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Research Paper

Assimilates transfer between fern and rhizome over an extended season in 2X and 8X asparagus

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Abstract
Asparagus (*Asparagus officinalis* L.) is a perennial vegetable crop with different ploidy levels. The translocation of assimilates in diploid (2X) and octoploid (8X) asparagus between rhizome and fern needs to be understood. Transfer of soluble sugars, pigments, and invertase activity were studied in the two-year-old asparagus from May to November 2019. Sucrose and glucose were the major soluble carbohydrates in the asparagus rhizome. In the case of 2X asparagus, there was a balance of total sugars in the fern and rhizome, as opposed to anthocyanin. May and September were critical stages in the consumption or storage of substances in 2X asparagus. Sucrose transferred from fern to rhizome from August to October in 2X and 8X. Total sugars in rhizome decreased from May to June in 2X and 8X. Invertase activity increased from May to June and after that declined in both 2X and 8X. The growth of fern was dominant to the rhizome until August and the loading of glucose in the rhizome was low. The discriminant analysis indicated the difference in the response pattern of the 2X and 8X asparagus, where the rhizome fructose, fern carotenoid, and rhizome glucose in 2X but the rhizome sucrose, rhizome total sugars, fern fructose, fern anthocyanin, fern invertase, and fern sucrose were the most influential variables in these two types of asparagus. The total sugars, sucrose, glucose, and fructose transfer patterns in diploid and octoploid asparagus can be used for nutrition management by asparagus growers.

Keywords: carbohydrate, invertase, ploidy, sucrose, total sugar

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Introduction

Garden asparagus (*Asparagus officinalis* L.) has a basic chromosome number of X=10 (2n=2X=20) in which a wide range of ploidy levels (2X, 4X, 6X, 8X, and 10X) have been detected (Moreno *et al*. 2006; Castro *et al.* 2013; Mousavizadeh *et al*. 2016). Polyploid asparagus has the characteristics of increased size, resistance to abiotic and biotic challenges, and poor fertility (Chen *et al.* 2014). It was clear that 2X asparagus like *A. officinalis*, *A. persicus*, and *A. verticillatus* were adapted to a similar area, which differs from that of 8X and 10X *A. officinalis* (Mousavizadeh *et al*. 2022). The lack of physiological similarity between diploids and polyploids has led to their differential spread in natural habitats (Manzaneda *et al.* 2012). Resistance to

diseases, spear diameter, strength, and productivity were found more in octoploid asparagus of "Morado de Hetor" (Moreno *et al.* 2006). Morphological changes (Moreno *et al.* 2006). Morphological changes and chlorophyll content enhancement were observed in polyploid asparagus compared to the diploids (Carmona-Martin *et al.* 2014).

Production of the commercial part of asparagus (spear) depends on photosynthesis during the last season with photosynthate stored in the rhizome (Pressman *et al.* 1993). Photosynthetic carbohydrates and soluble sugars transfer into the storage roots of asparagus in the first season and are used in the following season for spear production (Bhowmik and Matsui 2003). Biochemical characteristics of asparagus spear vary with temperature changes (Shou *et al.* 2007). Light and temperature during the growth season are important factors in asparagus sugar metabolism (Anastasiadi *et al.* 2020). Sugars are important for the taste and texture of asparagus spears (Takahashi *et al.* 2019). Carbohydrates produce energy in asparagus (Creydt and Fischer 2020). Carbohydrate content in asparagus is affected by the loss of crown storage carbohydrates over the harvest season (Bhowmik *et al.* 2001). Soluble and insoluble sugars had been quantified in asparagus spears grown in the field (Takahashi *et al.* 2019).

Based on the ploidy evolution, the asparagus growth pattern changed due to the transfer of carbohydrates from ferns into rhizomes which are necessary for the growth and production of the following season. Carbohydrate transfer pattern in the two-year old asparagus needs to be investigated from the beginning of growth to the beginning of dormancy. The objective of this study was to understand the transfer pattern of soluble sugars and pigments between fern and rhizome in two-year-old 2x and 8x asparagus.

Materials and Methods

Plant materials and field operation

Seeds (40-50 no) of diploid (2n=2x=20) and octoploid (2n=8x=80) asparagus were collected from the natural habitats of Iran as follows 2x asparagus was collected from the Alborz province- Karaj, 35°82' N, 50°92' E, ELV 200, cold Mediterranean climate; 8x asparagus was collected from the Mazandaran province- Gazanak, 35°53' N, 52°12' E, ELV 1670, semi-humid Mediterranean climate (Mousavizadeh *et al.* 2015, 2016).

Ploidy level was determined by estimating the relative DNA content using flow cytometry (Mousavizadeh *et al.* 2016). The seeds were placed on the absorbent filter paper in 10-cm Petri dishes and moistened with 10 mL distilled water. Germinated seeds were sown in a 128-cell tray (each cell 4 cm^2) filled with peat moss and perlite (1:1 v/v). The trays were kept in a greenhouse with 16 h light, 20 ^oC, and

supplied with a nutrient solution containing N20: P20: K20. Seedlings were grown for three months and then transplanted to the field.

The field experiment was conducted at the Gorgan University of Agriculture Sciences and Natural Resources Research Station, Gorgan

Table 1. Climactic data of the growth season in 2018-2019.

Month	Evaporation (mm)	Sunshine (h)	Average relative humidity (%)	Average maximum temperature (ºC)	Average minimum temperature (ºC)	Average temperature (ºC)	Days with precipitation	Precipitation (mm)
May	128.3	201.9	66	27.1	14.1	20.6	9	23.5
June	203.3	248.7	61	31.8	19.7	25.8	10	10.5
July	274.5	302.1	55	37.5	25.1	31.3	3	15
August	200	193.7	66	34.8	25.6	30.2	9	24.8
September	194.2	246	62	32.9	21.7	27.3	3	6.2
October	103.6	223	65	27.9	15.4	21.7	6	55.5
November	63.6	161.9	71	21.4	10.9	16.1	8	37.3

Table 2. Soil characters of the experimental site in 2018-2019.

Soil texture	Mg (ppm)	Fe (ppm)	K (ppm)	P (ppm)	Organic carbon (%)	Naturalized materials (%)	pH	EC (mmhos·cm^{-2})	Saturation point
Silty clay loam	457	14.1	186	11.3	0.84	2.25	7.9	0.6	51.5

(36º51' N, 54º16' E, and 13 m ASL), Iran in the growing season of 2017. Maximum and minimum temperature, sunshine hours, precipitation, and other metrological information were measured at a weather station located approximately 4 km from the experimental site.

The asparagus seedlings were sown in 3×7 m plots. Every plot consisted of three rows and seedlings were placed in 5 cm deep holes with 100 cm distance from each other. Plots had a 2 m distance between them. In all treatments, no fertilizer was added to the soil before and after sowing the seeds and during plant growth. Every plot consisted of 21 plants. Plants were irrigated with 25-40 mm of water weekly when there was no rain. Plants were grown for one year, and in the second year, physiological experiments were begun. Harvest took place in the middle of each month starting from May to November 2019 during an extended season. Extended season allows a crop to grow beyond its normal outdoor growing season and harvesting time frame. In the present study, the season was extended for seven months from May to November.

Total sugars

The measurement of total sugars was performed based on MacCready *et al.* (1950). A volume of 0.1 mL (100 µl) of concentrated alcoholic extract and 100 µL of distilled water

was mixed with 3 mL of anthrone and placed in a water bath (100 °C) for 20 min. When the sample temperature decreased, absorbance was recorded at 620 nm.

Glucose and fructose

Glucose content was determined with the dinitro salicylic acid (DNS) method, by spectrophotometry at 575 nm (Miller 1959). Fructose content was measured by the resorcinal method with spectrophotometry at 520 nm (Ashwell 1957).

Sucrose

The sucrose content was assessed by the method of Miller *et al.* (1959). For this purpose, 1.5 mL of concentrated alcoholic extract with 1.5 mL of dinitro-salicylic acid were mixed and placed in a water bath for 20 min at 90 °C. When the contents of the tube were hot, 0.5 mL of potassium sodium tartrate salt solution (40%) was added to the mixture. After cooling, the spectrum of dark-red solution was determined at 620 nm.

Carotenoids and chlorophyll

Carotenoids and chlorophyll a, b, and a+b content were extracted by acetone 80%, from 1 g of fern. The content of pigments was measured by the method of Arnon (1956), and spectrophotometry (UV/Vis 2100) was at 645 and 663 nm.

Anthocyanin

One g of asparagus tissue was extracted with 10 mL methanol, and incubated overnight at 4 °C in the dark. The slurry was centrifuged at 4000 g for 10 min. The anthocyanin was determined by a spectrophotometer (UV/Vis 2100) at 520 nm (Wanger 1979).

Proline

Proline content was obtained according to the Bates *et al.* (1973). Briefly, 0.5 g of fern was extracted in the 3% sulphosalicylic acid. Proline concentrations of samples were determined using ninhydrin acid in toluene with a spectrophotometer at 520 nm.

Invertase enzyme activity

To measure the the invertase enzyme activity, 5 grams of the fern was homogenized in the cool condition with 20% glycerol and brought to a volume of 100 ml. Then 5 ml of enzyme solution was added into a 100 ml flask and then 10 ml of buffer (1 M sodium phosphate buffer with pH=5) and 5 ml of sugar solution (2.5% sucrose) were added to it and incubated for 24 hours at 37 ^{o}C. After that 3 ml dinitrosalicylic acid reagent was added to the resulting mixture and placed in a bain-marie at the temperature of 100 ^{o}C for 5 minutes. After cooling down the tubes, 1 ml of potassium sodium tartrate was added to them and the amount of enzyme activity was read by spectrophotometry at 510 nm (King *et al*. 1997).

Data analysis

This experiment was conducted in a completely randomized design with two treatments and three replications (10 plants in each plot). The relationships between fern and rhizome carbohydrates and pigments were described with a linear model using mean values for both 2X and 8X genotypes in SAS (v. 9.1) software. Following that, influential assimilates that translocated in asparagus fern and rhizome were described using a stepwise discriminant analysis (Hardle and Simar 2007) in SPSS (ver. 21.0). The original groups were the seven months (May to November) from which the data for different traits were taken for the analyses. The variables with a standardized coefficient of ≥ 0.5 were selected as the most influential variables of asparagus fern and rhizome that contributed to the discriminant functions in explaining the total variation among different months in the growing season.

Results

Chlorophyll and anthocyanin

This experiment was performed in temperate climates with a harvest season of 8 to 10 weeks in each spring and a growing season until November. New spears of asparagus emerge at the end of March and quickly develop into ferns in a temperate climate. Right away the ferns develop, the soluble carbohydrates are formed and stored in the rhizome. According to the results of this study, the accumulation trend of chlorophyll a and chlorophyll b decreased from May to June, and then from June to October, the accumulation trend increased (Figure 1).

The anthocyanin content in the fern was higher than in the rhizome, which indicates that the reducing sugars in the rhizome were less than fern in both 2X and 8X. For anthocyanin, a peak was seen in September and a sharp decline in October, which is probably due to the reduction of non-reducing sugar consumption and its conversion to anthocyanin in October (Figure 2). This confirms the decrease in invertase activity in October in both 2X and 8X (Figure 8).

Based to the results, a positive relationship was observed between rhizome fructose and rhizome anthocyanin in both 2X and 8X asparagus (Figures 2 and 7). The anthocyanin content from July to August was higher in the 8X than 2X rhizome, whereas in June, the carotenoid content in the 2X fern was higher than the 8X ones (Figures 2 and 3). In August, in both 2X and 8X asparagus, the chlorophyll a to b ratio changed. This shows that the ratio a/b = 2 is not always established and is time-dependent (Figure 2). According to Chen *et al.* (2020), the number of chloroplasts and the content of chlorophyll a in the tetraploid Also, chlorophyll b and total chlorophyll were higher at that time than in diploid plants. The highest chlorophyll a and b

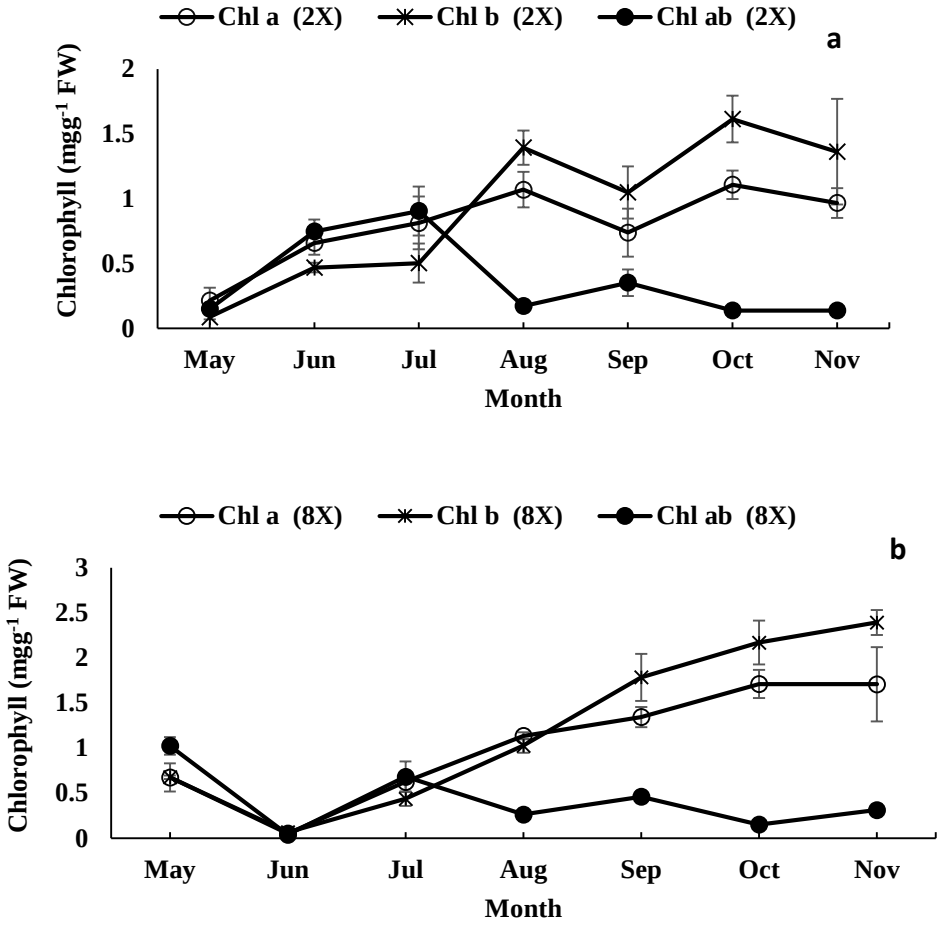

Figure 1. Changes in the chlorophyll content in the rhizome and fern of diploid (a) and octoploid (b) asparagus over an extended season. Vertical bars represent standard errors.

content (0.61 and 1.37 mg.g^{-1}) was recorded in tetraploid compared to diploid asparagus.

Sugars, invertase, and proline

According to the results, the movement of glucose in the 8X asparagus was different from the 2X asparagus (Figure 5). This independent behavior seemingly depended on the internal conditions of the asparagus plant and environment. From May to November, there was a steady decline in the fructose content of the 8X fern and rhizome (Figure 7). In the 8X asparagus, sugars accumulate in the rhizome. The 8X rhizome was thicker and had a storage texture. In the case of 2X asparagus, there was a balance of total sugar between the fern and rhizome, as opposed to the anthocyanin. May and September are two critical stages in the consumption or storage of substances in the 2X asparagus (Figure 4).

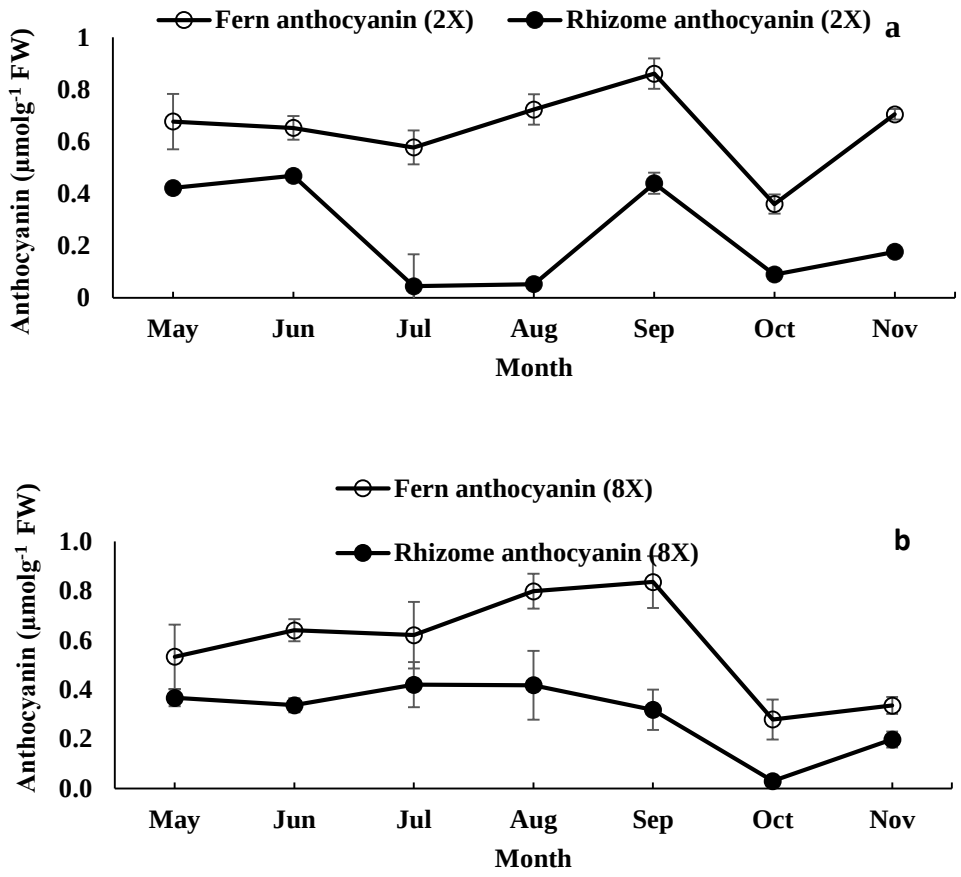

Figure 2. Changes in anthocyanin content in the rhizome and fern of diploid (a) and octoploid (b) asparagus over an extended season. Vertical bars represent standard errors.

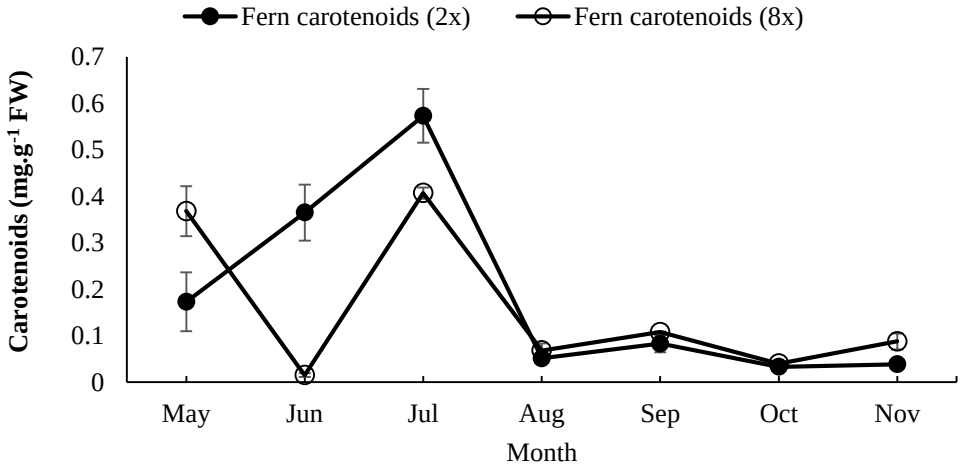

Figure 3. Changes in carotenoid content in the fern of diploid and octoploid asparagus over an extended season. Vertical bars represent standard errors.

Based on our observations, the vegetative growth of the fern was dominant on the rhizome until August and the storage in the rhizome was low. However, shortly after August in the 2X, the growth of the rhizome and fern was the same, and then the fern began to grow. In 8X, the balance between rhizome and fern growth continued until the end of November. In 8X from August to October, and 2X from October to November, total sugar transferred from the fern to the rhizome.

According to the results of this study, the process of invertase activity in both 2X and 8X until September was almost the same (Figure 8), which indicates that invertase activity is essential in maintaining the balance between shoot and rhizome and is a key enzyme in this balance. In 8X, the biochemical behavior of rhizome and fern was balanced, but in diploid, it was not. Invertase activity increased from May to June in the 2X rhizome (Figure 8), which led to energy production and sucrose movement in August (Figure 6).

The regular trend of increasing chlorophyll in 8X is due to increased growth and photosynthetic factors and as a result,

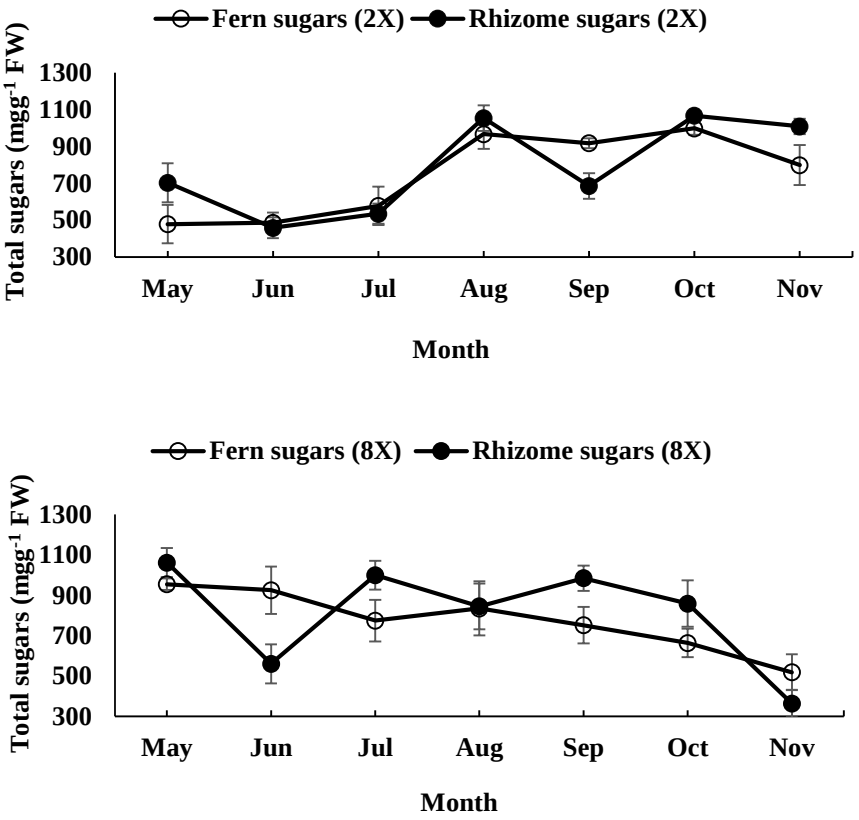

Figure 4. Changes in total sugar content in the rhizome and fern of diploid (a) and octoploid (b) asparagus over an extended season. Vertical bars represent standard errors.

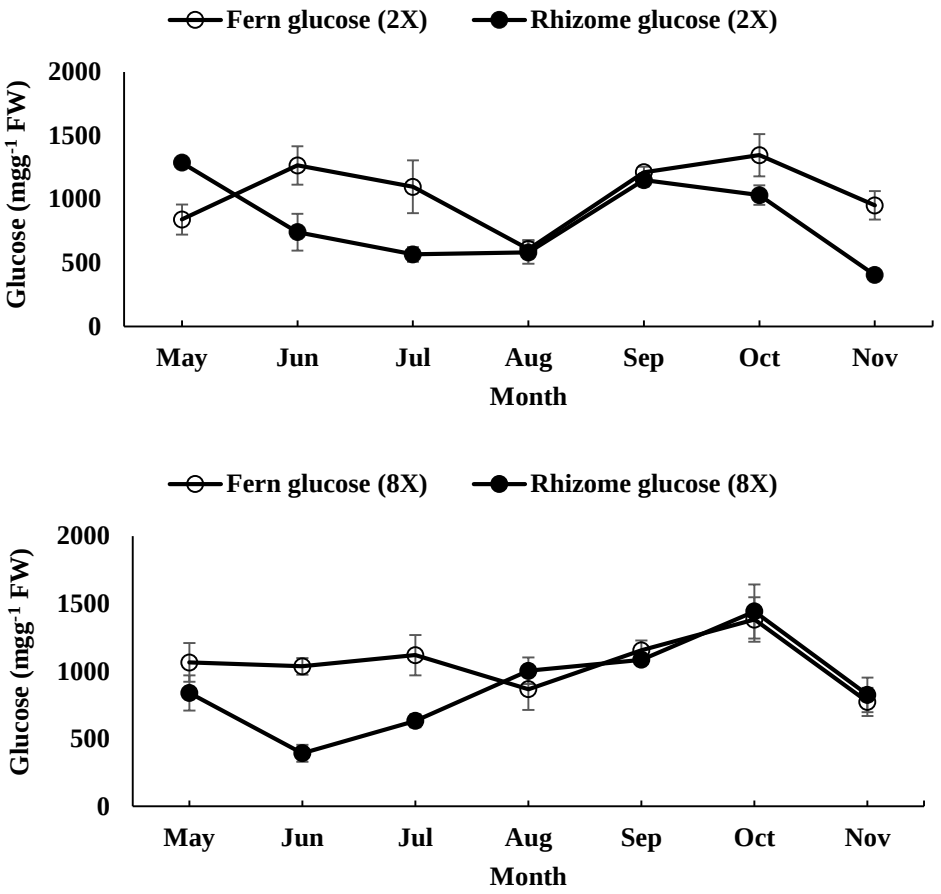

Figure 5. Changes in the glucose content of the rhizome and fern of diploid (a) and octoploid (b) asparagus over an extended season. Vertical bars represent standard errors.

more need for sugars. The growth trend for octoploid continues till November but in 2X, it remains stable from August. In the octoploid, both growth and consumption constantly increased. In August, a decrease in total chlorophyll (Figure 1), glucose (Figure 5), and fructose (Figure 7) was observed, which led to a decrease in plant growth in both 8X and 2X asparagus. Glucose and fructose in 2X asparagus show a sharper decreasing trend than 8X. Probably the stress and heat conditions of August have led to a decrease in the total chlorophyll because in August an increase in the proline content was seen in both 2X and 8X asparagus (Figure 9).

Transfer of sugars towards the rhizomes of 8X asparagus was in balance from May to July and then began to fluctuate with changing physiological conditions. In fact, from July to September, the fern section grows and the rhizome continues to grow until the end of the season (Figures 4 and 7). As the harvesting time prolonged, carbohydrate levels began to decrease again from September (Bhowmik and Matsui 2003). Carbohydrates decline during periods when fern is produced

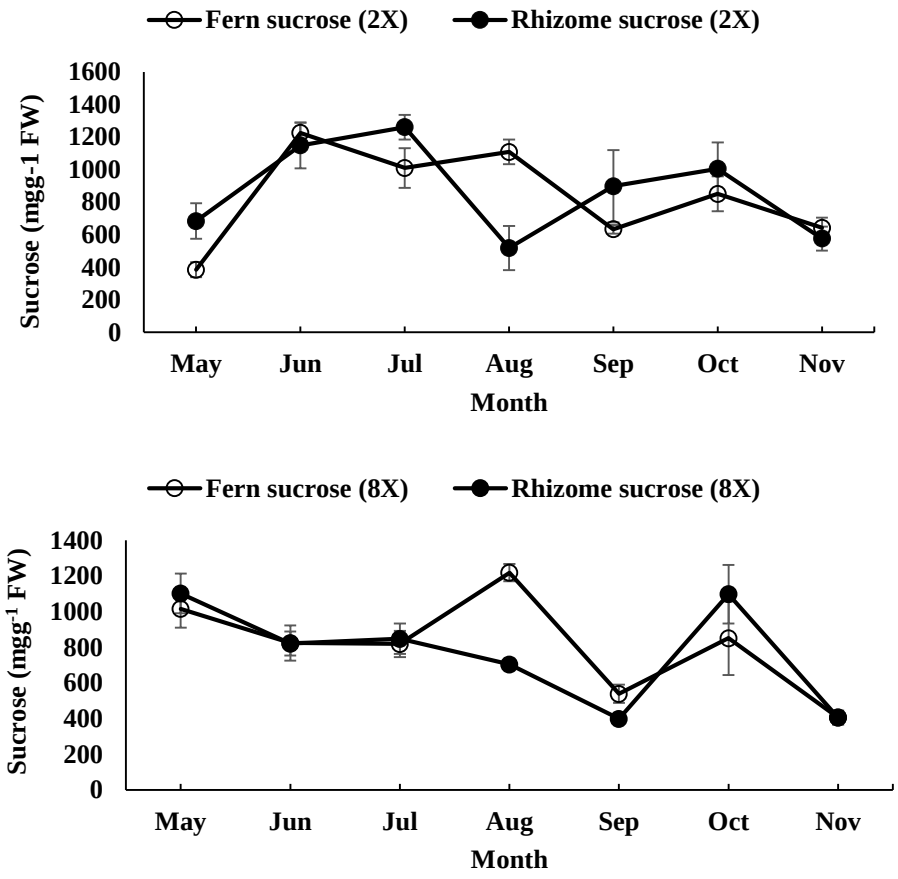

Figure 6. Changes in the sucrose content of the rhizome and fern of diploid (a) and octoploid (b) asparagus over an extended season. Vertical bars represent standard errors.

(Wilson *et al.* 1997). According to Bhowmik and Matsui (2003), root sugar stands unchanged until August and thereafter begins to decrease (Bhowmik and Matsui 2003). There is a relationship between glucose and fructose of spears with soil chemical properties (Takahashi *et al.* 2019). Based on Bhowmik and Matsui (2003), the activities of both the soluble and invertase increased until May and after that, they started to decline. In white asparagus, the impact of the starch and sucrose metabolism, unlike sugar content, was very low (Creydt and Fischer 2020). Shou *et al.* (2007) reported that the highest content of carbohydrates in spears was recorded at low temperatures. Carbohydrates typically are gathered in the root of the asparagus as fructans to produce energy during the following season for spear formation (Creydt and Fischer 2020).

The activity of invertase increased from May to June and after that, it declined in both 2X and 8X. The decline of sucrose content in 8X asparagus was occupied with the high invertase activity (Figure 6b and Figure. 8b). Bhowmik and Matsui (2003) also indicated that the downturn in the sucrose content was related to the rise in the invertase

activity. Increasing the invertase activity indicates that the hydrolysis of sucrose is related to the amounts of glucose and fructose (Takahashi *et al.* 2019).

Proline content increases under stress in plants. The increase in the proline content in 8X was similar in both fern and rhizome, but in 2X, the proline content in the fern increased in September (Figure 9). In many plants yield and stress resistance increase after the induction of polyploidy. Chen *et al.* (2014). reported that heat resistance in the tetraploid asparagus was higher than in the diploid.

Discriminant analysis

For the 2X and 6X asparagus, stepwise discriminant analysis resulted in six and eight variables that finally remained in the discriminant functions in the 2X and 8X asparagus, respectively. Only four variables were common in the discriminant functions of 2x and 8X types of asparagus. The eigenvalues and other related statistics are presented in Tables 3 and 4 for 2x and 8x asparagus, respectively. In both the 2X and 8X asparagus, the six discriminating functions, constructed by assimilates, were able to correctly classify

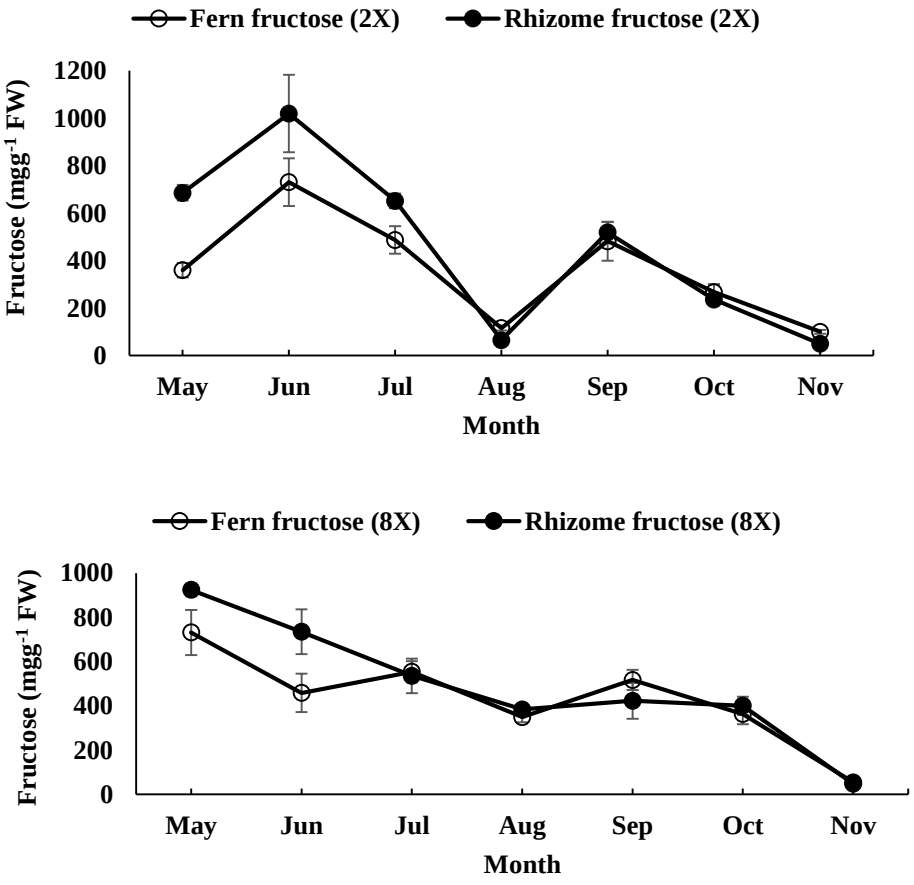

Figure 7. Changes in the fructose content in the rhizome and fern of diploid (a) and octoploid (b) asparagus over an extended season. Vertical bars represent standard errors.

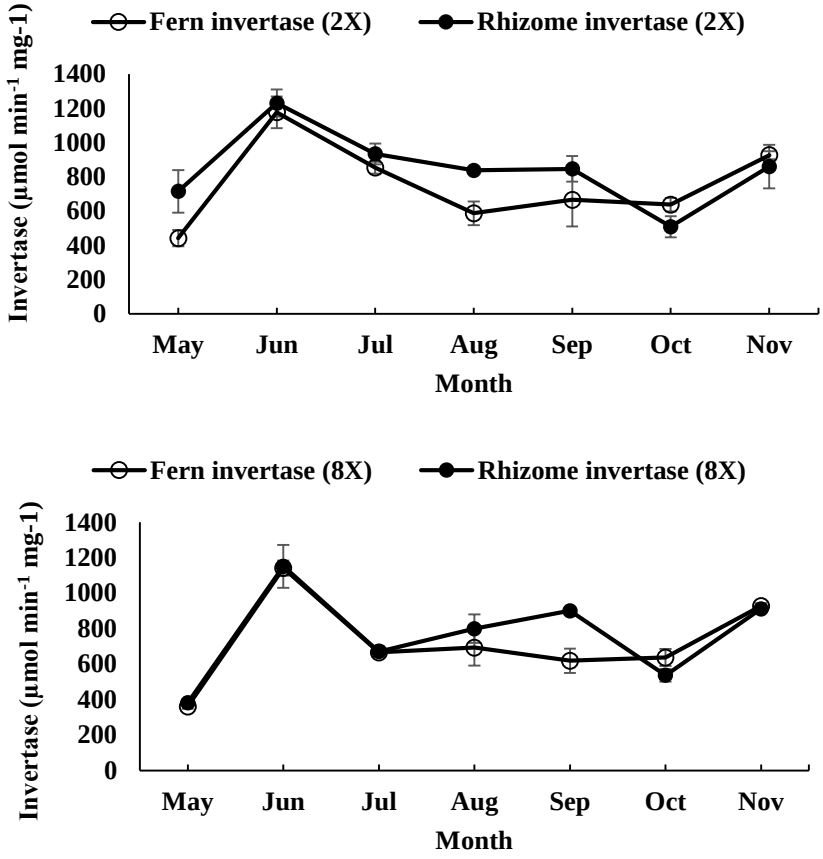

Figure 8. Changes in the invertase content in the rhizome and fern of diploid (a) and octoploid (b) asparagus over an extended season. Vertical bars represent standard errors.

100% of the original grouped cases. In the 2X asparagus, the first two functions significantly explained 88.2 and 8.1 percent of the total variation among the months. Since these two functions explained 96.3 of the total variation, they were selected to determine the most influential variables contributing to the differential responses of the asparagus in different months of the growing season. The pattern was different in the 8X asparagus. The first two functions significantly explained 36.5 and 32.4 percent of the total variation among the months (Summing to 68.9%). Therefore, they were selected to determine the most influential variables in each discriminant function.

The discriminant functions permit the comparison of important variables on 2X and 8X asparagus growth. The variables with the highest coefficients indicate their higher discriminating ability. Table 5 shows the standardized canonical discriminant function coefficients for the 2X and 8X asparagus. Since in the 2X asparagus, the first function explained 88.2% of the total variation among months, it seems that the rhizome fructose, fern carotenoid, and rhizome glucose, contributed most to the discrimination among

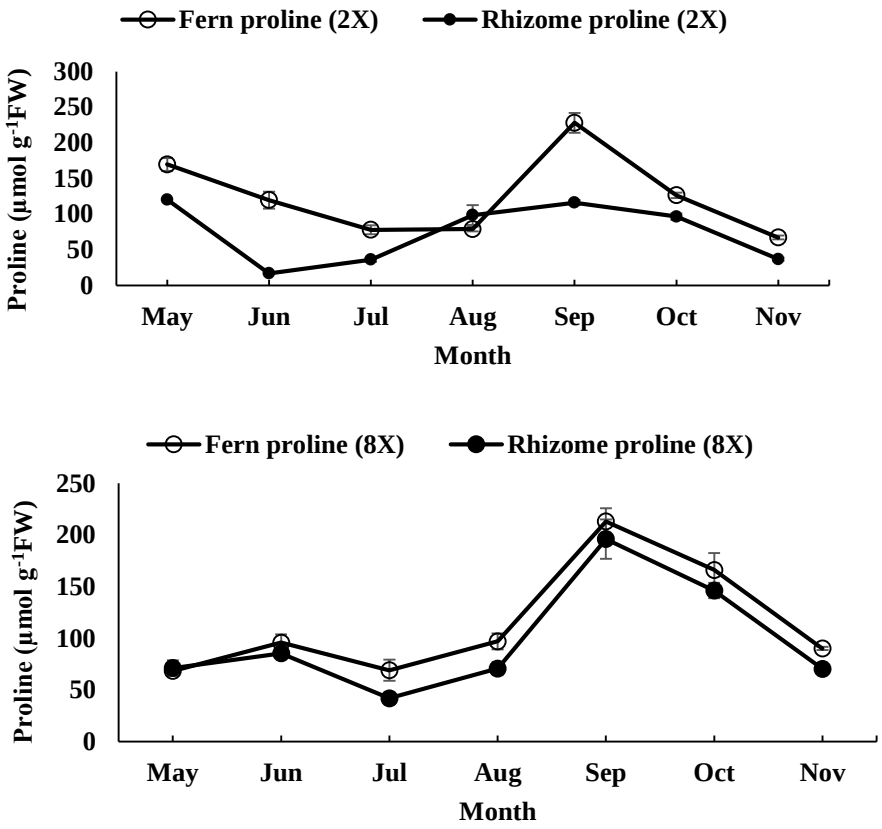

Figure 9. Changes in the proline content in the rhizome and fern of diploid (a) and octoploid (b) asparagus over an extended season. Vertical bars represent standard errors.

months (Table 5). However, the rhizome glucose had the opposite sign compared to the rhizome fructose and fern carotenoid. It means that the increase in rhizome glucose corresponds to the decrease in rhizome fructose and fern carotenoid of the 2X asparagus. The fructose required by the rhizome is always accompanied by the fern change. From July to September, a movement of glucose to the rhizome was observed in both 2X and 8X (Figures 4 and 5). The 8X asparagus pattern was different than 2X and the contribution of the two first functions to the total variation was almost similar. Therefore, taking the two functions together, it seems that the rhizome sucrose, rhizome total sugars, fern fructose, fern anthocyanin, fern invertase, and fern sucrose contributed most to discrimination among months. However, the rhizome sucrose had an opposite sign to the other above-mentioned influential characteristics and its increase was associated with the decrease in rhizome total sugars, fern fructose, fern anthocyanin, fern invertase, and fern sucrose (Table 4). The total sugars and glucose in the rhizome had a decreasing trend from May to June in both 2X and 8X (Figures 4 and 5). Bhowmik and Matsui (2003) reported that

sucrose, glucose, and fructose declined from March to May as the new shoots in asparagus ('E414' and 'Welcome' varieties) developed. Changes in fructose followed total sugars. According to Anastasiadi *et al.* (2020), changing of reducing sugars (glucose and fructose) to form sucrose was observed during the storage of asparagus spears.

The difference in the response of the two types of asparagus in terms of the assimilate transfer during the growing season can be emphasized also in Figure 10.

Table 3. First six canonical discriminant functions, which were selected by the stepwise discriminant procedure for use in the 2X asparagus analysis.

Function	p-value	Eigenvalue	% Variance	Cumulative %
1	0.0001	510.1	88.2	88.2
2	0.0001	46.7	8.1	96.3
3	0.0001	10.6	1.8	98.1
4	0.0001	6.4	1.1	99.2
5	0.0001	2.8	0.5	99.7
6	0.0002	1.7	0.3	100

100% of the original grouped cases were correctly classified.

Table 4. First six canonical discriminant functions, which were selected by the stepwise discriminant procedure for use in the 8X asparagus analysis.

Function	p-value	Eigen value	Variance %	Cumulative variance %
1	0.0001	134.8	36.5	36.5
2	0.0001	119.7	32.4	68.9
3	0.0001	81.6	22.1	90.9
4	0.0001	21.8	5.9	96.8
5	0.0001	9.1	2.5	99.3
6	0.001	2.6	0.7	100

100% of the original grouped cases were correctly classified.

Table 5. Standardized canonical discriminant function coefficients for the 2X and 8X asparagus.

2X			8X		
Variable	Function		Variable	Function	
	1	2		1	2
Fern anthocyanin	-0.241	-0.507	Fern anthocyanin	0.256	1.391
Fern carotenoid	1.497	0.245	Fern carotenoid	0.719	-0.748
Fern sucrose	0.800	-0.605	Fern sucrose	1.057	0.085
Rhizome fructose	2.169	0.055	Rhizome fructose	-0.661	-0.599
Fern glucose	0.630	0.707	Fern fructose	0.380	2.035
Rhizome glucose	-1.082	0.928	Rhizome sucrose	-1.673	-1.304
-	-	-	Rhizome total sugars	1.460	1.129
-	-	-	Fern invertase	-0.522	1.134

The coefficients ≥ 0.5 were regarded as the influential variables in the related function.

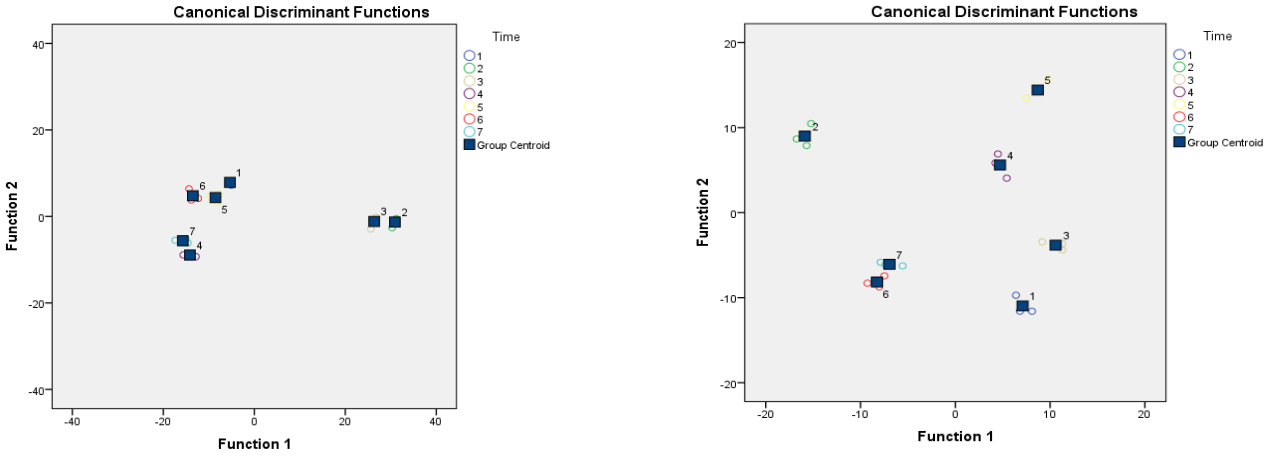

Figure 10. Distribution of the different months in terms of first and second canonical discriminant functions about the fern and rhizome assimilates of asparagus.

Conclusion

Based on the results of the present experiment, the accumulation of dry matter and stored carbohydrates in the fern and rhizome of asparagus was affected by seasonal temperature. With increasing temperature at the beginning of the growing season, the accumulation of dry matter and carbohydrates stored in spear and rhizome increased. Sucrose transferred from fern to rhizome from August to October in 2X and 8X. As the temperature dropped and the plant entered the dormant stage, the accumulation of dry matter and carbohydrates in the spear and rhizome also decreased, as at this stage soluble carbohydrates converted to insoluble compounds. Total sugars in the rhizome decreased from May to June in the 2X and 8X asparagus. The negative relationship between invertase and sucrose indicated that in asparagus, rhizome invertase only takes in the breakdown of sucrose. Invertase activity increased from May to June and after that declined in both 2X and 8X. The growth of fern was dominant to the rhizome until August and the loading of glucose in the rhizome was low. The results indicated that different types of asparagus ploidy have different patterns in the transfer of assimilates from fern to rhizome. The discriminant functions were capable of separating the response pattern of the 2X and 8X asparagus.

By conducting this research, total sugars, sucrose, glucose, and fructose transfer patterns in diploid and octoploid asparagus were determined, which can be used for nutrition and field management of asparagus growers. The transfer pattern of carbohydrates, pigments, and enzymes could be measured in the third year at the beginning of harvesting.

Author contribution

BE collected samples from natural regions and performed experiments. SJM drafted the manuscript and acted as the corresponding author. KM participated in the design and coordination of the study. All authors read and approved the final manuscript.

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Conflict of interest

The authors declare that they have no competing interests.

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انتقال آسمیلات‌ها بین ریزوم و اندام هوایی مارچوبه دیپلوئید و اکتاپلوئید در طول فصل رشد

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چکیده

مارچوبه (*Asparagus officinalis* L.) یک سبزی چند ساله با سطوح پلوئیدی متفاوت است. جابجایی مواد جذب شده در مارچوبه دیپلوئید (2X) و اکتوپلوئید (8X) بین ریزوم و اندام هوایی باید درک شود. انتقال قندهای محلول، رنگدانه‌ها و فعالیت اینورتاز در مارچوبه دو ساله از ماه می تا نوامبر ۲۰۱۹ مورد مطالعه قرار گرفت. ساکارز و گلوکز کربوهیدرات‌های محلول اصلی در ریزوم مارچوبه بودند. در مورد مارچوبه 2X برخلاف آنتوسیانین، تعادلی از قند کل در اندام هوایی و ریزوم وجود داشت. می و سپتامبر ماه‌های مهمی در مصرف یا نگهداری مواد در مارچوبه 2X بودند. ساکارز از اوت تا اکتبر در 2X و 8X از اندام هوایی به ریزوم منتقل شد. کل قند موجود در ریزوم از ماه می تا ژوئن در 2X و 8X کاهش یافت. فعالیت آنزیم اینورتاز از ماه می تا ژوئن افزایش یافت و پس از آن در هر دو مارچوبه 2X و 8X کاهش یافت. رشد اندام هوایی تا مرداد ماه بر ریزوم غالب بوده و بارگیری گلوکز در ریزوم کم بود. تجزیه تابع تشخیص تفاوت در الگوی پاسخ مارچوبه 2X و 8X را نشان داد زیرا ریزوم فروکتوز، کاروتنوئید اندام هوایی و گلوکز ریزوم در مارچوبه 2X ولی ساکارز ریزوم، قندهای کل ریزوم، فروکتوز اندام هوایی، آنتوسیانین اندام هوایی، اینورتاز اندام هوایی و ساکارز اندام هوایی تاثیرگذارترین متغیرها در این دو نوع مارچوبه بودند. الگوهای انتقال قندهای کل، ساکارز، گلوکز و فروکتوز در مارچوبه دیپلوئید و اکتوپلوئید را می‌توان در مورد مدیریت تغذیه توسط زارعین مارچوبه استفاده کرد.

واژه‌های کلیدی: اینورتاز، پلوئیدی، ساکارز، قند کل، کربوهیدرات