AIR TEMPERATURE AND SUNLIGHT INTENSITY OF DIFFERENT GROWING PERIOD AFFECTS THE BIOMASS, LEAF COLOR AND BETACYANIN PIGMENT ACCUMULATIONS IN RED AMARANTH (AMARANTHUS TRICOLOR L.)

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Manuscript received: June 25, 2009; Reviewed: December 22, 2009; Accepted for publication: January 12, 2010

ABSTRACT

The objectives of this study were to determine the effects of daily air temperature and sunlight intensity variations on biomass production, leaf color and betacyanin accumulations in red amaranth (Amaranthus tricolor L.). For this purpose, two improved cultivars; BARI-1 and Altopati were grown in seven different period (from April to October, 2006) under vinyl house condition in the experimental facilities of Gifu University, Japan. The mean daily temperatures fluctuated from 18 (growing month- April) to 29ºC (August), while the mean sunlight intensities varied from 850 (October) to 1257 µmol m^{-2} S^{-1} (August). The highest biomass yield and betacyanin accumulation was obtained in the warmer growing period (July and August) at 28 to 29ºC mean air temperatures and 1240 to 1257 µmol m^{-2} S^{-1} sunlight intensity. At the warmer growing period red amaranth produced red leaves with high color index, which enhanced the betacyanin accumulations. The biomass yield and betacyanin accumulations were reduced significantly in the growing period/month April and October under low temperature regimes (mean air temperature 18 and 19ºC, respectively). However, growing period's air temperature contributed more for biomass and betacyanin accumulations in red amaranth than sunlight intensity. Comparing two cultivars the biomass yield of BARI-1 was higher biomass yield than that of Altopati and Altopati highlighted with the higher betacyanin accumulations than that of BARI-1 in all growing period. Quantification of the effects of daily air temperature and sunlight intensity on biomass and betacyanin accumulation is important for growers producing these crops for fresh market and also optimize the best growing period. Therefore the influence of air temperatures and sunlight intensity should be considered while grown red amaranth for maximum yield with bioactive compounds like betacyanin and should be grown in between 28 to 29ºC air temperature and 1240 to 1257 µmol.m-2.S-1. of sunlight intensity.

KEY WORDS: Amaranthus tricolor, air temperature, sunlight intensity, biomass yield, betacyanin

INTRODUCTION

The main vegetable type of amaranth, Amaranthus tricolor L., seems to have originated in South or Southeast Asia [8] and then spread through the tropics and the temperate zone [11]. The two main types of amaranth grown as leafy vegetables are loosely termed red amaranth and green amaranth. The fresh tender leaves and stem of red amaranth are delicious when are cooked by boiling and mixing with condiments. The leaves and tender stems of red amaranth are rich in protein, minerals, vitamin A and C [21].

Mature leaves of A. tricolor contain red-violet pigments, betacyanins [16]. Betacyanin formation in Amaranthus cotyledons is a light controlled process, but it can also be induced by the plant hormones, and their analogs [14]. The active ingrendiets of betacyanin provides anti-inflammatory effects in our food as potential antioxidants [9, 5, and 19]. Interest in betacyanin has grown since antiradical activity were characterized, and the pigment is widely used as additive for food, drugs and cosmetic products, because of having natural properties and absence of toxicity [15]. Environmental air temperatures and sunlight can have significant impacts on accumulation of secondary plant compound like betacyanin. Betacyanin synthesis is enhanced at optimum growing temperatures and light, due to the light-sensitivity of certain enzymes involved in the synthetic pathway [7]. The change in leaf colors is dependent on many factors, such as shorter day lengths, temperature variance, and exposure to light and wind elements [18].

Seasonal temperature is an important climatic factor, which can have profound effects on the yield of crops. Changes in seasonal temperature affect the biological yield, mainly through phenological development processes. Significant variations of air temperature and sunlight can limit plant growth at both low and high regimes. Environmental conditions between these two extremes provide an optimum air temperature range for plant growth that allows for maximum productivity [1]. Red amaranth grows well in hot and humid weather. However, during winter, red amaranth growth and development is slow, compared with summer and rainy season [4]. The most important agronomic considerations for growers to optimize yield and quality are to select optimum air temperature and sunlight intensity.

However there is limited research to determine what impact air temperature and sunlight intensity has on the production of biomass and secondary plant compounds such as betacyanin pigments. The objectives of this study were to determine the effects of different air temperature and sunlight intensity on plant biomass production and accumulation of betacyanin pigment in leaves of red amaranth. Results of this study might be helpful to manage and manipulate the environmental factor to get maximum yield and nutritional values from red amaranth and also optimize the best sowing time in Japanese conditions.

MATERIALS AND METHODS

Place and Condition of the Experiment

Two improved red amaranth cultivars; BARI-1 and Altopati sample in Bangladesh were grown in the vinyl house to determine their total biomass, leaf color index and betacyanin content under different daily air temperature and sunlight intensity. About fifty seeds were sown under 1 cm depth soil in each planter pot. Seven sowing dates were considered and seeds were sown in the first week in each month from April to October 2006. Different growing period comprised the treatments, was arranged in a randomized complete block design with 3 replications. Plants were grown on nursery soil with element concentrations of N (nitrogen)- 0.28g.L^{-1}, P (phosphorus)- 0.65 g.L^{-1} and K (potassium)- 0.45 g.L^{-1} and soil pH 5.5-6.5. During the growing period (from germination to harvest) the light intensity was measured with a Quantum sensor light meter with Separate Sensor (QMSS) from. Each reading was expressed as μmol m^{-2} s^{-1}. The air temperature and humidity were recorded daily during the plant growing period in different growing months. Plant height, number of leaves, fresh and dry biomass were recorded at 15 days after sowing and continued at 20, 25 and 30 days after sowing, every time 9 plants were randomly selected from each planter.

Leaf color analysis

Color parameters- L* (lightness), a* (redness) and b* (yellowness) values were recorded from randomly selected leaf (30 days old plant) of each cultivar by a color meter (CS- Sharpener, Toppan, Japan) with three replications. Color index (CI) of leaf was calculated through the equation CI= 1000 x a*/L x b* [12].

Assay of Betacyanin

Leaves were collected from 30 days old plants and approximately 0.2 g leaf disks were taken by cutting roller (12 base) and homogenized with a mortar and pestle. Leaf paste was extracted with 20 ml 80% aqueous methanol containing 50 μM ascorbic acids and shaken for 30 minutes. The extraction was centrifuged at 14,000g for 10 min at 4°C, the supernatant was removed and the collected betacyanin was quantified by spectrophotometer (HITACHI U-1800, Tokyo, Japan) at 540 nm wavelength. Content of betacyanin was calculated using the extinction co-efficient of betacyanin (62 x 10^6 cm^2 mo^{-1}) described by Wyler et al., [22].

Table 1. Color Index, color parameters (L*, a*, b*) and betacyanin content in the leaves of Red amaranth cultivar BARI-1 and Altopati in different growing periods/months.

Sowing time	Mean Temp. (ºC)	Sunlight intensity $\mu mol \cdot m^{-2} \cdot S^{-1}$	Cultivar	Color Index (CI)	Lightness (L*)	Redness (a*)	Yellow ness (b*)	Betacyanin [µg betanin equivalent (g fresh weight)$^{-1}$]
April 4	18	975	BARI-1	93.37b	25.30a	9.54b	4.04c	383.23a
			Altopati	94.52b	25.54a	8.97b	3.71c	384.41a
May 5	21	1065	BARI-1	95.16b	26.83b	9.84c	3.85b	401.26b
			Altopati	98.94c	26.47b	9.46c	3.61b	402.15b
June 4	25	1208	BARI-1	102.99c	27.06c	10.60d	3.80ab	454.49d
			Altopati	99.61c	28.12c	9.85d	3.51a	455.08d
July 3	28	1240	BARI-1	104.98c	27.05c	10.67d	3.75a	470.75f
			Altopati	101.46d	28.06c	9.97f	3.50a	471.94f
August 3	29	1257	BARI-1	103.18c	27.04c	10.58d	3.79ab	464.25e
			Altopati	100.82d	28.03c	9.89e	3.5a	462.47e
September 4	24	1206	BARI-1	96.04b	26.82b	9.85c	3.82ab	421.08c
			Altopati	99.12c	26.47b	9.48c	3.61b	422.26c
October 5	19	850	BARI-1	90.14a	25.26a	9.3a	4.08c	382.93a
			Altopati	90.60a	25.37a	8.76a	3.81d	383.23a

Statistical Analysis

Data were analyzed using the PC software 'Excel Statistics' (Version 5.0, Esumi Co. Ltd., Japan) via ANOVA with Tukey's multiple comparison test ($P<0.05$) [13].

RESULTS AND DISCUSSION

Environmental Conditions

The temperatures and sunlight intensities were highly variable throughout the experiment season, while the variations of humidity were not so wide (Fig. 1). The distribution of air temperature and sunlight intensity in the month of July and August was the highest. These values in the April and October growing periods were lower with air temperature (18ºC and 19ºC, respectively) and sunlight intensity (975 to 850 µmol $m^{-2} S^{-1}$). Temperature and light intensity varied among the different growing period/month, and these differences in microclimate were implicated for the variability in growth pattern, biomass yield and betacyanin accumulations.

Biomass Yield and its Determinants

Variation of temperature and the sunlight intensity among different growing period significantly affected plant height (Fig.2). The mean data for growing period indicated that maximum plant height (BARI-1: 36.77cm; Altopati: 43.92cm) at 30 DAS was attained by cultivars sown on 03rd July, while minimum plant height (BARI-1: 21.39cm; Altopati: 20.24cm) observed in cultivars sown on 5th October. It is clear from the mean data that July sowing produced taller plant as compared to early (April and May) and later (September and October) sowing. It might be due to prevailing low temperature and sunlight intensity in early and late sowing. The correlation between plant height and mean growing period air temperature and sunlight intensity was highly significant (Fig. 4), which confirms air temperature and sunlight variability is a major factor influencing plant growth.

Number of leaves per plant plays a remarkable role in determining the biomass yield, which showed significant differences between the cultivars and among the growing period (Fig. 2). The mean data recorded for growing periods revealed that the maximum numbers of leaves (BARI-1: 20: Altopati: 19.66) per plant were produced at 30 DAS by plants sown on July, whereas the lower numbers of leaves (BARI-1: 10.22; Altopati: 8.44) per plant were obtained in plants sown on October. The leaf number increased linearly as the air temperature increased from 25 to 29ºC (June to August) and decreased linearly as the air temperature decreased from 18 to 24 ºC (April, September and October). Similarly the leaf number increased when sunlight intensity was higher from 1240 to 1257 µmol $m^{-2} S^{-1}$ and decrease when sunlight intensity was lower from 850 to 1065 µmol $m^{-2} S^{-1}$. So we assumed that, the leaf number responded strongly to changes in air temperature and sunlight intensity.

Like plant height and number of leaf, the fresh and dry yields were significantly affected by time of sowing (Fig. 3). The highest fresh and dry weight was observed at July followed by August, June, May and September. Considerably lower fresh and dry weight was observed at October growing period followed by April growing

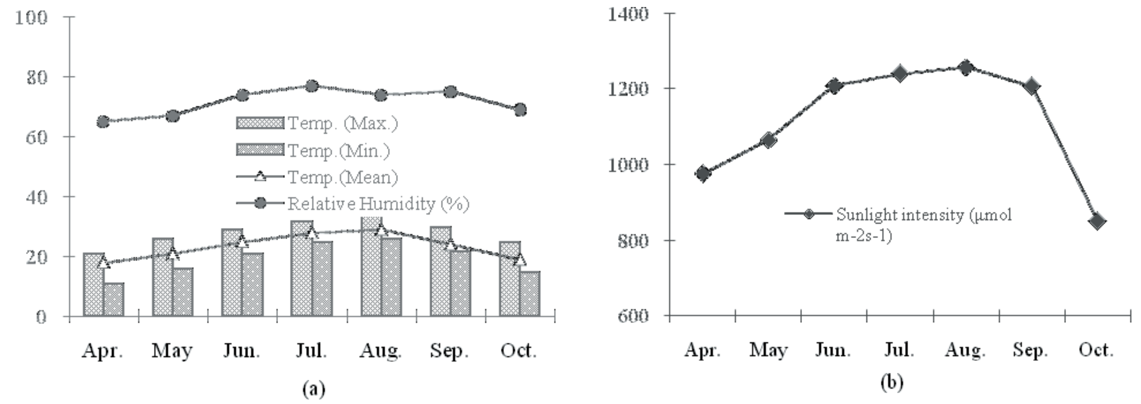

Fig. 1. Variations of air temperature, relative humidity and sunlight intensity of seven growing periods (April to October 2006); (a) Maximum, Minimum, Mean air temperature and relative humidity (%) and (b) sunlight intensity.

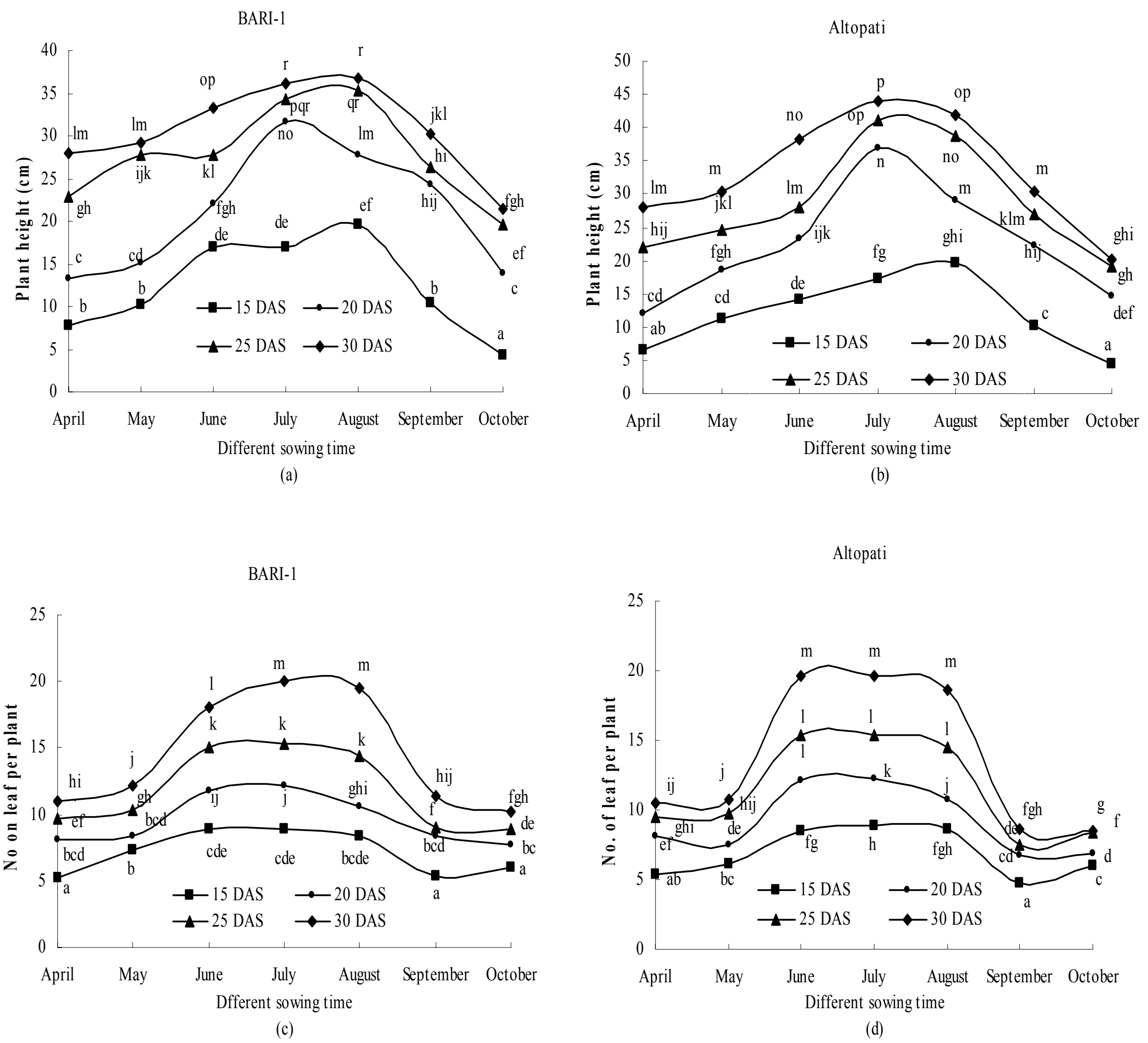

Fig. 2. Yield and yield contributing characters plant height (a) BARI-1, (b) Altopati; No. of leaf (c) BARI-1, (d) Altopati in different sowing times from April to October during 2006 growing season. Same letters on bars are not significantly different according to Tukey's test at $p < 0.05$.

period. The yield increased as the air temperature and sunlight intensity increased from 25 to 29ºC and 1240 to1257µmol m^{-2} S^{-1} respectively (June to August) and decreased as the air temperature and sunlight intensity decreased from 18 to 24ºC and 850 to 1065µmol m^{-2} S^{-1} respectively (April, September and October). Fresh and dry mass yield progressively decreased as sowing was early or late due to low temperature and sunlight intensity. Yield was reduced through a lower number of leaf set and smaller plant size.

BARI-1 produced higher fresh and dry biomass yield than Altopati in all growing period. This could be due to the different response of the cultivars to in photoperiod and temperature. The correlation among yield and growing period for average air temperature and sunlight intensity were highly significant, which indicate that the air temperature and sunlight intensity influence fresh and dry yield of red amaranth (Fig. 4.) Significant linear relationship between growing period and biological yield are consistent with the findings of Bange et al., [2] for the crop sunflower.

Temperature and light are linked through the processes of photosynthesis and respiration [6]. Under good light conditions high temperatures will mostly result in faster growth and higher yield. If light is weak, high temperatures are dangerous for plant growth [20]. Amaranthus tricolor L. is a C_4 plant and mostly characterized by high light intensities and high diurnal temperatures [10]. Low

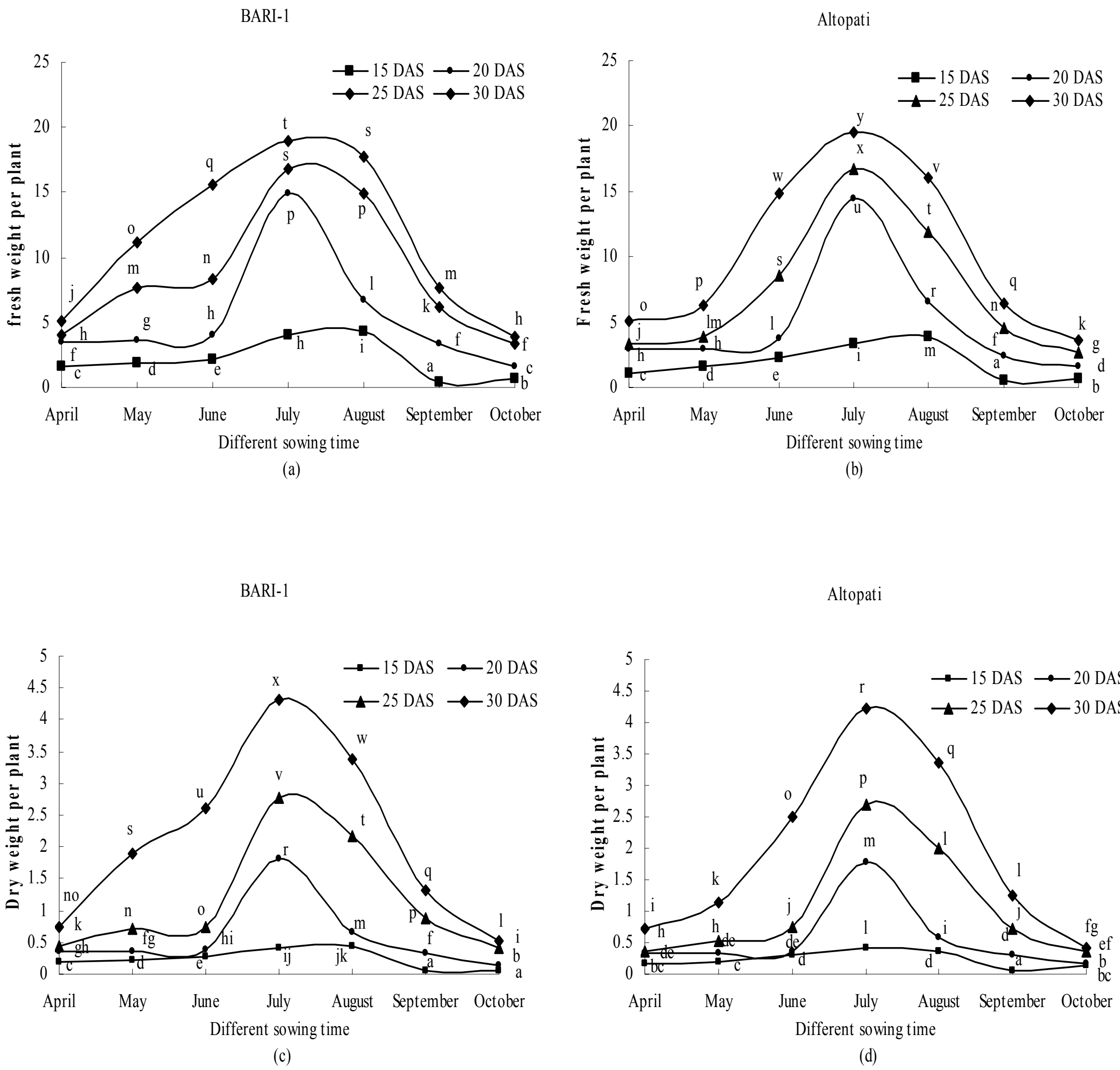

Fig. 3. Yield and yield contributing characters total fresh weight (a) BARI-1, (b) Altopati; total dry weight (c) BARI-1, (d) Altopati in different sowing times from April to October during 2006 growing season. Same letters on bars are not significantly different according to Tukey's test at $p < 0.05$.

temperatures are known to have a more negative effect on C_4 plants (such as Amaranthus) compared with C_3 plants [17]. We assumed that higher temperature and light intensity strongly influenced the fresh and dry matter yield in red amaranth.

Leaf Color and Betacyanin accumulations

The L*, a* and b* values and the color index of the leaf samples (Table 1) varied among the growing period. The cultivar BARI-1 and Altopati sown in July, August and September had the superior color index than those of rest growing period at 30 DAS. In contrast to color index lightness (L*), redness (a*) of the leaf of both cultivar showed better performance in the month of July and August than the other growing period. On the other hand leaf of both cultivars showed lower yellowness (b*) in the month of July and August than the other growing period. So higher redness (a*) in the month of June, July and August enhance the red leaf formation with higher color index at warmer climatic conditions. Growing period June, July and August gave the leaves superior leaf color index with higher redness under the higher sun light intensity and optimum temperature and April and October growing period gave the leaves lower color index under the lower sun light and temperature.

The major cause of leaf color might be changed by environmental factors such as light, temperature, and moisture and these factors regulated the physiology of plants by influencing the production and quality of flavonoids and groups of pigments. We observed the color index (CI) was highly correlated with the average air temperature and sunlight intensity of growing periods (Fig 5), which indicated that leaf color parameters of red amaranth was highly influenced by temperature and sunlight intensity. Low temperature and low sunlight intensity may damage leaves and induce color changes, so optimum temperature and sunlight intensity is desired to red leaf formation in red amaranth.

Betacyanin accumulations were highest in the July-sown red amaranth than those of rest growing period, although

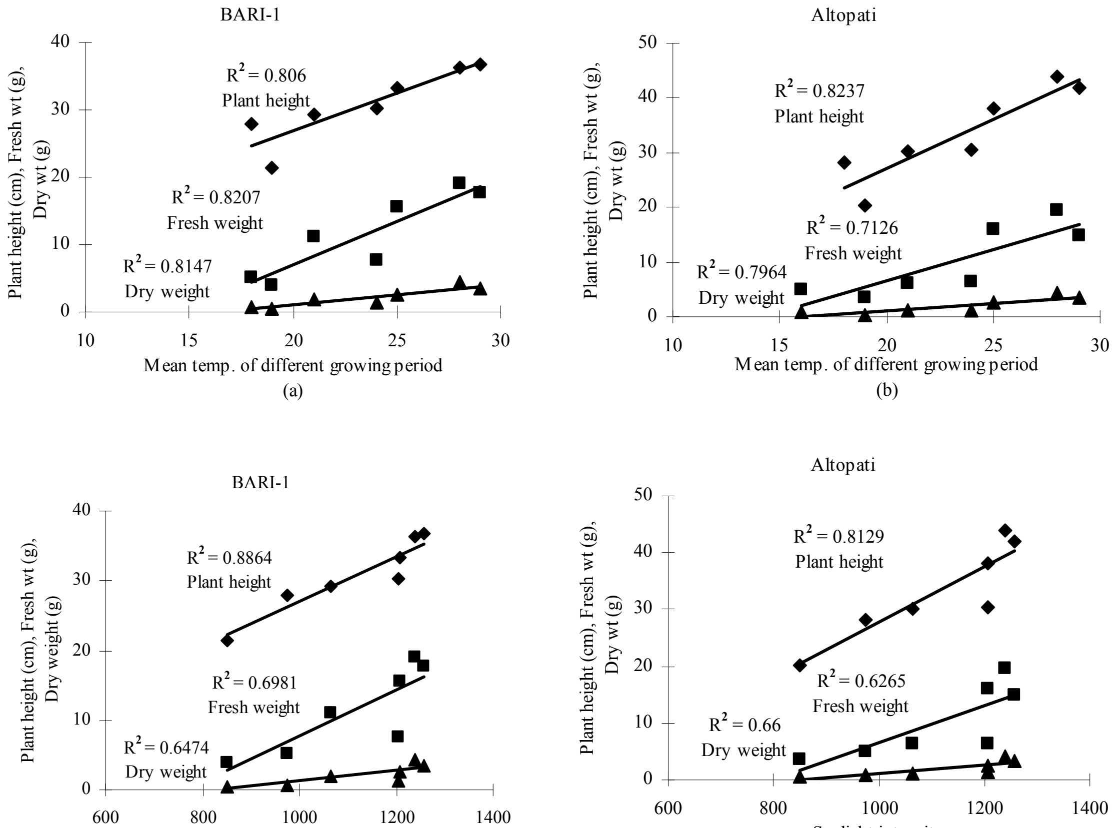

Fig. 4. Correlations among mean temperature with plant height, fresh weight and dry weight of (a) BARI-1 and (b) Altopati and correlations among sunlight intensity with plant height, fresh weight and dry weight of (c) BARI-1 and (d) Altopati at 30DAS after in different sowing times from April to October during 2006 growing season.

again the earlier (April and May) or later (September and October) growing period tended to have lower betacyanin content (Table 1). However, the concentrations of betacyanin were similar in the June, July and August sown red amaranth, and the concentration were significantly higher than April, May or October sown red amaranth. The pigment synthesis of many plants has been usually influenced by various factors including light; temperature and nutrient source and these factors fluctuate in different growing period [3].

The current study demonstrates that air temperatures and sunlight intensity influences the production of betacyanin pigments in the leaves of red amaranth. Betacyanin concentrations in leaves of red amaranth increased as the air temperatures increased from 24 to 29ºC while the betacyanin concentrations decreased in red amaranth as the air temperature decreased from 19 to 21ºC. Similarly betacyanin concentrations increased when sunlight intensity was higher from 1240 to 1257 µmol m^{-2} S^{-1} and the accumulation of betacyanin decrease when sunlight intensity was lower from 850 to 1065 µmol m^{-2} S^{-1}. Each betacyanin is a glycoside and consists of a sugar and a colored portion and their synthesis is promoted by light and optimum temperature [17] and since radiation and temperature were higher for the July, August and September sowing, therefore expected to be more abundant than for the April, May and October sowing.

In a different field experiment, red amaranth grown in the constant mean air temperature (27±2ºC) under manipulated sunlight intensity by shading net resulted narrow variations for biomass yield and betacyanin accumulations, where full sunlight intensity (1235 µmol m^{-2} S^{-1}) produced 30% more yield for dry and fresh biomass and 14% more betacyanin than lower light intensity (678 µmol m^{-2} S^{-1}) (data not shown). In the current study plants grown in 29ºC air temperature and full sunlight (1240 µmol m^{-2} S^{-1}) produced 60% more

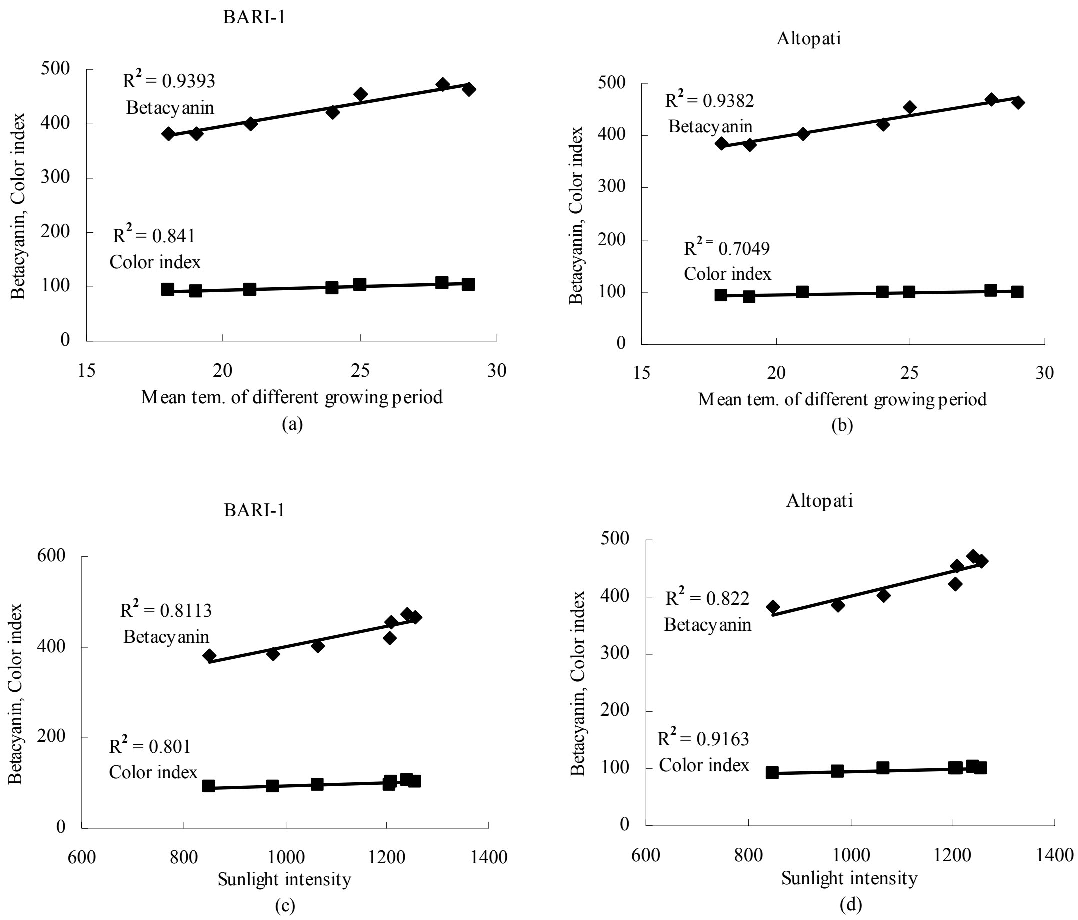

Fig. 5. Correlations among mean temperature with betacyanin and color index of (a) BARI-1 and (b) Altopati.and correlations among sunlight intensity with betacyanin and color index of (c) BARI-1 and (d) Altopati at 30DAS after in different sowing times from April to October during 2006 growing season.

fresh and dry biomass yield and 19% more betacyanin than plants grown in 18°C air temperature and 850 μmol m^{-2} S^{-1}sunlight intensity. Although 850 μmol m^{-2} S^{-1} sunlight is enough for optimum plant growth, we assumed that low air temperature significantly affected the biomass yield and betacyanin accumulations during comparatively cool climate-growing period. Hence we found, growing period's air temperature variations has higher effect than sunlight intensity for biomass and betacyanin accumulations in red amaranth. Therefore the influence of air temperatures and sunlight intensity should be considered while grown red amaranth for maximum yield with bioactive compounds like betacyanin and should be grow optimally under warm conditions and bright light.

As a conclusion, the results of this study showed that environmental factors, especially amount and effects of air temperature and sunlight intensity during plant growing period, significantly affected biomass yield, leaf color and betacyanin accumulations. Hyperbolic correlation coefficients of air temperature and sunlight intensity with the biomass productivity and betacyanin accumulations proved that, the air temperatures were more influential for biomass yield and betacyanin accumulations in red amaranth. The cooler growing condition (early and late growing periods) was exposed lower temperature and lower sunlight intensity caused the crop lower potential incidences of plant development. The warmer climatic conditions (optimum air temperature and sunlight intensity) increased plant height, leaf number, fresh and dry weight, leaf color and betacyanin accumulations. Comparisons of seven growing period, the superior yield and yield contributing characters were found in July followed by the order August> June> May> September> April> October. The growing periods June, July and August allowed the crop to avoid the problems with cold temperatures and lower sunlight intensity. Quantification the effects of air temperatures and sunlight intensity on biomass and betacyanin is important for growers producing red amaranth for fresh market and also optimize best sowing time in Japanese conditions. Therefore the influence of air temperatures and sunlight intensity should be considered while grown red amaranth for maximum yield with bioactive compounds like betacyanin and should be grown in between 28 to 29ºC air temperature and 1240 to 1257 μmol.m^{-2}.S^{-1} of sunlight intensity.

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