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Carbohydrate Content and It's Roles in Alternate Bearing in Olive

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Abstract: Changes in the soluble carbohydrate content of olive were determined during fruit ripening in on-and off-years. Soluble sugars increased up to 90 days after fruit set and decreased during fruit ripening. In on-year, a marked temporary increase was evident at the beginning of fruit color changes. Glucose, fructose and mannitol were the main sugars of soluble fraction of fruits. The content of glucose was higher than the other sugars in fruits. In leaves, mannitol, glucose and fructose were the major components of soluble sugars. Mannitol was higher than the other sugars and the content of mannitol in off-year was lower than that of on-year.

Key words: Oive, carbohydrate, alternate bearing

INTRODUCTION

Alternative bearing is a spread wide phenomenon, occurring in both deciduous and evergreen trees (Ulger, 2004; Goldschmidt, 2005). Flower induction in olives starts during late winter and requires winter chilling and the presence of leaves (Monselise and Goldschmidt, 1982). Olive has a very marked alternative bearing pattern (Lavee, 1996; Seyyednejad, 2001). Studies on changes in carbohydrate components of leaves during on (bearing) and off (non-bearing) years cycle have shown that, sugars are much higher at the beginning of a on-than of a off-year (Seyyednejad, 2001). Competition between vegetative and reproductive organs during the on-year causes reduced production of new branches, producing a smaller number of flowers. The strong growth in the off-year again allows large amounts of flower to be initiated the next year (Monselise and Goldschmidt, 1982).

Changes in soluble carbohydrate composition of leaf and fruit tissues have been previously investigated, that showed, in leaves, mannitol, glucose and fructose are the major components of alcohol soluble sugars, with a different pattern during on- and off- years (Seyyednejad *et al*., 2001; Ebrahimzadeh *et al*., 2000).

However, studies have not been done during the fruit development in on-and-off years in Khoozestan province. Therefore the objective of present study was to examine the changes of components of soluble carbohydrates in the olive leaves and fruits during fruit development in on- and off-years in order to gain more information about the physiology of olive tree and investigate the possible roles of carbohydrates in alternate bearing in different conditions.

MATERIALS AND METHODS

Leaf and fruit samples from tree, Manzanillo and Dezfull cv.s grown in Khoozestan, Iran, were prepared during fruit ripening at 3 month intervals from 75 to 165 days after fruit set, from region of Safi abad garden of Dezfull in Khoozestan province at 2001 and 2002 years. The samples were lyophilized for 48 h, powdered in omni mixer and maintained at -20°C. For determination of sugars, 1 g of powder was extracted repeatedly using 10 mL of 80% ethanol, centrifuged and the supernatant collected (Patumi *et al*., 1990).

Phenol-sulfuric acid method (Dubois *et al*., 1956) was utilized for determination of total sugar content. The monosaccharides were also analyzed quantitatively and qualitatively by GLC of their TMS ether derivatives (Sweeley *et al*., 1963).

In this method up to 10 mg of standard sugars were treated with 2 mL of anhydrous pyridine, 1 mL of Hexamethyldisilazane (HMDS) and 1 mL Trimethyl chlorosilane (TMS).

The reactions were carried out in plastic stoppered vials. The mixture was shaken vigorously for about 30 sec and put in ban Mary at 60°C for 1h, then allowed to stand for 5 min or longer at room temperature, prior to chromatography. One microliter samples was then injection into a gas chromatograph with flame ionization detector (Shimadzu GC 17-A) for analysis. A 50% phenyl-polysilphenylene-siloxane capillary column was used under the following temperature program condition: Column 140-160°C for 5 min, 160-180°C for 8 min, 180-250°C for 7 min, 250°C for min, detector and injector: 250 and 200°C, respectively. Carrier gas was N_2 with a flow rate of 77 bars.

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RESULTS

Changes in the content of soluble sugar in leaves and fruits after fruit set during on- and off-years are shown in Table 1.

Content of soluble sugars during fruit development increased up to 90 days after fruit set and decreased subsequently. In on-year, a marked temporary increase in sugars was noticed at the beginning of fruit color change. The content of total sugars in off-year was higher than that of on-year.

Glucose, fructose and mannitol were the main constituents of alcohol soluble sugars of fruits during ripening both in on-and off-years (Nergiz and Engiz, 2000) (Table 2). This is accordant with other researches (Lavee, 1988; Seyyednejad, 2001; Ulger, 2004; Rinaldi *et al*., 1994).

Besides, the content of glucose was higher than that of fructose and mannitol, the result that has also been reported previously (Fernandez and Diez, 1971; Ebrahimzadeh *et al*., 2000; Marsilio *et al*., 2001). Glucose and fructose vary to each other in fruits during ripening in on-year. Furthermore during the decrease of glucose and fructose, the amount of mannitol increased.

In leaves, the amount of fructose was very low and between the two other sugars, glucose and mannitol which are the main monosaccharides of alcohol soluble fraction, there were a negative relationship both in on-and off-years and the mannitol was the most abundant, especially in on-year. This is accordant with previous our researches (Seyyednejad *et al*., 2001).

In general, the fruit is fed by translocation of metabolites produced in the leaves. The accumulated reducing sugars which were found in the fruits (glucose and fructose) are not translocated in the phloem of higher plants. It has been shown, however, that in some plants the polyols serve as translocatable carbohydrates, in addition to sucrose and other non-reducing oligosaccharides (Wonder and Lavee, 1988). Thus the mannitol in the olive fruit as well as mannitol and other polyols in the fruits of many other plants groups might be of specific importance in the metabolic transformation and synthesis of specific storage material.

Our experiments showed that, during fruit development (from 75 to 135 days of fruit set), the amount of sugars was similar among the fruit in on- and off-years, but in leaves glucose and fructose were lower and mannitol was higher in on-year. In the later stages (from 135 to 165 days of fruit set), the amount of mannitol increased in fruit following the augmentation of this sugar in the leaves.

The level of soluble and insoluble carbohydrates in photosynthesizing leaves are principally the result of balance between the rate of carbon assimilation and the rate of carbon exported, under the influence of the various sinks. The balance between the carbon assimilated in leaves and the carbon for export is regulated by two different mechanisms (Drossopoulos and Niavis, 1988; Seyyednejad *et al*., 2001). Our results showed that, the content of these compounds in leaves was higher in off-year. Thus, we deduce that the rate of carbohydrate transport in off-year is low. As the carbohydrate content of the fruit similar in both on-and off-years, we can conclude that the new shoots lack the capacity for carbohydrate mobilization. This observation has previously been reported for apple (Monselise and Goldschmidt, 1982).

Table 1: Total soluble sugars in leaf and fruit

	Time	Leaf							Fruit						
Samples	(days after fruit set)	75	90	105	120	135	150	165	75	90	105	120	135	150	165
cv. Manzanillo	On-year	5.3	5.5	4.0	2.0	2.8	3.2	2.2	5.3	6.5	5.3	4.7	3.0	3.5	3.0
	Off-year	5.8	5.3	4.9	5.1	3.7	3.9	4.5	5.7	6.3	5.5	5.0	3.0	3.6	2.8
cv. Dezfull	On-year	6.3	5.2	4.9	2.4	3.7	4.1	2.9	6.3	6.5	6.0	5.5	4.0	4.4	4.0
	Off-year	6.7	6.3	5.8	6.2	4.2	4.5	5.0	6.2	6.5	6.1	5.8	3.4	4.6	4.2

Table 2: Changes in content of soluble sugars in leaves and fruits

	Leaf						Fruit					
	Sample											
Time	Manzanillo			Dezfull			Manzanillo			Dezfull		
(day after fruit set)	Glucose	Fructose	Mannitol	Glucose	Fructose	Mannitol	Glucose	Fructose	Mannitol	Glucose	Fructose	Mannitol
75	26.29 (37)	26.33 (2.71)	37.77 (19)	34 (29.18)	1.31 (13.96)	60 (33.81)	30.56 (49)	2.99 (15.33)	17.32 (1.93)	46.2 (51.57)	19 (1.44)	18.8 (39.4)
90	18 (31.72)	18.64 (3.54)	38.39 (25)	25 (22.77)	1.6 (10)	66 (43)	36.51 (52)	2.77 (18.91)	20.75 (3.59)	50 (52.94)	30 (0.92)	13 (37.09)
105	33.96 (38.57)	26.85 (0.87)	29.17 (22)	42 (27.46)	1.05 (7.59)	45.7 (38.6)	32.37 (56.69)	3.4 (4.22)	25 (4.09)	59.37 (52.93)	18.7 (1.44)	12.58 (39.4)
120	29 (21.31)	10.1 (1.42)	35 (32.87)	30 (60)	13.28 (6.12)	55 (27)	56.99 (41.82)	11.58 (30.23)	16.33 (22)	44.24 (56.78)	34.16 (3.47)	3 (37.06)
135	21.39 (25.11)	5.53 (6.35)	40.51 (31)	20 (62.78)	3.23 (10.94)	67 (24.98)	36.84 (42.23)	27.35 (31)	17.45 (19.61)	56.14 (48.6)	16.24 (15.75)	17.23 (14.52)
150	23.73 (30)	2.22 (10.34)	37.16 (29)	10 (68.31)	2.44 (13.71)	75 (20)	37.02 (27.43)	28 (36.44)	23.34 (20.86)	29.21 (45.82)	12 (23.28)	19 (19.35)
165	11 (37.1)	1.43 (7.87)	52.16 (25)	5 (59.48)	0.5 (11.11)	80 (28.46)	44 (46.21)	33.22 (17.31)	15.91 (37.59)	18 (45.7)	9 (22.37)	30 (12.6)

Number in parenthesis is the values in off-year

Table 3: Test statistics of different kind of sugars

		Soluble sugar	Glucose	Fructose	Mannitol
Most etreme differences	Absolute	0.29	0.36	0.21	0.50
	Positive	0.29	0.36	0.14	0.50
	Negative	0.00	-0.14	-0.21	0.00
Kolmogorov-smirnov Z		0.76	0.95	0.57	1.33
Asymp.Sig.(2tail)		0.62	0.33	0.91	0.05

Table 4: Mean values of different kind of sugar in leaf and fruits

		Mean			
Kind of sugar	Frequencies	t	Leaf	Fruit	Sig
Soluble sugar	28	4.51	4.88	-1.08	0.29
Glucose	28	31.43	44.54	-3.71**	00.0
Fructose	28	7.89	16.80	-3.47**	0.00
Mannitol	28	39.94	19.96	5.29**	0.00

Table 5: Mean±SD of different kind of sugars in on and off years

		Mean		SD			
Kind of sugar	Frequencies	Off	On	Off	On	t	Sig
Soluble sugar	28	5.02	4.38	1.12	1.38	1.93*	0.05
Glucose	28	43.59	32.39	13.15	14.17	3.07**	0.00
Fructose	28	11.74	12.96	9.84	11.32	-0.43	0.69
Mannitol	28	25.31	34.58	11.27	20.88	-2.07*	0.04

Since the level of certain elements in off-year is lower than that of the on-year (Villemur, 1984) the decrease in mannitol transport could be attributed the low level of these elements. The biosynthetic pathway of mannitol in *Apium graveolens* (Umbeliferae) has already been established (Simirnoff, 1995).

Thus based on the present result in our study, we conclude that the same pathway is used during the on-year and mannitol is accumulated. But during off-year, the amount of glucose is more than that of mannitol, this pathway not be operative. The possible pathway could be the conversion of fructose-6-P to glucose-6-P and free glucose.

Moreover, for improve of our results, we did some statistic tests.

Table 3 shows that, in leaf organ, only mannitol have significant difference in two varieties. This shows that translocated sugar from leaf to fruit is mannitol.

Table 4 shows that there are significant differences between mean of glucose, fructose and mannitol in two organs (leaf and fruit). The result of Table 5 shows that there are significant difference between amount of soluble sugar and mannitol in two years, also, mean of glucose in two years have significant difference. Amount of soluble sugar and glucose in former year is higher than later year, but amount of mannitol in later year is higher than former year, this shows that translocation of this sugar to fruit disorder in off-year.

So, on the basis of present results, there are two hypothesis for possible role of carbohydrate in alternate bearing. One of them is transport speed of manitol reduce from leave to fruit in off-year and cause to accumulate manitol in leave. The other is transformation glucose to fructose and manitol disorder in off-year.

More over there, comparisons of these results with previous results on some cultivars from different wether condition, show that, climate does not have important effect on alternate bearing.

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