

INFLUENCE OF STORAGE CONDITIONS ON ANTHOCYANINS, CAROTENOIDS AND COLOR QUALITY OF RED AMARANTH (*Amaranthus hybridus* L.) OF KASHMIR VALLEY

Gazala Qazi*¹, Shoukat Ara¹ and Hina Qazi²

¹Division of Environmental Sciences,

Sher-e-Kashmir University of Agricultural Sciences & Technology of Kashmir,

Shalimar, Srinagar, Jammu & Kashmir – 190 025 (India)

²Division of Textile Science and Apparel Designing,

SNDT Women's University, Santacruz, Mumbai-400049

ABSTRACT

Red Amaranth (*Amaranthus hybridus* L.) belonging to family Amaranthaceae is a tall, annual herb found in a variety of habitats including cultivated vegetable fields, roadsides, wastelands, etc. The objective of the present study was to evaluate the dye yielding potential by analyzing the total anthocyanins, total carotenoids, and the color characteristics of the above ground portion of *Amaranthus hybridus* L. of Kashmir valley. The stability and color quality of pigments (anthocyanins and carotenoids) were assessed by quantifying the pigments and recording their color quality under ambient and refrigerated storage conditions at 10 days interval at 0, 10, 30, 40, 50, 60, 70, 80 and 90 days after collection. Results revealed that the total anthocyanin content (mg/100 g) and total carotenoid content (mg/100 g) are in the range of 72.98 to 244.39 mg/100 g and 3.08 to 39.12 mg/100 g. Highest pigment content was recorded during 0 days of storage. With increase in days of storage pigment content decreased in both plant material as well as extract while as pigment degradation was more in the plant material as compared to the plant extract. L^* , a^* , b^* , Chroma (C^*), Hue angle (H°) and total color change (ΔE) changed with increase in days of storage. The L^* value ranged from 28.01 to 35.78, a^* , b^* , H° , C^* and ΔE values ranged from 26.72 to 32.58, -9.27 to -5.62, -15.88 (344.12°) to -11.88 (348.12°), 27.30 to 33.87, 63.15 to 73.06. With increase in days of storage a^* , C^* and ΔE values were increased while as L^* , b^* and H° values were decreased. From the present study it was concluded that Red Amaranth contained superior levels of anthocyanin content, thus it can be used as a raw material for extraction of magenta edible food color.

Key words : Anthocyanins, Carotenoids, Hunter Lab, *Amaranthus hybridus* L., Kashmir

I. INTRODUCTION

Red Amaranth (*Amaranthus hybridus* L.) belonging to family Amaranthaceae is a tall, annual herb found in a variety of habitats including cultivated vegetable fields, roadsides, wastelands, etc. Pigments from plant tissue include chlorophylls, carotenoids, flavonoids, anthocyanins, quinones, and betacyanins [1]. *Amaranthus* pigments have been used to color beverages, bread, and other foods [2]. [3] reported that a red-colored extract of

amaranth leaves had been used to color foods throughout the world. Pigments from vegetable amaranth (*A. tricolor*) are approved for food use in China [4], yet *Amaranthus* pigments have not been used as commonly as red beet pigments. *Amaranthus* pigments are very similar to beet pigments (betanine) in structure and properties. Thus, likely applications include: yogurt, sherbert, ice cream, frozen fruit desserts, candies, frostings, puddings [5]; bacon, sausage [6]; and beverages and canned fruit [7]. There has been little information published on food applications of *Amaranthus* pigments.

The color of a food substance indicates freshness and safety that are indices of good aesthetic and sensorial values [8]. In the ancient times wide varieties of food colorants were derived from natural sources-plant, animal or mineral. The British chemist Sir William Henry Perkin created the first synthetic dye, mauveine, in 1856 by oxidizing aniline. By the end of the century, eighty synthetic dyes were produced. With the discovery of synthetic dyes the use of natural food dyes declined in the middle of nineteenth century. Industrialization of the food system, including a rise in food processing, has increased the use of food additives including food dyes as synthetic food dyes are less expensive and have intense hue than natural edible dyes but have general toxicity [9]. Synthetic food dyes and other additives may contribute to hyperactivity and other disturbed behavior in children [10, 11, 12]. Interest in anthocyanin-rich foods and extracts has intensified because of their possible health benefits. Anthocyanins are potent antioxidants and may be chemoprotective. The increased stability of these pigments together with their added value due to potential beneficial effects opens a new window of opportunities for use of these extracts in a variety of food applications. Anthocyanins represent an option for use, with the potential to substitute artificial red dyes, since they present brilliant, attractive colors, besides possessing several recognized functional properties [13].

In view of harmful impact of synthetic dyes on human health and environment and beneficial effect of natural food colorants, the present investigation was carried out to study the natural dye yielding potential of Red Amaranth (*Amaranthus hybridus* L.) of Kashmir valley.

II MATERIAL AND METHODS

The experimental material, comprising of the plant material of Red Amaranth (*Amaranthus hybridus* L.) were collected from the local areas of Srinagar, Kashmir.

2.1 Total anthocyanin content (mg/100g)

Anthocyanins were calculated as per the method of [14].

2.1.1 Calculations

The total anthocyanin content was calculated using following equations :

$$\text{Total optical density} = \frac{\text{Optical density} \times \text{Volume made}}{\text{Weight of sample}} \times 100$$

$$\text{Total anthocyanin content (mg/100 g)} = \frac{\text{Total optical density}}{98.20} \times 100$$

2.2 Total carotenoid content (mg/100g)

Total carotenoids were estimated as per [15]. To remove the interference of the chlorophyll during carotenoid estimation, the filtrate obtained at the end of the above procedure was evaporated to dryness and the residue was dissolved in the minimum quantity (10 ml) of ethanol. Afterwards, 60% of KOH (1ml for 10ml) was added to it. The whole mixture was boiled for about 5-10 minutes. The equal amount of distilled water was added to it and the solution obtained was partitioned twice with petroleum ether. The combined ether layers were evaporated and the residue obtained was dissolved in 25 ml hexane. A pinch of anhydrous sodium sulphate was added to it and resultant mixture was filtered through Whatman No.1 filter paper and the optical density (O.D) at 449 nm was recorded using hexane as blank.

2.2.1 Calculations

The total carotenoid content was calculated using following equation :

$$\text{Total carotenoid content (mg/100 g)} = \frac{\text{Optical density} \times \text{Volume made}}{250 \times \text{Weight of sample}} \times 100$$

2.3 Evaluation of CIE L*a*b* values

CIE L*a*b* values of the dyed and undyed fabrics was determined by chromometer (Model CR-2000, Minolta, Osaka, Japan) equipped with 8 mm measuring head and AC illumination (6774 K) based on CIE system (International Commission on Illumination). The meter was calibrated using the manufacturer's standard white plate. L*, a* and b* coordinates, Chroma (C*) and hue angle (h°) values were calculated by the following equations:

$$\text{Chroma} = (a^{*2} + b^{*2})^{1/2}$$

$$\text{Hue (h}^\circ) = \tan^{-1} (b^*/a^*)$$

Total color change was calculated from the L*, a* and b*coordinates by applying the following equation:

$$\text{Total Color change } (\Delta E) = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2]^{1/2}$$

Where, $\Delta L^* = L^* \text{ sample} - L^* \text{ standard}$

$\Delta a^* = a^* \text{ sample} - a^* \text{ standard}$

$\Delta b^* = b^* \text{ sample} - b^* \text{ standard}$

III RESULTS AND DISCUSSION

3.1 Anthocyanin content (mg/100g)

Anthocyanin content in Red Amaranth (*Amaranthus hybridus* L.) ranged between 72.98 to 244.39 mg/100g (TABLE 1; Fig. 1a). Highest pigment content (244.39 mg/100g) was recorded on day 0 of storage and lowest (72.98 mg/100g) on 90th day of storage. [16] reported 103.6±10.4 mg/100g anthocyanins in the seeds and sprouts of *Amaranthus cruentus* v. Aztek and 90.83±9.2 mg/100g in *Amaranthus cruentus* v. Rawa. Values recorded in the present study are quite higher as the anthocyanin content in living tissues increases with

exposure to sunlight. In addition to the anthocyanins, Red Amaranth also contains betacyanin pigments. Red-violetbetacyanins have been reported from the leaves of vegetable Amaranth (*Amaranthus tricolor* L.) [17, 18, 19, 20].

Decreasing trend was recorded in TABLE 1 and every storage interval was significantly different from each other. The drastic decrease of anthocyanins was because of rapid degradation. Anthocyanin molecules are unstable and very sensitive to technological processing particularly when heat is involved. [21] have reported degradation of anthocyanin in Phalsanector, concentrate, squash and crush during entire period of storage. [22] reported anthocyanin loss in thermal processing and storage in osmodehydrated and pasteurized cherries. Similar findings have been reported by [23] in plum juice, [24] in cherry nectar, [25] in black currant nectar, [26] in strawberry jam, [27], in bilberry and strawberry jam, [28] in cherry, plum and raspberry. Anthocyanin degradation during refrigerated storage is believed to be due to native enzymes, particularly polyphenoloxidase.

3.2 Total carotenoid content (mg/100 g)

The total carotenoid content in Red Amaranth (*Amaranthus hybridus* L.) ranged between 3.08 to 39.12 mg/100g (TABLE 1; Fig. 1b). Highest (39.12 mg/100g) pigment content was recorded during 0 days of storage and lowest (3.08 mg/100g) on 90th day of storage in plant material under ambient storage conditions. Decreasing trend was recorded in TABLE 1 and every storage interval was significantly different from each other. [29] reported total carotenoid contents of 46-90 mg/100g DW in 15 varieties of Amaranth. Values recorded in the present study are lower as they are expressed in terms of fresh weight. Beta-carotene to the tune of 3.29 mg/100g have been reported from the leaves of Red Amaranth (*Amaranthus hybridus* L.) by [30]. [31] reported 113.6±9.3 and 131.3±10.7mg/100g DM total carotenoids in *Amaranthus hybridus* L. from different geographical regions in South Africa.

3.3 Color parameters (L*, a*, b*, C* and h°)

L*, a*, b*, Chroma (C*), Hue angle (H°) and total color change (ΔE) changed with increase in days of storage (TABLE 2). With increase in days of storage a*, C* and ΔE values were increased while as L*, b* and H° values were decreased. The L* value of Red Amaranth (*Amaranthus hybridus* L.) ranged between 28.01 to 35.78. The L* value showed the decreasing trend with increase in storage time in both ambient as well as refrigerated conditions, showing darker color with storage. The a* value ranged between 26.72 to 32.58. The a* value showed the increasing trend with storage in both storage conditions reflecting reddening of plant sample. The b* value ranged between -9.27 to -5.62. The b* value showed the decreasing trend with increase in storage time in both storage conditions. The H° ranged between 344.12 to 375.41°, with increase in days of storage the hue changes from purplish red to magenta. These results are in conformity with those of [32] who reported the hue angles in the range of 339 to 356° for *Amaranthus* pigments. Further the C* ranged between 27.30 to 33.87 showing increasing trend. ΔE indicating the total colour difference between two samples ranged between 63.15 to 73.06. It showed the increasing trend with increase in storage and the total colour difference (ΔE) between plant material stored under refrigerated conditions was smaller than those stored under ambient conditions.

3.3.1 Trend of hue angle (H°) changes

The hue angle (H°) of the selected plant species during 0 and 90 days of storage under ambient and refrigerated are shown in TABLE 2. The less changes in hue angle (H°) were recorded under refrigerated conditions as compared to the ambient storage conditions in selected plant species. Thus, among the two storage conditions, the refrigerated condition is the best condition for maintaining the hue angle (H°).

Table 1: Changes in Anthocyanin Content (mg/100 g) and Total Carotenoid Content (mg/100 g) at Different Storage Intervals of Plant Material and Extract of Red Amaranth (*Amaranthus hybridus* L.) Under Ambient (12-28 °C) and Refrigerated (4 °C) Storage Conditions

Storage intervals (days)	Red Amaranth (<i>Amaranthus hybridus</i> L.)							
	Anthocyanin content (mg/100 g)				Total carotenoid content (mg/100 g)			
	Plant material		Extract		Plant material		Extract	
	Ambient	Refrigerated	Ambient	Refrigerated	Ambient	Refrigerated	Ambient	Refrigerated
D ₀	244.39 (15.66)	244.39 (15.67)	244.39 (15.66)	244.39 (15.66)	39.12	39.12	39.12	39.12
D ₁₀	222.33 (14.94)	229.12 (15.17)	234.21 (15.33)	237.61 (15.44)	32.24	34.69	35.90	37.27
D ₂₀	212.15 (14.60)	215.54 (14.71)	224.03 (15.00)	230.82 (15.22)	28.61	31.94	33.22	34.20
D ₃₀	195.18 (14.00)	201.96 (14.24)	208.75 (14.48)	220.63 (14.89)	25.60	29.23	30.90	31.42
D ₄₀	179.90 (13.45)	191.78 (13.88)	201.96 (14.25)	213.84 (14.66)	21.97	25.27	26.87	28.40
D ₅₀	151.05 (12.33)	166.32 (12.93)	178.20 (13.38)	188.39 (13.76)	17.84	21.30	23.10	25.40
D ₆₀	130.67 (11.47)	156.14 (12.53)	166.32 (12.93)	178.20 (13.39)	14.70	17.07	19.94	22.24
D ₇₀	117.10 (10.86)	132.38 (11.55)	139.17 (11.84)	161.23 (12.74)	10.64	12.70	17.36	18.87
D ₈₀	93.34 (9.71)	108.62 (10.46)	120.50 (11.02)	128.98 (11.40)	5.00	9.07	13.77	15.93
D ₉₀	72.98 (8.59)	93.34 (9.71)	105.22 (10.30)	117.10 (10.86)	3.08	6.20	9.77	12.07
C.D(p<0.05)	0.305	0.278	0.254	0.252	1.283	1.546	1.393	1.299

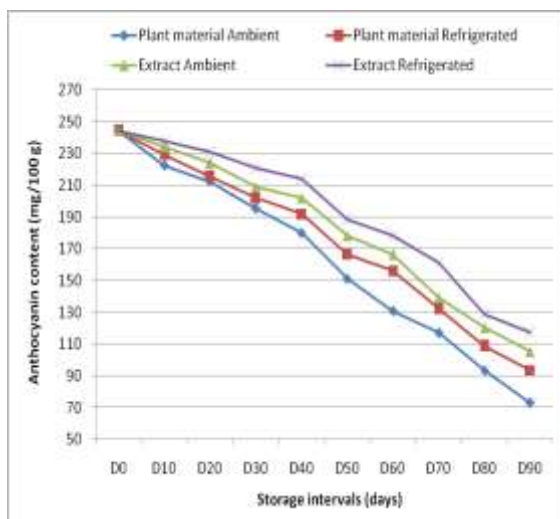
*Figures in parentheses are square root transformed means

Table 2: Chromaticity of the Red Amaranth (*Amaranthus hybridus* L.) under Ambient (12-28 °C) and Refrigerated (4 °C) Conditions

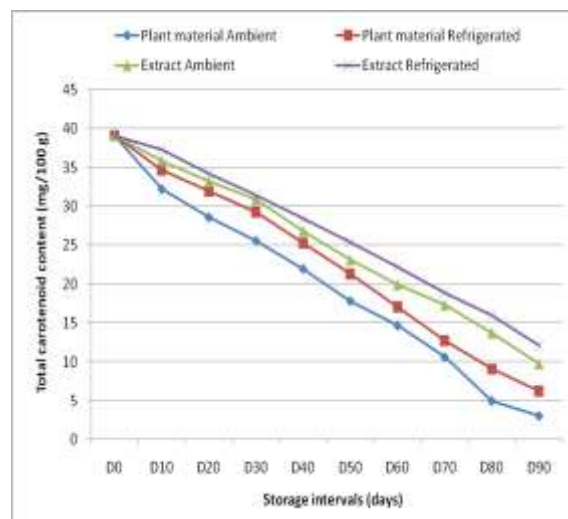
Storage intervals (days)	Red Amaranth (<i>Amaranthus hybridus</i> L.)											
	Color coordinates						Hue angle (H°)	Chroma (C*)		Total color change (ΔE)		
	L*		a*		b*							
	A	R	A	R	A	R	A	R	A	R		
D ₀	35.78	35.78	26.72	26.72	-5.62	-5.62	-11.88 (348.12)	-11.88 (348.12)	27.30	27.30	63.15	63.15
D ₁₀	35.58	35.73	27.7	26.97	-6.11	-5.82	-12.44 (347.56)	-12.18 (347.82)	28.37	27.59	63.80	63.32
D ₂₀	34.7	35.42	28.5	27.34	-7.05	-6.28	-13.89 (346.11)	-12.94 (347.06)	29.36	28.05	65.04	63.80

D ₃₀	34.14	34.76	28.92	27.69	-7.25	-6.45	-14.07 (345.93)	-13.11 (346.89)	29.81	28.43	65.74	64.56
D ₄₀	33.34	34.30	29.38	28.20	-7.45	-6.76	-14.23 (345.77)	-13.48 (346.52)	30.31	29.00	66.68	65.23
D ₅₀	31.72	33.70	29.95	28.49	-7.94	-7.01	-14.85 (345.15)	-13.82 (346.18)	30.98	29.34	68.43	65.92
D ₆₀	30.85	33.10	30.72	28.80	-8.42	-7.33	-15.33 (344.67)	-14.28 (345.72)	31.85	29.72	69.60	66.62
D ₇₀	29.51	32.83	31.37	29.21	-8.69	-7.59	-15.48 (344.52)	-14.57 (345.43)	32.55	30.18	71.11	67.07
D ₈₀	28.95	31.84	31.73	29.81	-8.95	-7.97	-15.75 (344.25)	-14.97 (345.03)	32.97	30.86	71.80	68.26
D ₉₀	28.01	30.94	32.58	30.22	-9.27	-8.33	-15.88 (344.12)	-15.41 (375.41)	33.87	31.35	73.06	69.28
C.D(p<0.05)	0.348	0.184	0.102	0.104	0.083	0.085						

A= Ambient; R = Refrigerated



a) Anthocyanin content (mg/100 g)



b) Total carotenoid content (mg/100 g)

Figure 1a & b : Changes in anthocyanin content (mg/100 g) and total carotenoid content (mg/100 g) at different storage intervals of plant material and extract of Red Amaranth (*Amaranthus hybridus* L.) under ambient (12-28 °C) and refrigerated (4 °C) storage conditions



Pic. 1: Red Amaranth (*Amaranthus hybridus* L.)



Pic. 2: Red Amaranth (*Amaranthus hybridus* L.) extracts

IV CONCLUSION

From the present study it could be concluded that Red Amaranth (*Amaranthus hybridus* L.) contained superior levels of anthocyanin content, thus it is recommended to be used as a raw material for extraction of reddish to pink edible food colors when stored in extract form under refrigerated conditions. However further research must be done in identifying the new areas of demand and application.

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