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Biochemical composition of broccoli cultivars at harvest and during post-harvest storage

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Abstract

The fresh edible heads of four broccoli cultivars grown in randomized block design with six replications at the Vegetable Science research farm of the university were evaluated for different biochemical constituents of nutritional quality significance at harvest and post-harvest stages. Significant variations in moisture, protein, total lipids, carbohydrates, total chlorophyll, total carotenoids, ascorbic acid, total phenols, glucosinolates, calcium, iron, sodium, potassium and phosphorus contents were observed to range from 89.56 to 91.24 %, 3.16 to 3.86%, 243.3 to 288.3 mg/100g , 5.4 to 5.6%, 0.13 to 0.42 mg/g, 0.36 to 1.40mg/g, 58.80 to 81.31mg/100g, 0.87 to 1.18%, 264.1 to 344.2 mg/100g, 54 to 79.8 mg/100g, 0.90 to 1.61mg/100g, 23 to 26 mg/ 100g, 122.8 to 324.6 mg/ 100g and 52.00 to 79.00mg/100g , respectively at harvest stage (DAS=129). At post harvest storage stage(s) viz. I, II and III each at interval of two days after harvest stage , various quality parameters were observed to vary significantly from 87.78 to 90.02%, 3.02 to 3.82%, 228.6 to 283.3 mg/100g, 4.8 to 5.4 %, 0.04 to 0.38 mg/g, 0.29 to 1.38 mg/g, 47.24 to 78.45 mg/100g, 1.16 to 1.49%, 250.1 to 340.5 mg/100g, 53.30 to 79.50 mg/100g, 0.9 to 1.6 mg/100g, 22.16 to 26.0 mg/100g, 122.0 to 324.5 mg/100g and 52.0 to 79.0 mg/100g, in that order. On the basis of genotypic ranking, the cultivar Palam Vichitra followed by Palam Samridhi emerged out as versatile cultivar(s) at harvest stage when the performance of various cultivars was compared at harvest and post harvest storage intervals.

Key words: Cultivars, broccoli, ascorbic acid, carbohydrates, glucosinolates, phenols.

Introduction

Broccoli is believed to be the first of the Cole crops to evolve from the wild species of Kale or cabbage and was cultivated by the Romans. It was introduced in England in the early 16th century known as 'Italian asparagus' or 'sprout cauliflower' (Bill Hankin and Debb Schmetzer, 2001). In India, broccoli has established amongst amateur gardeners and commercial production have also picked up near metropolises. In the state of Himachal Pradesh systematic breeding programme was initiated in the Department of Vegetable Science, CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur more than a decade ago, which has resulted into release of improved indigenous variety of green sprouting broccoli 'Palam Samridhi'. Furthermore, marginal hill farmers with small holdings are benefited from sprouting type genotype as two to three harvesting can be taken from these genotypes (Gautam, 2001). Broccoli is a cool weather crop that does poorly in hot summer weather. It is high in vitamin C and dietary fiber and contains multiple nutrients with potent anti-cancer properties due to presence of secondary metabolites called glucosinolates. Broccoli leaves are also edible and contain more ascorbic acid and β- carotene exhibiting a positive effect on visual activity contained in these vegetables (Biermann Medical News, 2000). Literature revealed wide variations in the biochemical constituents of freshly harvested heads of broccoli genotypes (Kalia, 1995 and Dogra and Awasthi, 2009).

However, there appears still inadequate information systematically about the status of dietary nutrients including secondary metabolites viz., phenols and glucosinolates in broccoli cultivars particularly during post-harvest storage conditions. As such, an attempt was made to evaluate various biochemical constituents both in the fresh edible heads as well as at post- harvest storage stages of promising broccoli cultivars commonly grown in northern India.

Materials and Methods

The edible heads of four broccoli cultivars; Palam Samridhi, Palam Vichitra, Palam Kanchan, Palam Haritika were procured from the research farm of the Department of Vegetable Science of the University grown in randomized block design with six replications following the recommended package of cultural practices. The broccoli heads were plucked from randomly selected plants at the stage of marketable

maturity, called harvest stage (DAS=129). After harvest, the heads were kept under normal room temperature during the month of January and analysed at interval of two days for one week, called as post-harvest storage stage(s) –I, II and III. The heads of each plant were plucked, pooled and used for biochemical estimations by following standard procedures. Moisture, protein and ascorbic acid were estimated by the procedure given in AOAC (1970). Carbohydrate content was estimated by method followed by Yemm *et al.*, (1954), whereas total lipids were determined by the procedure of Folch *et al.* (1957). Colorimetric estimation of total chlorophyll and carotenoids was carried out by adopting the method of Singh (1977) and Davies (1976), respectively. Fresh heads of broccoli were used for glucosinolates determination by following the procedure given by Tholen *et al.* (1989). The dried, ground samples were used for total phenols estimation as described by Bray and Thorpe (1954). The powdered samples were digested in triacid mixture (nitric acid, percholric acid and sulphuric acid in 10:2:1 ratio) for the analysis of various minerals in the digested samples. Calcium, potassium, sodium and iron were estimated using flame photometer (model Systronics-3491), atomic absorption spectrophotometer (model AA-137) as described in A.O.A.C. (1975). Phosphorus content was determined colorimetrically by adopting the procedure of Jackson (1967). The entire data was analyzed statistically by using analysis of variance as described by Panse and Shukhatme (1984).

Results and Discussion

The data presented in Table 1 revealed wide variations in the biochemical constituents of edible heads of broccoli cultivars at harvest and post harvest storage stages- I, II and III, respectively.

The status of moisture content roughly indicates the degree of maturity and accumulation of fibre in vegetables. It is an important criterion contributing towards consumers' acceptability of the crop harvest including vegetables. It is evident that moisture content varied from 89.56 to 91.24, 88.70 to 90.02, 88.18 to 89.81 and 88.17 to 88.44 per cent at harvest and post- harvest storage stages- I, II and III, accordingly. The loss in moisture content might be attributed to respiration process during storage after harvest as evident from the loss of moisture content in the broccoli heads uniformly at post harvest storage stages over harvest stage. It is worth while to indicate that among various cultivars Palam Kanchan exhibited the highest value consistently for moisture content both at harvest and all the post-harvest stages. The range of variations in moisture content in the present study are corroborated with those reported by other workers to range from 87.85 to 89.9 per cent in moisture content in this crop (Kalia, 1995).

Proteins are of vital importance to living organisms as these constitute an important component of tissues as well as the body fluids imparting structural, regulatory and immune functions. The protein content of different cultivars was observed to vary significantly at harvest stage from 3.16 to 3.86 *per cent.* The highest value was showed by Palam Haritika accompanied by Palam Vichitra, Palam Samridhi and Palam Kanchan in that order. However, on storage at room temperature a decreasing trend in protein content was noticed to vary from 3.12 to 3.82, 3.08 to 3.77, 3.02 to 3.75 per cent at post-harvest storage stages- I, II and III, respectively. Nevertheless, altogether the highest value was showed by Palam Kanchan in all the stages of post-harvest storage, whereas the maximum protein content was exhibited by Palam Haritika at post harvest storage stages - I, II and Palam Samridhi merely at post harvest stages- III on this aspect, but there was marginal decrease in protein content in all the cultivars at post- harvest storage stages over the harvest stage. Similar trend of variability for protein content in broccoli cultivars at the time of marketable maturity have also been reported elsewhere (Gautam, 2001). Zhuang *et al.* (1997) suggested that increased temperature accelerated soluble protein losses. It was observed that at the end of storage period of 144 hours at 23^0C, protein content decreased by 70 per cent in broccoli.

The edible heads of broccoli cultivar(s) evaluated for total lipids showed wide variability at various harvest stages to range from 243.3 to 288.3, 240 to 283.3, 233.3 to 280 and 228.6 to 276.6 mg/100g at harvest and post- harvest storage stages- I, II and III, accordingly. The harvest stage exhibited maximum lipid content, whereas slight decreasing trend was observed in the lipid content at post- harvest stages. Variation in the total lipids content at various post - harvest

Table 1. Biochemical composition of edible heads of various broccoli cultivars at harvest and post harvest stages (Values on dry weight basis)

Parameters / Cultivars	Palam Samridhi				Palam Vichitra				Palam Kanchan				Palam Haritika				CD (P=0.05)
	I	II	III	IV	I	II	III	IV	I	II	III	IV	I	II	III	IV	
*Moisture (%)	90.52	88.70	88.18	88.16	89.96	89.30	88.48	87.78	91.24	90.02	89.81	88.44	89.56	89.01	88.28	87.95	0.25
Protein (%)	3.62	3.60	3.58	3.55	3.85	3.80	3.77	3.75	3.16	3.12	3.08	3.02	3.86	3.82	3.77	3.72	0.004
*Total lipids (mg/100g)	286.6	283.3	276.6	270.0	288.3	283.3	280.0	276.6	243.3	240.0	233.3	228.6	256.6	255.0	250.0	243.3	2.7
Carbo-hydrates (%)	5.6	5.4	5.2	5.0	5.5	5.3	5.2	5.1	5.5	5.2	5.1	4.8	5.4	5.1	5.0	4.9	0.03
*Total chloro-phyll (mg/g)	0.42	0.38	0.34	0.30	0.13	0.08	0.06	0.04	0.22	0.18	0.14	0.10	0.39	0.35	0.31	0.27	0.005
*Total caro-tenoids (mg/g)	1.35	1.03	0.88	0.69	0.36	0.33	0.32	0.29	1.40	1.38	1.36	1.33	0.88	0.84	0.80	0.76	0.04
*Ascorbic acid (mg/100g)	81.31	78.45	75.63	71.04	58.80	55.55	51.39	47.24	67.45	63.12	60.26	55.23	75.54	71.80	67.74	64.12	0.20
Total phenols (%)	1.18	1.28	1.38	1.49	0.87	1.16	1.25	1.32	1.11	1.20	1.29	1.39	1.10	1.19	1.29	1.39	0.008
*Glucosino-lates (mg/100g)	309.7	307.4	303.8	298.1	292.6	290.1	286.5	280.5	264.1	258.1	254.1	250.1	344.2	340.5	335.3	329.8	0.21
Calcium (mg/100g)	54.0	53.50	53.50	53.50	79.80	79.50	78.80	78.80	61.93	61.60	61.50	61.25	54.18	54.08	54.08	53.30	0.28
Iron (mg/100g)	0.90	0.90	0.90	0.90	1.61	1.60	1.60	1.60	1.40	1.40	1.40	1.40	0.91	0.90	0.90	0.90	NS
Sodium (mg/100g)	26.0	26.00	26.00	25.6	25.0	25.00	24.80	24.6	23.0	23.00	23.00	22.16	23.10	23.00	23.00	22.78	0.25
Potassium (mg/100g)	123.8	123.0	123.0	122.0	314.0	314.0	314.0	314.0	324.6	324.5	324.5	324.5	124.0	124.0	124.0	124.0	0.23
Phosphorus (mg/100g)	61.05	61.00	61.00	60.00	79.00	79.00	79.00	79.00	52.00	52.00	52.00	52.00	62.00	62.00	62.00	62.00	0.19

*Values on fresh weight basis. Stages of heads harvest: Harvest stage (129 DAS), Post-Harvest stage-I (129 DAS+ 2 days of storage at normal room temperature condition), Post - harvest stage-II (129 DAS+ 4 days storage at normal room temperature condition), Post - harvest stage-III (129 DAS+ 6 days storage at normal room temperature condition)

storage duration also revealed that cultivar Palam Vichitra emerged superior over other cultivars. The range of variation observed for total lipids content in the accompanying study is in accordance with the findings of Willis *et al.* (1998), who had reported that flower heads of broccoli cultivars contained total lipids content from 880 to 980 mg/100g at harvest stage (on fresh weight basis in the edible portion of the broccoli).

Dark green varieties of broccoli contain significantly more chlorophyll than grey-green or violet varieties. The high concentration of β-carotene, which has a positive effect on our visual activity, has been reported in the literature in these vegetables (Biermann Medical News, 2000). Significant variations in total chlorophyll content at various harvest stages among the broccoli cultivars were noticed. At harvest stage, variations from 0.13 to 0.42 mg/g in total chlorophyll content and 0.08 to 0.38, 0.06 to 0.34, 0.04 to 0.30 mg/g at post - harvest storage stages- I, II and III, respectively were observed on comparing all the head harvest stages. The harvest stage exhibited the maximum chlorophyll content. Among various post- harvest storage stages, the post - harvest stage- I possessed maximum chlorophyll content, whereas post - harvest stage - II and III followed the decreasing trend. The maximum value for this parameter was exhibited by Palam Samridhi accompanied by Palam Haritika, Palam Kanchan and Palam Vichitra in that order in all the stages of post- harvest. The findings of the study are altogether in consonance with the observations of other researchers on this aspect (Gautam, 2001). Paradis *et al.* (1995) observed that during storage at 4^0C, chlorophyll contents were almost stable, whereas storage at 20^0C was associated with chlorophyll losses.

Carotenoids are widely recognized as one of the important nutritional components essentially needed for regulating metabolic processes in human beings. Significant variation in the total carotenoids was also observed to range from 0.36 to 1.4 mg/g with its highest value in Palam Kanchan followed by Palam Samridhi, Palam Haritika and Palam Vichitra at harvest stage. Variations in total carotenoids were observed from 0.33 to 1.38, 0.32 to 1.36 and 0.29 to 1.33 at post harvest storage stages- I, II and III in that order. However, the minimum value was observed in Palam Vichitra, while the maximum value was possessed by Palam Kanchan. The trend of variation observed in total carotenoids content in the present study is in conformity with the findings of Krumbein *et al.* (1999). Studies conducted earlier also revealed that on an increased temperature at 16.5^0C, the total carotenoids level declined from 30 to 60 *per cent* (Biermann Medical News, 2000).

The presence of high concentrations of ascorbic acid is reported to exhibit positive effect on collagen synthesis and visual activity (Biermann Medical News, 2000). Besides, it is a cofactor in the enzymatic conversion of glucosinolates to anti-carcinogenic compounds, such as sulforaphane. As such the ascorbic acid content in broccoli cultivars was evaluated and data presented in Table 1 revealed significant variation from 58.80 to 81.31, 55.55 to 78.45, 51.39 to 75.63 and 47.24 to 71.04 mg/100gin this parameter at harvest and post- harvest storage stages - I, II and III, accordingly. The highest and lowest values of ascorbic acid were exhibited by Palam Samridhi and Palam Vichitra. The findings of the study on this aspect corroborated with the observations made by other investigators on cole crops including broccoli (Rognerud and Roennevig, 1976). Nevertheless, post- harvest storage studies showed marginal declining trend over harvest stage. Paradis *et al.* (1995) reported that on storage at 4^0C ascorbic acid was generally stable, whereas storage at 20^0C was associated with ascorbic acid losses.

Phenolics form one of the prominent secondary plant metabolites in plants including edible parts of its produce and exhibit anti-oxidant characteristics. However, when present beyond permissible limit in various plant parts, theses are broadly considered as anti- nutritional factor in human diet as prolong/ excess intake of these eatables are reported to impair absorption of proteins and trace elements in human body. In spite of the fact that total phenols level in broccoli heads was very low, nevertheless, significant variation in total phenols among various broccoli cultivars at harvest stage was observed to range from 0.87 to 1.18 per cent. Palam Vichitra exhibited the lowest value for this parameter, while Palam Samridhi showed the highest value at the harvest stage. Variability at post- harvest storage stages – I, II and III with regard to total phenols among different broccoli cultivars was found from 1.16 to 1.28, 1.25 to 1.38 and 1.32 to

1.49 per cent, respectively. The minimum value was observed by Palam Vichitra, while its maximum value was noticed in the cultivar Palam Samridhi at all the post- harvest storage stages under investigation. Variations in total phenols among various broccoli cultivars were more or less close to those reported by Vinson *et al.* (1998). Leja *et al.* (2001) observed that low temperature treatment resulted in an increase in the total phenol content after seven day storage of non- packaged heads.

Leafy vegetables and lots of other fruits and vegetables besides other nutritionally desirable constituents, contain a cocktail of compounds called 'phytochemicals', which protect humans against a wide range of ailments including cancers. These chemicals in cole crops known as glucosinolates are largely responsible for the taste and smell of brussels sprouts. These compounds are probably also involved in the plants' defences against predators and fungal attacks. As such, the status of glucosinolates of broccoli cultivars studied at various harvest intervals was evaluated and found to range significantly from 264.1 to 344.2, 258.1 to 340.5, 254.1 to 335.3 and 250.1 to 329.8 mg/100g at harvest and post- harvest storage stages, in that order. Palam Haritika emerged the best cultivar on this aspect over the others. Harvest stage showed maximum glucosinolates content, while post- harvest storage stages exhibited slightly decreasing trend over harvest stage. The trend of variability for glucosinolates was in agreement with those reported by other investigators on this aspect earlier (Hansen *et al.*, 1995). Further studies conducted by Krumbein *et al.* (1999) found that content of glucosinolates decreased with advanced development of broccoli head. However, on rise in average daily temperature to more than 16.5^{0}C, the glucosinolates content of broccoli was reported to fall from 30 to 60 *per cent* (Biermann Medical News, 2000).

The carbohydrates are primarily considered as readily available source of energy and form an important structural component of the body tissues. The study undertaken on this aspect revealed marginal variation in carbohydrate content among different cultivars (from 5.4 to 5.6 per cent) at harvest stage. The lowest value was observed by Palam Haritika and Palam Samridhi showed its highest value. At post-harvest storage stages – I, II and III, carbohydrate content varied from 5.1 to 5.4, 5.0 to 5.2 and 4.8 to 5.1 *per cent* in that order. The maximum value of this parameter was showed by Palam Samridhi and Palam Vichitra followed by Palam Haritika and Palam Kanchan. However, slight decrease in carbohydrates was found in different post- harvest storage stages when compared with harvest stage. Variations in carbohydrates have been reported by other workers and values obtained in the present investigation were found to be in consonance with the findings of Peirce (1987). Kubota *et al.* (1997) reported that levels of soluble sugars were reduced significantly in dark- stored plantlets.

Besides presence of nutraceutical compounds in substantial amounts edible broccoli heads also form rich source of minerals with special reference to phosphorus, potassium, calcium, sodium and iron content. Calcium is essentially required for various physiological functions such as nerve conduction, muscle contraction and relaxation etc. Among different broccoli cultivars, calcium content varied from 54.00 to 79.80 , 53.50 to 79.50, 53.50 to 78.80 and 53.30 to 78.80 mg/100g at harvest and post-harvest storage stages – I, II and III, respectively. The highest value was noticed at harvest stage, but merely marginal decrease in calcium content at post- harvest storage stage(s) was found and the cultivar Palam Vichitra emerged out to be high calcium containing cultivar.

Phosphorus is an essential element required for several life supporting processes. As the structural component, in association with calcium it is required for the formation and maintenance of skeleton and teeth. Hence, the broccoli cultivars were evaluated for phosphorus content and values on this aspect were found to range significantly from 52.0 to 79.0 mg/100g at harvest and post- harvest storage stages. Nutritionally, essential iron content was also determined and its value varied from 0.90 to 1.61 and 0.90 to 1.60, 0.90 to 1.60, 0.90 to 1.60 mg/100g at harvest and post - harvest storage stages - I, II and III, accordingly. Variation in potassium content was observed from 122.8 to 324.6, 123.0 to 324.5, 123.0 to 324.5 and 122.0 to 324.5 mg/100g, whereas differences in sodium content were noticed from 23.0 to 26.0, 23.0 to 26.0, 23.0 to 26.0 and 22.16 to 25.6 mg/100g in various broccoli cultivars at harvest and post-harvest storage stages- I, II and III, in that order.

The similar trend of genotypic variability in broccoli cultivars for various minerals under study has been reported elsewhere (Peirce, 1987; Farnham *et al.*, 2000).

Table 2. Ranking of broccoli cultivars in search of multipurpose cultivars at harvest and post harvest stage (s)

Parameter (s)	Harvest				Post Harvest stage – I				Post Harvest stage - II				Post Harvest stage - III			
	I	II	III	IV	I	II	III	IV	I	II	III	IV	I	II	III	IV
Protein (%)	3	2	4	1	3	2	4	1	3	2	4	1	3	1	4	2
Carbohydrates (%)	1	2	2	4	1	2	3	4	1	1	2	4	2	1	4	3
Ascorbic acid (mg/ 100g)	1	4	3	4	1	4	3	2	1	4	3	2	1	4	3	2
Carotenoids (mg/ 100g)	2	4	1	3	1	4	3	2	1	4	3	2	2	4	3	1
Phenols (%)	4	1	3	2	3	1	4	2	4	1	2	2	4	1	2	1
Glucosinolates (mg/ 100g)	2	3	4	1	2	3	4	1	2	3	4	1	2	3	4	1
Calcium (mg/ 100g)	4	1	2	4	4	1	2	3	4	1	2	3	3	1	2	4
Iron (mg/ 100g)	4	1	2	3	4	1	2	3	4	1	2	4	4	1	2	4
Total	21	18	21	19	19	18	25	19	20	17	22	19	21	16	24	19
Cumulative ranking	3	1	3	2	2	1	3	2	3	1	4	2	3	1	4	2

Cultivars: I = Palam Samridhi, II = Palam Vichitra, III = Palam Kanchan and IV = Palam Haritika

The study indicated that cultivar(s) in proteins; and in lipids emerged promising for individual quality traits at harvest stage. Besides identifying genotypes superior in individual quality traits to be useful for breeding programme, attempts were made to carry out overall genotypic ranking in respect of prominent nutritional quality attributes as well as anti - oxidant characteristics taken together viz., protein, carbohydrate, ascorbic acid, total phenols, glucosinolates, total carotenoids, iron and calcium content in broccoli cultivars that were ranked in ascending order (Table 2). Based on cumulative rating, the overall superior multipurpose cultivars were identified as Palam Vichitra, Palam Haritika, Palam Samridhi and Palam Kanchan in order of excellence at harvest stage, whereas among various post- harvest storage stages, broccoli heads stored at room temperature for two days i.e., post harvest storage stage - I were adjudged to be the most suitable with regard to retention of nutrients over other post harvest storage stages. Nevertheless, further studies are warranted to evaluate broccoli cultivars in relation to antioxidant (free radical scavenging) potential and flower heads yield performance taken together for two consecutive seasons before specific recommendations are made pertaining to versatile cultivars for commercialization for overall health benefits.

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