Vitis 47 (3), 191–192 (2008)

Research Note

Tetraploid sucrose-accumulating grapevines

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K e y w o r d s : HPLC, sucrose accumulator, sugar composition, tetraploid.

Introduction: Fructose and glucose are primary sugars of grape (*Vitis* spp.), and sucrose presents in trace amounts (Winkler *et al*. 1974, Shiraishi 1993, Davis and Robinson 1996). However, some American hybrid cultivars such as 'Bath', 'Buffalo', 'Fredonia' and 'Steuben' contain large amounts of sucrose (Lott and Barrett 1967, Takayanagi and Yokotsuga 1997; Shiraishi 2000). The early ripening and strong, sweet taste of these diploid sucrose-accumulating cultivars are of great interest in table grape breeding to broaden variability among seedlings. However, the diploid berries are in general smaller than the tetraploid ones, and consumers' acceptance of large-sized grapes is high. Polyploid breeding has been very successful in Japan since 1970, and the tetraploid cultivars with large berries offer the possibility of creating unique table grapes with a striking appearance (Reisch and Pratt 1996). Many tetraploid genotypes have also been introduced by crossbreeding and *in vitro* colchicine treatment of diploid cultivars in our breeding program at Fukuoka Agricultural Research Center, Japan (Notsuga *et al*. 2000). As breeding materials, these tetraploid genotypes are valuable additions to the gene pool for developing future table grapes. By screening sugar variants, the tetraploid genotypes were generally found to be hexose accumulators (Fig. 1 A), but some tetraploid sucrose accumulators were observed as follows (Fig. 1 B): 'Buffalo 4X' (TSA-1), 'Steuben 4X' (TSA-2), 'Red Port 4X' (TSA-3), and Seedling 11-7 (TSA-4). In this report we document the origins of TSA-1,2,3 and -4, and describe their agro/biochemical traits between 2006 and 2007.

Materials and Methods: Four tetraploid sucrose-accumulating genotypes, TSA-1 to -4 (*Vitis labruscana* × *V. vinifera* interspecific hybrids) were grown in a greenhouse between 2006 and 2007 at the Fukuoka Agricultural Research Center, Fukuoka, Japan. Own-rooted, one vine per genotype was used and trained onto a horizontal trellis system. The following agro/biochemical traits were measured during growth: time of full maturity, single berry weight, skin color, soluble solids content (SSC), acidity, total sugar content, and sugar composition. Two to three bunches in each genotype were selected. Three berries per bunch were sampled, and were selected from the top, middle, and bottom of the bunch. After microwave heat treatment for 20 s, the juices were extracted from the berries by hand pressing and centrifuged at 5000 × *g* for 10 min. SSC was measured by a hand refractometer and expressed as °Brix. Acidity was determined by potentiometric titration. Sugar composition was determined using a Shimadzu LC-10A HPLC system equipped with a RID-10A refractive index detector. Juice (1 ml) was diluted to 20 ml with deionized water and filtered through a 0.45 µm filter. The column (Cosmosil Sugar-D, 4.6 × 250 mm, Kyoto, Japan) was operated at 42 ºC. The mobile phase was a mixture of acetonitrile and water (75:25, v/v), and the flow rate was 1.0 ml·min^{-1}. The injection volume was 20 µl.

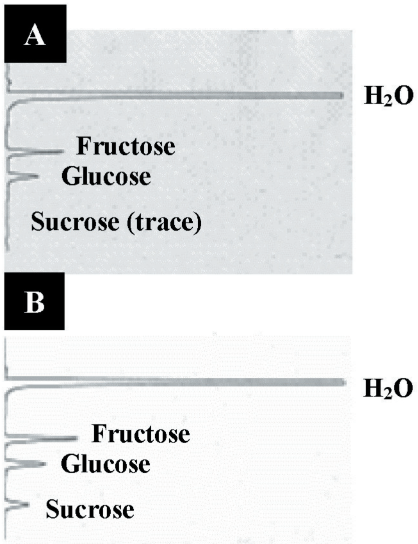

Fig. 1: Typical HPLC profiles of sugar composition in tetraploid grape genotypes. **A**: Hexose accumulator (cv. Kyoho) **B**: Sucrose accumulator (TSA-4: breeding line 11-7).

Results and Discussion: O r i g i n : TSA-1 to -3 were induced between 1984 and 1990 via *in vitro* chromosome doubling of the diploid sucrose-accumulating cultivars 'Buffalo', 'Steuben', and 'Red Port', respectively (Fig. 2).

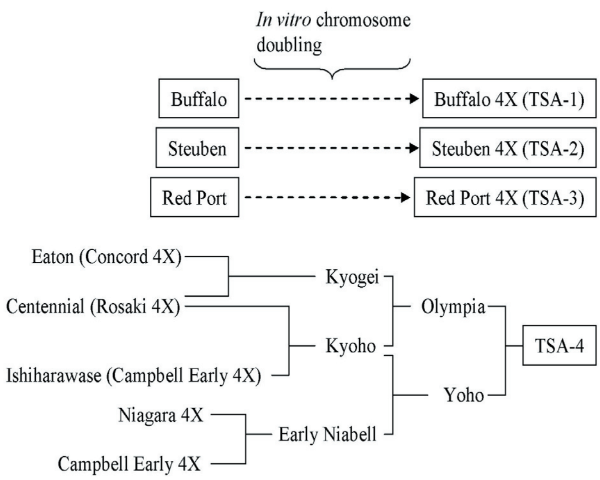

Fig. 2: Pedigrees of sucrose-accumulating tetraploids, TSA-1, TSA-2, TSA-3 and TSA-4. ▭: Sucrose accumulator.

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TSA-4 was introduced in 1990 from an interspecific cross between 'Olympia' and 'Yoho' (Fig. 2). It is interesting that the occurrence of TSA-4 resulted from successive parental crosses between hexose accumulators. Inheritance of sucrose accumulation among diploid- and tetraploid-grapevines is not clear, but genetic experiments are ongoing in our laboratory. Of the seven seedlings derived from 'Olympia' × 'Yoho', two plants alone were sucrose accumulators, and the remaining five were hexose accumulators. In a population of 23 F_2 population (self-seedlings of TSA-4), fifteen plants were fruited and all sucrose accumulators. These results suggest that the trait of sucrose accumulation may be controlled by recessive genes. In 2007, a total of 1111 seeds was obtained by self-pollination of TSA-4 for further genetic analysis of F_2 population.

A g r o / b i o c h e m i c a l t r a i t s : The Table shows comparison of agro/biochemical traits between tetraploid sucrose-accumulating grape genotype and its source cultivar. TSA-1 and TSA-4 reached full maturity in early August, and TSA-2 and TSA-3 in mid-August. Single berry weight ranged from 5.1 g (TSA-1) to 9.2 g (TSA-4) with a mean of 6.7 g. The berry weights of induced autotetraploids (TSA-1 to -3) were 1.3 to 1.5 times greater than those of source diploid cultivars, agreeing with a previous report (NOTSUKA *et al*. 2000). The allotetraploid TSA-4 is a more promising breeding material for production of large-sized berries with a maximum berry weight of 10.5 g. All genotypes had well-colored bunches, and skin color was blue-black, except for TSA-3 that was red. SSC ranged from 15.0 °Brix (TSA-3) to 18.4 °Brix (TSA-4) with an average of 17.4 °Brix. A similar trend was found in the total sugar content. Acidity ranged from 0.48 % (TSA-2) to 0.67 % (TSA-3) with an average of 0.54 %. On a percentage basis, fructose and glucose presented in an approximately 1:1 ratio. Sucrose varied from 18.8 % (TSA-3) to 22.2 % (TSA-4) with an average of 20.4 %. In their review of sugar composition, LOTT and BARRETT (1967) indicated that the sucrose content of nine diploid sucrose accumulators varied from 15.2 % (Bath) to 32.6 % (Sweet Blue), and averaged 25.5 %. Sucrose content of tetraploids was not always higher than that of diploids, suggesting that the effect of polyploidy is not clear for sucrose accumulation in grape berries. Progeny tests revealed that tetraploid sucrose-accumulating genotypes were essentially tetraploids in their breeding behavior. These genotypes with high sucrose content, which are early- to mid-season in maturity, are suitable for developing tetraploid table grapes.

T a b l e

Comparison of agro/biochemical traits between tetraploid sucrose-accumulating grape genotype and its source cultivar (in bracket)

Sucrose accumulator	Time of full maturity	Berry weight (g)	Skin color	SSC (°Brix)	Acidity (%)	Total sugar content (g·100 ml^{-1} juice)	Sugar composition (%)		
							Fru.	Glu.	Suc.
TSA-1	Early Aug.	5.1	Blue-black	18.1	0.50	16.4	41.0	39.5	19.5
(Buffalo)	Early Aug.	4.0	Blue-black	19.0	0.52	17.1	40.7	38.3	21.0
TSA-2	Mid. Aug.	6.3	Blue-black	17.9	0.48	16.2	42.0	36.8	21.2
(Steuben)	Late Aug.	4.2	Blue-black	17.2	0.46	15.2	38.8	38.2	23.0
TSA-3	Mid. Aug.	5.9	Red	15.0	0.67	13.3	39.7	41.5	18.8
(Red Port)	Mid. Aug.	4.0	Red	15.2	0.63	13.9	45.3	42.4	12.2
TSA-4	Early Aug.	9.2	Blue-black	18.4	0.50	17.5	39.0	38.8	22.2
(Olympia)	Late Aug.	12.0	Red-brown	18.0	0.45	16.5	49.1	50.9	trace
(Yoho)	Mid. Aug.	8.2	Red	17.0	0.57	15.5	50.8	49.2	trace

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Received December 10, 2007