International Journal of Chemical Studies

P-ISSN: 2349–8528
E-ISSN: 2321–4902
IJCS 2019; 7(5): 271-275

Received: 06-07-2019
Accepted: 10-08-2019

Priyanka HL
Department of Horticulture, College of Horticulture UHS Campus, GKVK, Bengaluru, Karnataka, India

Sakthivel T
Indian Institute of Horticulture Research, Bengaluru, Karnataka, India

Shivashankar KS
Indian Institute of Horticulture Research, Bengaluru, Karnataka, India

Dinesh MR
Indian Institute of Horticulture Research, Bengaluru, Karnataka, India

Vinay GM
Department of Horticulture, College of Horticulture UHS Campus, GKVK, Bengaluru, Karnataka, India

Correspondence
Priyanka HL
Department of Horticulture, College of Horticulture UHS Campus, GKVK, Bengaluru, Karnataka, India

Biochemical characterization of *Annona squamosa* varieties and half sib progenies

Priyanka HL, Sakthivel T, Shivashankar KS, Dinesh MR and Vinay GM

Abstract
Annona squamosa Linn, commonly known as Sugar apple, belonging to the family Annonaceae, is said to show varied medicinal effects. The plant is traditionally used for the treatment of epilepsy, dysentery, cardiac problems, worm infestation, constipation, hemorrhage, antibacterial infection, fever, ulcer and also has insecticidal properties. Hence, the present investigation was carried out to determine the chemical composition and also to determine the superior physico chemical properties of the selected *Annona squamosa* varieties and half sib progenies for selection and further breeding purposes at ICAR-Indian Institute of Horticultural Research, Hesaraghatta Lake Post, Bengaluru. The biochemical analysis of *Annona squamosa* varieties and half sib progenies pulp extract revealed the highest values with respect to TSS (31.44^0B), acidity of pulp (0.425%), total sugars (24.52g/100g) and reducing sugars (21.19g/100g) was recorded in hybrid Arka Sahan, TSS: Acidity ratio in accession 3/2 (211.31), ascorbic acid content (13.667% to 34.775%), non-reducing sugars content was recorded in the accession 8/3 (5.37g/100g). The total phenol content ranged between 57.7 and 81.76mg GAE/100g, total flavonoid between 9.94 and 20.44mg Catechin equivalent/100g and antioxidant capacity between 62.03 and 89.64 mg AEAC/100g, justifying the use of this plant to treat many ailments in folk and herbal medicine and also for the selection purpose.

Keywords: *Annona squamosa* varieties and half sib progenies, biochemical composition.

Introduction
The genus *Annona* is the most economically important among the Annonaceous family, containing 120 species. Among the edible *Annona*s, the cherimoya (*Annona cherimola*), sweetsop/sugar apple/custard apple (*Annona squamosa*), and *Annona atemoya* (a hybrid of *Annona cherimola* and *Annona squamosa*) are commercially significant and economically important fruits in several tropical and sub-tropical countries. Other species of *Annona*ceae are Ramphal (*Annona reticulata*), Soursop (*Annona muricata*) and Pond apple (*Annona glabra*).
Annona squamosa is native to Tropical America but its exact native range is unknown. The sugar apple species name 'squamosa' refers to the knobby appearance of the fruit. Sugar apple is a small tropical tree originating in the New World tropics, probably Central America. It is one of the major Annonaceous fruits grown commercially in India with an area of 44,000 hectares with a production of 3,67,000MT (Anon, 2017) [3]. The plant parts also contain numerous amounts of bioactive chemical substances such as acetogenins, alkaloids, terpens, flavonoids, cyclopeptide, annomuricatin and oils (Pinto *et al*., 2005) [14]. These compounds are very useful medicines because some acetogenins have anti-tumoral, antifertility, abortifacient, insecticidal, antibacterial, immuno-suppressant, pesticidal or antihelminthic properties. The plant is traditionally used for the treatment of epilepsy, dysentery, cardiac problems, worm infestation, constipation, hemorrhage, antibacterial infection, dysuria, fever, and ulcer. Ethanolic extracts of leaves and stem are reported to have an anticancerous activity. *A. squamosa* has phytopharmacological properties (Saleem *et al*., 2009) [16].
Nutritional and commercial potential knowledge of the native species could be an economic alternative to the livelihood of native population from different regions (Silva *et al*., 2013) [17]. There is an old saying "Eat your food like your medicines or else you will eat your medicines like food". Although, there is a crescent increase in studies with native fruits as well as the development of new food products based on them, the information about the nutritional potential of the Annonaceous fruits are limited or, often, do not exist, occurring a lack on scientific investments in this area (Souza *et al.*, 2012) [20]. thus the current investigation was taken up to assess the biochemical profiling of selected *Annona squamosa*

varieties and half sib progenies.

Materials and Method

The experiments were carried out at ICAR-Indian Institute of Horticultural Research, Hesaraghatta Lake Post, Bengaluru during 2015-2017. The fruits were harvested at full maturity and kept in laboratory for 2 days for ripening. Fresh pulp from ripe fruits was used for the biochemical analysis. The experiment was done in a completely randomized design using three replications and data generated through experiments were statistically analyzed by using the OPSTAT (Gomez and Gomez, 1983) [7]. The list of ten varieties of *Annona squamosa* L. and 25 half sib progenies of Balanagar are given in table 1.

Table 1: List of Ten varieties of *Annona squamosa* L. and 25 half sib progenies of Balanagar selected for the biochemical profiling

1.	Arka Sahan	11.	1/1	21.	4/1	31.	8/3
2.	Balanagar	12.	1/5	22.	4/2	32.	8/4
3.	Barbados	13.	1/10	23.	4/10	33.	8/7
4.	Mammoth	14.	2/1	24.	4/11	34.	8/8
5.	Red Sitapal	15.	2/2	25.	4/12	35.	8/11
6.	Washington 98797	16.	2/10	26.	5/1		
7.	Washington 107005	17.	2/13	27.	5/8		
8.	APK 1	18.	3/1	28.	6/1		
9.	Taiwan	19.	3/2	29.	6/18		
10.	Raidurg	20.	3/3	30.	7/1		

Total soluble solids [° Brix] of fruit pulp of all the genotype was recorded by using a digital Refractometer (Digital refractometer, DBX-55).

Titrable acidity (%): The acidity of the 10 g pulp samples was determined by diluting an aliquot of the sample with distilled water and titrating with 0.1N NaOH using phenolphthalein as an indicator. The end point appeared as light-pink colour. The calculated acidity was expressed as per cent anhydrous citric acid (Ranganna, 1986) [15]. The TSS: acid ratio was calculated by dividing the TSS by titrable acidity.

Ascorbic acid content was determined by 2, 6-Dichlorophenol indophenol (DCPIP) method (AOAC, 967.21) (Association of Official Analytical Chemists, 2006). Ten grams of fruit pulp was mixed thoroughly with 4% oxalic acid solution, squeezed through a muslin cloth and volume was made up to 50 ml. Vitamin C content present in the solution was estimated by titrating a known quantity of the extract against DCPIP. The end point is the appearance of pink colour. Vitamin C content was calculated as mg of ascorbic acid equivalents per 100 g fresh weight using a standard curve of L-Ascorbic acid.

Total Sugars, Reducing and non-reducing sugars by Nelson-Somogyi Method (Somogyi, 1952 and Krishnaveni *et al* 1984) [19, 10]. The extract was taken and titrated against 10ml of mixed Fehling solution A and B using methylene blue as indicator. The results were ex-pressed as percent of reducing sugar. The sugar extract was hydrolyzed with concentrated hydrochloric acid and titrated against 10 ml of mixed Fehling's solution (5 ml Fehling A + 5 ml Fehling solution B) using methylene blue as indicator. Results were expressed as per cent total sugar. The amount of non-reducing sugar was calculated by subtracting reducing sugars from total sugar and multiplying the difference by factor 0.95 as suggested by AOAC (1980).

Antioxidant activity (Diphenyl-1-picryl hydrazyl radical scavenging ability (DPPH): Extract was prepared by taking sample (5g) with 50 ml of 80% methanol. 0.2 ml of extract was taken in test tube, 0.3 ml of acetate buffer was added followed by 2.5 ml of DPPH solution and mixed. The absorbance of the solution was read spectro photometrically at 517 nm after 30 minutes of incubation (A1). The absorbance of DPPH solution without sample (A2) was taken. The difference in the absorbance of DPPH solution with and without sample (A2 – A1) was calculated and the decrease in absorbance with sample addition was used for calculation of antioxidant activity. A standard curve was developed using different concentrations of ascorbic acid (20-100 µg/ml). The results were expressed as ascorbic acid equivalent antioxidant capacity (Kang *et al*. 2002). the difference in absorbance of DPPH solution with and without ascorbic acid (b1-a1) was calculated and Divided the concentration of the ascorbic acid by the difference in absorbance to arrive at the amount of ascorbic acid per unit absorbance (µg/OD) - (a). The antioxidant concentration in the sample extract (µg/ml) was calculated by multiplying the absorbance of sample with (a) - (b).

Total phenols: The analysis of total phenols was carried out by Folin-Ciocalteu spectrophotometric method suggested by Singleton and Rossi (1965) [18]. Gallic acid equivalence method was used for determining the phenol content in the fruit juice. Total phenolics content was expressed as Gallic acid equivalents (GAE) in mg per 100g fresh weight of pulp

Total flavonoids: 5g of sample was Homogenized with 20 ml of methanol (80%) in a pestle and mortar 2-3 times. the extract was Pooled and the volume was made up to 50 ml. 1.0 ml of extract was Taken in tubes and 0.3 ml of 5% $NaNO_2$ was added. after 2 min 0.3 ml of 10% $AlCl_3$ was added. After another 2 min, 3.4 ml of NaOH was added and allowed to stand at room temperature for 10 minutes. The absorbance was Read at 510 nm against blank. Catechin was used as standard (Chun *et al*., 2003).

Results and Discussion

A considerable variation was observed in some of the physico-chemical and antioxidant properties of studied selected *Annona squamosa* varieties and half sib progenies. Some of the physical characteristics of the pomegranate fruits are presented in Table 2.

Table 2: Biochemical characters of different Varieties and accessions of *Annona squamosa*

Varieties and accessions	TSS (^{0}B)	Titrable acidity (%)	TSS: Acidity	Ascorbic acid (mg/100g)	Total sugars (g/100g)	Reducing sugars (g/100g)	Non reducing sugars (g/100g)	Total Phenols (mgGAE/ 100g)	Total Flavanoids (mg Catechin equivalent /100g)	DPPH (mgAEAC/ 100g)
Arka Sahan	31.44	0.43	78.43	30.12	24.53	21.19	3.34	68.89	13.55	73.71
Balanagar	24.98	0.19	132.30	24.93	20.58	17.39	3.19	74.95	20.44	87.69
Barbados	22.07	0.25	88.27	34.78	17.52	14.78	2.74	64.01	13.35	67.21
Mammoth	24.87	0.26	95.91	27.18	19.78	16.23	3.55	64.28	14.77	64.28
Red Sitapal	23.70	0.14	176.21	22.67	18.65	14.75	3.90	73.01	7.84	81.05
Washington 98797	24.50	0.28	89.32	18.78	16.55	12.84	3.72	58.52	11.78	62.03
Washington 107005	24.27	0.23	104.20	26.95	19.24	14.87	4.37	63.56	10.88	73.09
APK 1	24.97	0.20	127.03	21.33	19.57	15.31	4.26	73.54	13.14	79.42
Taiwan	24.74	0.22	114.86	23.71	18.94	13.91	5.03	68.64	18.96	77.56
Raidurg	22.67	0.20	120.56	26.62	18.08	13.35	4.72	73.02	20.71	84.70
1/1	24.57	0.17	144.41	16.67	19.57	14.80	4.78	70.49	19.11	78.25
1/5	24.90	0.17	149.20	25.45	19.45	15.00	4.45	71.33	19.32	77.75
1/10	27.10	0.14	189.73	26.33	18.27	14.25	4.02	70.10	12.76	75.01
2/1	23.20	0.38	62.53	25.68	18.21	14.22	3.98	76.49	13.32	84.91
2/2	24.33	0.14	187.44	22.67	19.18	14.75	4.44	61.84	11.34	72.36
2/10	23.20	0.24	96.00	24.79	17.30	13.83	3.48	63.42	11.90	76.10
2/13	26.40	0.24	109.31	13.67	20.05	15.59	4.46	64.52	12.89	70.33
3/1	23.40	0.15	168.62	19.67	18.08	14.40	3.68	121.09	16.24	133.20
3/2	25.47	0.12	211.31	16.87	19.56	14.73	4.83	118.99	16.65	140.40
3/3	24.80	0.14	183.69	18.99	19.29	14.13	5.16	104.90	16.39	119.59
4/1	24.57	0.15	201.47	25.22	19.56	15.22	4.34	75.07	15.39	83.32
4/2	24.37	0.14	175.91	17.67	19.37	15.37	4.00	75.79	14.90	90.19
4/10	24.80	0.21	122.94	25.14	15.70	12.42	3.28	77.46	21.76	84.44
4/11	24.13	0.13	182.83	24.17	17.37	12.93	4.44	79.41	11.05	88.94
4/12	21.83	0.37	167.16	16.99	13.68	10.48	3.20	67.70	20.36	68.38
5/1	21.60	0.15	148.05	26.42	15.98	11.26	4.71	65.00	10.73	76.70
5/8	22.83	0.11	216.22	24.33	16.67	12.25	4.42	57.70	12.21	66.35
6/1	24.47	0.16	157.28	23.24	19.52	14.59	4.94	66.68	16.15	75.35
6/18	23.40	0.14	163.87	22.33	18.15	13.40	4.75	67.82	17.00	81.39
7/1	22.63	0.11	199.83	13.67	17.23	12.22	5.01	73.12	15.82	78.96
8/3	22.83	0.14	164.24	18.64	17.24	11.88	5.37	66.73	12.80	71.74
8/4	22.17	0.16	141.98	15.67	15.75	11.95	3.80	66.90	13.71	76.26
8/7	23.63	0.17	136.45	16.99	15.98	12.05	3.93	69.26	14.25	74.11
8/8	23.43	0.15	160.50	18.09	17.49	13.05	4.44	66.20	11.32	78.78
8/11	23.73	0.14	169.81	23.44	17.38	12.20	5.18	62.43	12.59	71.79
CD @ 5%	1.82	0.14	60.57	4.43	2.46	3.02	0.64	5.04	3.36	22.87
SEm	0.64	0.05	21.43	1.56	0.87	1.07	0.23	1.78	1.19	8.09

When evaluating a fruit for consumer acceptance a breeder is not concerned with soluble solids alone but with perceived sweetness, which is determined largely by the relative levels of total soluble solids and acids in the fruits. The total soluble solids are composed of all the soluble solids which are present in the fruits. In desert fruit like custard apple, the fruits having perfect sugar-acid blend are preferred by consumers. Different kinds of organic acids and the extent of their concentration play an important role in the flavor of a fruit. Usually high acidity gives better blend and flavor.

The average TSS content for different *Annona squamosa* varieties and half sib progenies, highest TSS (31.44 ^{0}B) and acidity of pulp (0.43%) was recorded for hybrid Arka Sahan whereas lower was recorded for accession 5/1. The variation in the values of TSS may be due to various factors like climatic condition and genetic characters of the genotype. The similar results were reported by in annona. Jalikop and Kumar (2000) [9] reported the similar results that in Arka Sahan, interspecific hybrid the TSS was more than 30 ^{0}B. Mathakar (2005) [12] in *Annona squamosa* L. Because acidity in fruits plays an important role in taste, color, and microbial stability of the fruit juice and determines maturity it can be concluded that *Annona* fruits can have better acceptability for the consumers.

For perfect blend, sugar and acid ratio is one of the important parameter which determines the taste of fruit. The TSS and acidity of fruit both have contribution towards fruit taste and flavour. The highest TSS: Acidity ratio was observed in accession 3/2 (211.31 ^{0}B). Anon (2006) [1] reported variation for TSS, acidity and ascorbic acid in seventeen accessions of custard apple germplasm in Tamil Nadu.

Average Ascorbic acid content for different accessions of *Annona squamosa* was observed in the range of 13.667% to 34.775%. The variation in the values of ascorbic acid may be due to various factors like climatic condition and genetic characters of the genotype. Pareek *et al.* (2011) [13] also claimed 11.5 ± 5.5, 30.0, 29.4 ± 3 and 37.38 ± 4.62mg of ascorbic acid per 100g of pulp of cherimola, custard apple, sour sop and sugar apple, respectively. Boake *et al.* (2014) [6] obtained 20.33 and 63.67 mg ascorbic acid per 100 g of sweetsop and soursop fruits, respectively. The high values of ascorbic acid in *Annona* signify the potential use of the fruit as a good source of ascorbic acid. The recommended daily intake (RDI) of ascorbic acid is about 30 mg/day for adults

and 17 mg/day for children. Therefore, these fruits could be considered as good sources of ascorbic acid for purposes of human nutrition.

Sugars are the primary products of photosynthesis and perform multiple roles in plants as energy and carbon transport molecules, hormone like signalling factors, from which plants make proteins, polysaccharides, oils and woody materials. It is also responsible for resistance mechanism against biotic stresses. Sugars play important role in osmotic adjustment and in providing protection against various types of stresses. Sugar content of fruit is the only factor which determines the sweetness of pulp. The highest total sugars (24.53 g/100g) and reducing sugars content (21.19 g/100g) was recorded in the Arka Sahan while the highest non reducing sugars content was recorded in the accession 8/3 (5.37 g/100g).

Jalikop and Kumar (2000) [9] observed a total sugar in Arka Sahan was 22.80 per cent. Mathakar (2005) [12] observed the total sugar content varied from 14.75 to 22.88 percent in *Annona* hybrids. The total sugar content varied from 21.02 to 24.88 percent in cultivars of custard apple (Anon., 2007) [2]. Vinay *et al*. (2016) reported that Arka Sahan had highest total sugars and reducing sugars. The higher concentration of reducing sugars indicating that the hybrid has more sweetness and lower concentration indicating that lower sweetness. Benkeblia *et al.* (2014) [5] also reported similar result of 11.98 to 17.88 mg reducing sugar. Jadhav (2008) [8] reported that non-reducing sugars varied from 1.50 to 2.95 percent in Annona hybrids. The non-reducing sugars percentage of cultivars of custard apple under Rahuri conditions ranged from 2.17 to 3.50 per cent (Anon., 2007) [2].

Significantly higher total phenol was recorded accession 3/1 (121.09 mgGAE/100g) and the lowest was observed in 5/8 (57.70 mgGAE/100g). Higher phenol content can be a potential source of disease resistance and also for therapeutic purpose in pharmaceuticals. Flavonoids are a group of compounds contributing to the total antioxidant capacity of fruits and vegetables. Flavonoid content was found in a very less quantity. Significantly the highest total flavonoid was recorded in variety Balanagar (20.44 mg Catechin equivalent/100g) and the lowest was observed in Red Sitaphal (7.84 mg Catechin equivalent/100g). The antioxidant activity is an essential biological property of great interest because it reduces the toxic effects of oxidants and thus prevents the cellular damage caused by free radicals. The total antioxidant capacity was the highest antioxidant capacity was recorded in accession 3/2 (140.40 mg AEAC/100g) and the lowest was observed in variety Washington 98797 (62.03 mg AEAC/100g).

In addition to the antioxidant capacity, phenolics can influence the flavour determining fruit astringency and bitterness (Silva *et al*. 2013 and Anuragi *et al*., 2017) [17, 4]. Study of Benkeblia *et al.* (2014) [5] also revealed 9.83 to 18.71 mg phenols per gram of fruit pulp of custard apple (*A. reticulata*). Mariod *et al.* (2010) [11] and Vijayaraghavan *et al*. (2013) reported 7.81 to 125.0 µg/ml antioxidant activities using DPPH assay in *A. squamosa* extracts.

Conclusion

From the study it can be revealed that different selected *Annona squamosa* varieties and half sib progenies can act as important source of compounds with a high potential to protect health. *Annona* species possessed relatively higher amount of antioxidants, were sweet enough and also possessed higher quantity of antioxidants. Thus, it can be suggested that the fruits of these species can be good source of natural antioxidants.

This study showed considerable variation in some of the bio chemical properties of annona cultivars. The variation could originate from the annona cultivar and agro-climatic as well as environmental conditions. This study provides important data for compositional information of the fruits (e.g. reducing sugars, vitamin C, titrable acidity, antioxidant activity and etc.), emphasizing that annona fruit can be a good source of nutrients.

In conclusion, a comparison of the results obtained by us with those found in other studies reveals that annona fruit contains important amounts of antioxidant and high amount of nutrients that play a valuable role in people's daily diet. The variability in the biochemical characteristics is mostly useful for improving the quality of Annona and thus helpful for the exploitation of heterosis.

Acknowledgements

Author is grateful to the Dean and campus Head, College of Horticulture Bangalore, My research topic advisory committee members and Indian Institute of Horticulture Research Bangalore-India for providing financial support and various laboratory facilities.

References

1. Anonymous. Biennial Report (2004-2005). XIII Group Workers Meeting. AICRP on Arid Zone Fruits, Cen. Inst. Arid Hort., Bikaner (Rajasthan), India, 2006.
2. Anonymous. A report on characterization and documentation of custard apple germplasm. AICRP on Arid Zone Fruits, Department of Horticulture, Mahatma Phule Krishi Vidyapeeth, Rahuri. (Maharashtra), India, 2007.
3. Anonymous. Horticulture at a glance, 2017, http://nhb.gov.in/statistics/Publication/Horticulture%20At%20a%20Glance%202017%20for%20net%20uplod%20(2).pdf
4. Anuragi H, Jain BT, Dhaduk HL, Kumar S. Genetic Association Studies for Fruit Yield and its Components and Qualitative Phytochemical Screening in Promising *Annona* Genotypes. Res. J. of Agric. Sci. 2017; 8(1):222-227.
5. Benkebliaa N. Emanuel MA, Richards RM. Variation of total and reducing sugars, total phenols and total anthocyanins in custard apple (*Annona reticulata*) during three "On tree" ripening stages. Acta Hort. 2014; 1047:147-152.
6. Boake AA, Faustina D, Jacob K, Ibok O. Dietary fibre, ascorbic acid and proximate composition of tropical underutilized fruits. Afr. J. Food Sci. 2014; 8(6):305-310.
7. Gomez KA, Gomez AA. Statistical procedures for agricultural research. Int. Rice Res. Inst., Philippines, 1983.
8. Jadhav SB. Evaluation of Annona (*Annona spp.*) hybrids. Ph.D. (Hort.) Thesis, Mahatma Phule Krishi Vidyapeeth, Rahuri, India, 2008.
9. Jalikop SH, Kumar SP. New fruit varieties for arid regions. Pomegranate 'Ruby' and custard apple 'Arka Sahan'. Indian Hort. 2000; 45(2):19-20.
10. Krishnaveni S, Balasubramanian T, Sadasivam S. Sugar distribution in sweet stalk sorghum. Food Chem. 1984; 15:229.
11. Mariod AA, Elkheir S, Ahmed YM, Matthaus B. *Annona squamosa* and *Catunaregam nilotica* seeds, the effect of

the extraction method on the oil composition. J. Am. Oil Chem. Soc. 2010; 87:763-769.
12. Mathakar TD. Assessment of custard apple (*Annona squamosa* L.) hybrids. M.Sc. (Agri.) thesis, Mahatma Phule Krishi Vidyapeeth, Rahuri (Maharashtra), India, 2005.
13. Pareek S, Yahia EM, Pareek OP, Kaushik RA. Postharvest physiology and technology of *Annona* fruits. Food Res. Int. 2011; 44:1741-1751.
14. Pinto AC, De Q, Cordeiro MCR, Andrade SRM, De Ferreira FR, Filgueiras HA *et al*. Annona species. Int. Cen. Underutilized Crops, UK, 2005.
15. Ranganna S. Manual of analysis of fruit and vegetable product. Tata McGraw Hill Publishing Co. Ltd., New Delhi, 1986.
16. Saleem TSM, Hema Basnett, Ravi V, Bhupendra Shrestha, Verma NK, Patel SS, *et al*. Phyto-pharmacological review of *Annona squamosa* Linn. Natural Products. 2009; 5(2):85-88.
17. Silva EP, Vilas Boas EVB, Xisto ALP. Characterization and development of marolo (*Annona crassiflora*, Mart.). Ciência e Tecnologia de Alimentos. 2013; 33(4):1-10.
18. Singleton VL, Rossi JA. Colorimetry of total phenolics with phosphomolybdic phosphotungstic acid reagents. Am. J. Enol. Viticult. 1965; 16:144-158.
19. Somogyi M. Estimation of sugars by colorimetric method. J. Biol. Chem. 1952; 200:245.
20. Souza PM, Elias ST, Imenoni LA, Paula JE, Gomes SM, Guerra ENS *et al*. Plants from Brazilian cerrado with potent tyrosinase inhibitory activity. Plos One. 2012; 7(11):1-7.
21. Vinay GM, Praneeth YS, Chithiraichelvan R, Upreti KK, Venugopalan R. Biochemical changes during off-season flowering in custard apple (*Annona squamosa* L.) cv. Arka Sahan induced by pruning and defoliation. Inter. J. Agri. Sci. 2016; 8(59):3314-3317.