0.24 ± 0.14* | 0.3 ± 0.18* | 0.17 ± 0.1* | 0.10 ± 0.05* | 0.20 ± 0.12 | 0.15 ± 0.03* | 0.18 * ± 0.005* | 0.21 ± 0.08* | 0.19 ± 0.22* | 0.27 ± 0.14* | 0.36 ± 0.20* |
| ***Fruits*** | ***0.28***±0.12 | ***0.30***± 0.14* | ***0.40***± 0.11* | 0.41 ± 0.21 | 0.45 ± 0.24* | 0.47 ± 0.26* | 0.20 ± 0.12* | 0.26 ± 0.15* | 0.28 ± 0.16* | 0.16 ± 0.06* | 0.22 ± 0.1* | 0.34 ± 0.12* | 0.36 ± 0.27* | 0.41 ± 0.24* | 0.52 ± 0.31* |
| ***Seeds*** | ***0.25***±0.11 | ***0.32***± 0.05* | ***0.45***± 0.15* | 0.36 ± 0.21* | 0.46 ± 0.20* | 0.51 ± 0.28* | 0.35 ± 0.20* | 0.48 ± 0.26* | 0.54 ± 0.29* | 0.20 ± 0.16* | 0.34 ± 0.19 * | 0.43 ± 0.21* | 0.28 ± 0.21* | 0.37 ± 0.28* | 0.55 ± 0.30* |
| | ***Aqueous extract*** | | | | | | | | | | | | | | |
| ***Leaves*** | ***0.22***±0.21 | ***0.28***± 0.16 | ***0.30***± 0.28 | 0.22 ± 0.18* | 0.29 ± 0.14* | 0.34 ± 0.21* | 0.12 ± 0.05* | 0.22 ± 0.11* | 0.26 ± 0.14* | 0.16 ± 0.06* | 0.21 ± 0.10 * | 0.27 ± 0.12* | 0.17 ± 0.14* | 0.24 ± 0.16* | 0.34 ± 0.17* |
| ***Fruits*** | ***0.25***±0.06 | ***0.32***± 0.14* | ***0.38***± 0.17 | 0.36 ± 0.21* | 0.42 ± 0.29* | 0.49 ± 0.29* | 0.20 ± 0.15* | 0.30 ± 0.18* | 0.32 ± 0.16* | 0.18 ± 0.06* | 0.28 ± 0.1* | 0.37 ± 0.21* | 0.21 ± 0.28* | 0.38 ± 0.27* | 0.48 ± 0.29* |
| ***Seeds*** | ***0.28***±0.12 | ***0.30***± 0.16 | ***0.38***± 0.18 | 0.34 ± 0.16* | 0.48 ± 0.21* | 0.54 ± 0.27* | 0.24 ± 0.12 | 0.26 ± 0.16* | 0.34 ± 0.2* | 0.20 ± 0.16* | 0.32 ± 0.19 * | 0.40 ± 0.26* | 0.28 ± 0.22* | 0.31 ± 0.29* | 0.51± 0.32* |

***Values are mean±S.E.M. n=3***

****significance is set at p>0.5.***

***# Conc µg/ml***