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Review

Metabolism and Accumulation of Sugars Translocated to Fruit and Their Regulation

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Photoassimilates needed for fruit development are supplied from leaves, converted in fruit to substances relating to the specific quality of the fruit, then accumulate in the fruit. There are various regulation steps in the process from photoassimilate synthesis in leaves to sugar accumulation in fruit: photosynthesis, synthesis of translocation sugars, loading of translocation sugars, their translocation, their unloading, their membrane transport, their metabolic conversion, and compartmentation in vacuoles. Thus, it is important to clarify the mechanism and regulation of each step in fruit development. In this review, mainly the metabolic conversion of translocation sugars and their regulation at the genetic level in fruit are described because the metabolic conversion in fruit contributes greatly to produce the sink activity needed for fruit development.

Key Words: fruit, sink activity, sorbitol, sucrose, sugar metabolism.

Introduction

Photoassimilate in fruit depends mainly on supply from leaves, but some fruits in their early stages of development can supply it by their own photosynthesis. Fruit is a heterotrophic organ. However, specific substances in some fruits are generated in the fruit by themselves from photoassimilates etc., and can accumulate in fruit. Therefore, they depend on sugars translocated from leaves. The force attracting translocation sugars in fruit is called the sink strength. If the sink strength of the fruit is weak, the fruit can not grow sufficiently: growth is retarded or sometimes the fruit shrinks because of sugar deprivation. This review discusses the physiological conditions influencing sink strength during the various steps between photoassimilate synthesis in leaves and its accumulation in fruit (Fig. 1).

1. Photosynthesis: Photoassimilate is synthesized by photosynthesis in leaves. 2. Synthesis of translocation sugars: Photoassimilate is converted to translocation sugars, such as sucrose and sorbitol. 3. Loading: Sucrose and sorbitol are carried in the phloem by a transporter. 4. Translocation: Sugar can flow from high to low sugar concentration according to the pressure flow theory. 5. Unloading: Sugars translocated in fruit tissue are carried out from phloem tissue. 6. Membrane transport: Sugars unloaded apoplastically are taken up in cells by a transporter on the plasma membrane. 7. Metabolic conversion: Translocation sugars unloaded in fruit by the symplasmic or apoplastic pathway are converted to various substances; first, invertase and sucrose synthase metabolize sucrose, sorbitol dehydrogenase metabolizes sorbitol, and then sucrose phosphate synthase synthesizes sucrose. 8. Compartmentation: Sugars in cells are mainly compartmented in vacuoles. High accumulation of sugars in vacuoles produces a high osmotic pressure that stimulates influx of water into vacuoles, leading to a high turgor pressure that is the driving force for cellular enlargement. Physiological steps for controlling the sink strength of fruit are mainly unloading, membrane transport, metabolic conversion, and compartmentation, if sufficient photoassimilate is supplied. Among these four major steps, the role of enzymes in metabolic conversion seems to be the most important for producing the sink strength of fruit because this step correlates closely with unloading and compartmentation of sugar. Thus, in this review, I want to describe mainly "metabolic conversion".

1. Types of translocation sugars in fruit

Sucrose is well known as a translocation sugar in plants. Zimmermann and Ziegler (1975) listed the translocation sugars of various plant species. In horticultural crops, sorbitol, raffinose, stachyose,

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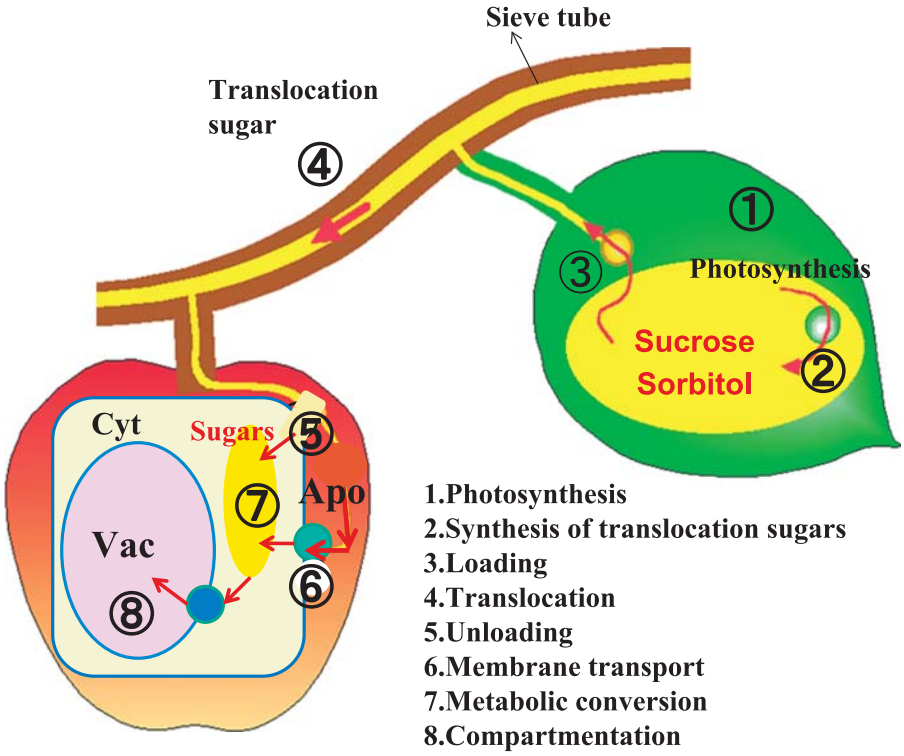

Fig. 1. Various physiological steps between photoassimilate synthesis in leaves and its accumulation in fruits for growth and development. Apo; apoplast, Cyt; cytoplasm, Vac; vacuole.

mannitol as well as sucrose are well known (Table 1). Each translocation sugar is converted to several sugars in fruit. For example, sucrose is converted to glucose and fructose by invertase or to fructose and UDPglucose by sucrose synthase. Sorbitol is converted to fructose or glucose by sorbitol dehydrogenase. Raffinose and stachyose are converted to galactose, glucose, and fructose by α-galactosidase and invertase. Mannitol is converted to fructose by mannitol dehydrogenase. I will review sucrose and sorbitol and recommend other reviews for mannitol (Loescher and Everard, 1996) and raffinose and stachyose (Keller and Pharr, 1996).

Table 1. Various translocation sugars in horticultural crops.

Horticultural crops	Translocation sugars
Citrus, Persimmon, Tomato, Grape, Strawberry, Banana, etc.	Sucrose
Apple, Peach, Pear, Cherry, Plum, Loquat, etc.	Sorbitol, Sucrose
Melon, Cucumber, Pumpkin, Squash, etc.	Raffinose, Stachyose, Sucrose
Celery, Parsley, Olive, etc.	Mannitol, Sucrose

This table was made from the result by Zimmermann and Ziegler (1975).

2. Metabolic conversion of translocation sugars in fruit

1) Sucrose-metabolizing enzymes and their regulation

(1) Loading and unloading of sucrose

Sucrose translocated in fruit is generally generated in leaves by sucrose phosphate synthase (SPS). SPS was first isolated in spinach leaves and its properties (Huber and Huber, 1996) and subsequent regulation of activity by light were clarified (Comparot et al., 2003). Sucrose synthesized in leaves is loaded into phloem by the H^+-sucrose co-transporter whose presence was predicted by Giaquinta (1977). Riesmier et al. (1994) cloned the gene of this transporter and clarified its function. Loaded sucrose is translocated to fruit flesh through the phloem and is unloaded into the parenchyma tissue. Two types of unloading in fruit are 1) the symplasmic pathway in which sucrose passes through plasmadesmata and 2) the apoplastic pathway in which sucrose is taken out in the apoplast and then transported into cells again across the plasma membrane by a transporter. In the symplasmic pathway, sucrose synthase (SuSy), cytoplasmic neutral invertase (CNIV), and vacuolar acid invertase (VAIV) mainly convert sucrose, and in the apoplastic pathway, cell wall-bound (apoplastic) acid invertase (BAIV) has an important role. In storage organs, the presence of a symplasmic pathway through the plasmadesmata was suggested, and later the presence of the apoplastic pathway in fruit flesh was detected in citrus fruit (Koch and Avigne, 1990) and apple fruit (Zhang et al., 2004). Some reports suggested both symplasmic and apoplastic pathways are important in fruit (Ruan and Patrick, 1995). For example, the symplasmic pathway functions actively in the early development stages of the tomato fruit, and

the apoplastic pathway is active in the later development stages. Conversely, sugar beet tap root uses mainly the apoplastic pathway with active BAIV in the early development stages and the symplasmic pathway with active SuSy in the late development stages (Godt and Roitsch, 2006). In walnut fruit, the seed pericarp transports sugar by the symplasmic pathway and the fleshy pericarp transports sugar by the apoplastic pathway (Wu et al., 2004). Fruit can use both pathways depending on the species, organs and the developmental stage. Recently, translocation of assimilate was easily detected in eggplant fruit by using a positron-emission tracer imaging system (Kikuchi et al., 2008). Analysis of various fruits is expected in the future.

(2) Purification and cDNA cloning of sucrose-metabolizing enzymes

i) Invertase: Invertase is divided into three types according to the cellular localization; BAIV, VAIV or soluble acid invertase (SAIV), and CNIV. Invertase has been purified and its cDNA has been cloned from many plants. Here, protein purification and full-length cDNA cloning of invertase from mainly fruits are reviewed. SAIV (Konno et al., 1993) and two kinds of acid invertase (AIV) (Nakagawa et al., 1971) from tomato fruit, two kinds of SAIV from Japanese pear fruit (Hashizume et al., 2003) and BAIV and SAIV from apple fruit (Pan et al., 2005b) were purified. Full-length cDNA of SAIV was cloned from fruits such as tomato fruit (Ohyama et al., 1998). *Lin 5*, *Lin 6*, and *Lin 7* of BAIV pallalogues in tomato (Godt and Roitsch, 1997), *GIN1* and *GIN2* of VAIV pallalogues in grape berries (Davis and Robinson, 1996), BAIV in papaya fruit (Zhou et al., 2003a) and *PsSAIV1* and *PsSAIV2* of VAIV pallalogues in Japanese pear fruit (Yamada et al., 2007).

ii) Sucrose synthase: The characteristics and function of SuSy were clarified by using mainly maize. The reaction is generally considered to incline towards sucrose degradation to supply UDPglucose. Some reports have described the properties of purified SuSy in fruit. Moriguchi and Yamaki (1988) suggested that SuSy purified from peach fruit has a high affinity to UDPglucose compared with other SuSy and has a role in sucrose synthesis. Tanase and Yamaki (2000a) purified two isozymes, SSI and SSII, from Japanese pear fruit, and indicated that the reaction of SSI inclines towards sucrose breakdown like the general type, while the reaction of SSII inclines towards sucrose synthesis like SuSy of peach fruit. Further, Tanase et al. (2002) found that SSI is phosphorylated, but SSII is unphosphorylated, which is consistent with the previous report by Huber et al. (1996) that phosphorylation of SuSy has a high affinity for UDP and stimulates sucrose breakdown. SuSy was also purified from tomato fruit (Islam, 2001) and banana fruit (do Nascimento et al., 2000), and the reaction inclined towards sucrose breakdown (supply of UDPglucose) by using kinetic analysis. Full-length cDNA was cloned from fruits, such as *SuSy3* and *SuSy4* from tomato fruit (Chengappa et al., 1998; Wang et al., 1993), *CaSUS1* and *CaSUS2* from coffee fruit (Geromel et al., 2006), and *CitSUS1*, *CitSUSA*, and *CitSUS2* from citrus fruit (Komatsu et al., 2002).

iii) Sucrose phosphate synthase: SPS was purified from spinach leaves. It has a role in switching between sink and source function of leaves by its transcriptional regulation (Chavez-Barcenas et al., 2000). SPS was also regulated by post-translational modification by phosphorylation and by a negative or positive effector like glucose-6-P or Pi (Huber and Huber, 1996). However, the properties of SPS in fruit are not clear because of no report of SPS purified from fruit. SPS activity in apple leaves was inhibited by sorbitol-6-P and the inhibition has a role in switching between the synthesis of sucrose and the synthesis of sorbitol as a translocation sugar (Zhou et al., 2002). Full-length cDNA was cloned from citrus fruit, including *CitSPS1*, *CitSPS2*, and *CitSPS3* (Komatsu et al., 1996, 1999), *CmSPS1* from melon (Yu et al., 2007), from kiwifruit (Langenkämper et al., 2002), from Asian pear (Itai and Tanahashi, 2008) and from coffee (Privat et al., 2008).

(3) Changes in expression of sucrose-metabolizing enzymes with fruit development

Sucrose translocated in fruit is generally broken down to glucose, fructose, or UDPglucose by SuSy or invertase, then SPS functions actively to resynthesize sucrose in the fruit. In citrus fruit, VAIV, BAIV, SuSy, and SPS activities were assayed from each tissue and at each stage of development (Lowell et al., 1989). SuSy activity was highest among the four enzyme activities in the vascular bundle system, and SPS activity was highest in juice sac tissue. Both activities of VAIV and BAIV of the four enzymes were highest in the young stage of fruit development, but decreased rapidly with the formation of juice sacs. An increase in SuSy activity in the enlargement stage in which the fruit has just started to enlarge was shown in fruit of *Citrus unshiu* (Kubo et al., 2001). Further, *CitSUS1* and *CitSUSA* of *Citrus unshiu* were detected in edible tissue of the fruit, and *CitSUS1* expression decreased with fruit development, but *CitSUSA* expression increased (Komatsu et al., 2002). The expression of *CitSPS1* and *CitSPS2* in *Citrus unhsiu* fruit corresponded to sucrose accumulation, but the expression of *CitSPS3* was not detected in edible tissues (Komatsu et al., 1999).

Tomato fruit that accumulates hexose increased AIV activity with fruit maturation, but fruit accumulating sucrose decreased AIV activity markedly (Stommel, 1992). However, definite increases in SuSy and SPS activities did not occur with fruit maturation. Comparing tomato fruit cultured in the cool season with tomato fruit cultured in the warm season, activities of AIV and SuSy were higher in the cool season when more sugars accumulate, but SPS activity did not change (Islam and Khan, 2001). Therefore, SuSy and AIV are considered

to have a role in sugar accumulation. BAIV pallalogues, *Lin5*, *Lin6*, and *Lin7* are present in tomato fruit, and *Lin6* was expressed specifically in sink organs and its expression greatly increased with increases in sink activity (Godt and Roitsch, 1997). Four AIV pallalogues are present in the tomato; the expressions of two of these pallalogues were correlated closely with sugar accumulation in fruit (Husain et al., 2003).

In pear fruit, changes in activities and mRNA levels of sucrose-metabolizing enzymes SuSy, SPS, VAIV, and BAIV were investigated in relation to fruit development (Yamada et al., 2006). The fluctuation pattern of SuSy activity with NAD-dependent sorbitol dehydrogenase (NAD-SDH) activity paralleled to that of the relative growth rate (RGR); therefore, SuSy may be important for sugar uptake. AIV activity is higher in young fruit, but is low in mature fruit, and the mRNA of one isoform is expressed in the mature stage. SuSy isozymes SSI and SSII are present in Japanese pear fruit; SSI activity was high in young fruit but decreased with fruit development, and SSII activity which was not detected in young fruit, increased rapidly with sugar accumulation in the mature stage (Tanase and Yamaki, 2000). Moriguchi et al. (1992) examined seasonal changes in SuSy, SPS, and AIV activities by comparing Asian pear fruits that accumulate mainly sucrose with Asian pear fruits that accumulate mainly hexose. In pear fruit that accumulated mainly sucrose, SuSy and SPS activities increased with fruit maturation, but in pear fruit that accumulated less sucrose these activities did not increase. AIV activity decreased rapidly with fruit maturation in the fruit of both sugar accumulation types. Ito et al. (2002) examined changes in SuSy and AIV activities related to NAD-SDH and NADP-dependent sorbitol dehydrogenase (NADP-SDH) activities in the buds of Japanese pear tree and indicated that AIV activity with NAD-SDH activity increased markedly with bud growth. SuSy and AIV activities with NAD-SDH activity were also investigated in endocarps, mesocarps, and the seeds of peach fruit during fruit development (Lo Bianco and Rieger, 2002). SuSy and AIV activities are high in young fruit, and the activities are especially high in seeds in the hardening stage. Moriguchi et al. (1991) reported the role of SuSy and other related enzymes in sucrose accumulation in peach fruit.

In grape berries, VAIV participates actively in hexose accumulation (Davis and Robinson, 1996). The expressions of the VAIV genes *GIN1* and *GIN2* were high in young fruit, then decreased but remained constant in the mature stage. The expression of the genes and enzyme synthesis occurred several weeks before the onset of hexose accumulation. AIV in papaya fruit is more important than SuSy or SPS for fruit maturation because the mRNA of AIV increases with fruit maturation (Zhou and Quebedeaux, 2003; Zhou et al., 2000). AIV in strawberry fruit participates actively in hexose accumulation. VAIV activity increased together with hexose accumulation during maturation, and BAIV activity was high in young fruit but decreased with fruit maturation (Ranwala et al., 1992). The increase in SuSy activity in coffee fruit participates actively in sucrose accumulation during fruit maturation (Geromel et al., 2006).

In melon fruit, the relationship between activities of SPS, SuSy, and AIV and sucrose accumulation during fruit development were examined using various musk melon fruits (Hubbard et al., 1989). SPS activity increased markedly with fruit maturation and corresponded to the sucrose accumulation. Thus, the strength of SPS activity determines the amounts of accumulated sucrose. When the activities of SuSy and SPS and sucrose amounts were compared between seeded (accumulating a lot of sucrose) and seedless melon fruit (accumulating a little sucrose), SuSy activity in seeded fruit increased in the mature stage, but did not in seedless fruit, while SPS activity in seeded fruit increased before sucrose accumulation, but decreased gradually in seedless fruit (Hayata et al., 2001a). This suggests that both enzymes are important for sucrose accumulation in melon fruit. In banana fruit, SuSy activity and expression of SuSy mRNA were high to accumulate starch in immature fruit, and SPS activity increased markedly to convert starch to sucrose during fruit maturation (do Nascimento et al., 2000; Hubbard et al., 1990). In pineapple fruit, SuSy and AIV activities were high in young fruit but decreased with fruit development, while SPS activity was not as high as their activities in young fruit, but remained constant during fruit maturation to accumulate a lot of sucrose (Chen and Paull, 2000). SPS activity increased to accumulate sucrose during fruit maturation of pumpkin fruit (Tateishi et al., 2004) and kiwifruit (Langenkämper et al., 1998).

(4) Regulation of sucrose-metabolizing enzymes by phytohormones and stresses

Table 2 shows the regulation of sugar-metabolizing enzymes by phytohormones and stresses, except for sugar in fruit or other sink organs. Regarding phytohormonal effects, AIV, SuSy, and SPS activities were increased by GA_3, ABA, cytokinine, and brassinolide, by GA_3, ABA, IAA, and brassinolide and by GA_3, cytokinine, and brassinolide, respectively. Conversly, AIV activity was decreased by IAA. SuSy mRNA shows different expression in an anoxia condition, that is, one isoform increases the expression and another isoform decreases the expression. In a hypoxia condition the expression of SuSy mRNA increased. AIV mRNA shows different expression in its gene family in a hypoxia condition like SuSy mRNA. SPS mRNA expression increased with a low temperature treatment. NaCl stress increased SPS and SuSy activities, while osmotic stress increased AIV, SuSy, and SPS activities. There are many reports about hormonal and stress effects of AIV, SuSy, and SPS activities, but the reverse effect among species is rare. Thus, it may be

Table 2. Regulation of sucrose-metabolizing enzymes by phytohormones and stress.

Phytohormone	Materials	Enzyme activity	(↑/↓)[z]	mRNA level (↑/↓)	Reference	Comment
GA_3	Pea pod	AIV	↑	—	Estruch and Beltran, 1991	
	Pea	BAIV	↑	↑	Wu et al., 1993	
	Pea subhook	AIV	↑	↑	Miyamoto et al., 2000	
	Watermelon seed	SuSy	—	*Cuss1* ↑	Kim et al., 2002	
	Banana fruit	SPS	↑	—	Miranda et al., 2003	
	Chickpea seedling	SuSy	↑	—	Kaur et al., 2000	with cytokinin
		SPS	↑	—	Kaur et al., 2000	
ABA	Sorghum grain	SuSy	↑	—	Bhatia and Singh, 2002	
	Grape berry	VAIV, BAIV	↑	—	Pan et al., 2005a	
	Apple fruit	BAIV	↑	—	Pan et al., 2006	
	Strawberry	SuSy	↑		Saito et al., 2009	
IAA	Chickpea seedling	SuSy, SPS	↑	—	Kaur et al., 2003	
	Sorghum grain	SPS, AIV	↑	—	Bhatia and Singh, 2002	
	Melon fruit	SuSy, SPS	↑	—	Li et al., 2002	by 4-CPA
Cytokinin	Melon fruit	AIV	↑	—	Hayata et al., 2001b	by CPPU
		SPS	↑	—	Li et al., 2002 by CPPU	
	Tomato fruit	AIV	—	*Lin6* ↑	Godt and Roitsch, 1997	by zeatin
Brassinolide	Cucumber leaf	SuSy, SPS, AIV	↑	—	Yu et al., 2004	by epibrassinolide
Stress						
Anoxia	Maize carnel	SuSy	—	*Sh1* ↑	Zeng et al., 1998	
	Pond weed	SuSy	—	*PdSS1* ↑, *PdSS2* ↓	Harada et al., 2005	
Hypoxia	Maize carnel	AIV	—	*Inv1* ↑, *Inv2* ↓	Zeng et al., 1999	
		SuSy		*Sh1* ↑ *SUS1* ↑	Zeng et al., 1998	
	Tomato root	SuSy	↑	—	Germain et al., 1997	
Low temperature	Kiwifruit	SPS	—	↑	Langenkämper et al., 1998	
NaCl	Tomato	SPS	↑	—	Carvajal et al., 2000	
	Chickpea seedling	SuSy, SPS	↑	—	Kaur et al., 2003	
Drought	Peach leaf	SuSy	↓	—	Lo Bianco et al., 2000	
	Maize	AIV	—	*Inv2* ↑	Kim et al., 2000b	
Osmotic	Sweet potato	SuSy, AIV	↑	—	Wang et al., 2000	
		SPS	—	↑	Wang et al., 2000	

[z] Increase, ↑; decrease, ↓

effective to use them to control plant growth and development.

Regarding sugar regulation (sugar sensing) in maize, AIV generates hexose-based sugar sensing by supplying hexose, which induces cell division and induces related gene expressions, but SuSy minimizes hexose-based sugar sensing and induces gene expression for storage and maturation. Both AIV and SuSy form isozymes and were regulated reciprocally, that is, one isozyme was induced by a high sugar concentration, but the other enzyme was repressed by a high sugar concentration and induced by starvation (Koch, 1996, 2004). Although there are some reports of sugar sensing in fruits (Kanayama et al., 1998; Schaffer and Petreikov, 1997), reports of sugar sensing in fruit that accumulate a lot of sugar are rare.

2) Sorbitol-metabolizing enzymes and their regulation

Kanayama (2009) reviewed the cloning, properties and functions of sorbitol enzymes. Therefore, the present review targets mainly the role of sorbitol enzymes in fruit development, and describes briefly the cloning and properties of sorbitol enzymes.

(1) Loading and unloading of sorbitol

Sorbitol occurs in leaves from dephosphorylation of sorbitol-6-P that is synthesized from glucose-6-P by sorbitol-6-P dehydrogenase (S6PDH). Then, sorbitol is loaded into phloem by the H^+/sorbitol co-transporter and is translocated to the fruit. Sorbitol unloaded in the fruit is converted to fructose or glucose by NAD-SDH or NADP-SDH. S6PDH was purified from loquat fruit (Hirai, 1979) and the gene was cloned from apple leaves (Kanayama et al., 1992). Sorbitol-6-P-specific phosphatase was purified from apple leaves (Zhou et al., 2003b). The sorbitol transporter which was not related directly to phloem loading of sorbitol, was first cloned from the cherry (Gao et al., 2003). The H^+/sorbitol co-transporter localized in phloem tissues for sorbitol loading was cloned from apple leaves (Watari et al., 2004).

(2) Purification and cloning of sorbitol dehydrogenase

NAD-SDH was found in apple fruit (Negm and Loescher, 1979). It was purified completely from

Japanese pear fruit and the detailed properties were clarified (Oura et al., 2000). Yamada et al. (1998) cloned full-length cDNA from apple fruit. Thereafter, it was cloned in some other fruits (Ito et al., 2005; Kim et al., 2007; Nosarszewski et al., 2004; Ohta et al., 2005; Park et al., 2002; Yamada et al., 2001). NAD-SDH forms a gene family (Ito et al., 2005; Nosarszewski et al., 2004; Park et al., 2002). In the strawberry, which belongs to the Rosaceae family and in which the fruit does not use sorbitol as a translocation sugar, NAD-SDH activity was detected and its cDNA was cloned (Duangsrisai et al., 2007). NAD-SDH present in Rosaceae, such as apples, pears, peaches, has a lower Km value (about 100 mM) for sorbitol degradation than the Km value (several M) for sorbitol synthesis, and so easily converts translocated sorbitol to fructose. However, NAD-SDH from maize kernel has a Km value of 100 mM for sorbitol synthesis and can synthesize sorbitol from fructose without S6PDH in kernel tissue (Doehlert, 1987). NAD-SDH in strawberry fruit is not the type of NAD-SDH found in Rosaceae, but is the type found in maize, and so it can produce sorbitol from fructose (Duangsrisai et al., 2008).

(3) Changes in the expression of sorbitol-metabolizing enzymes with fruit development

Translocated sorbitol is converted to fructose by NAD-SDH or to glucose by NADP-SDH. NAD-SDH activity in apple fruit was high in young fruit, decreased with fruit enlargement and increased again with fruit maturation (Yamaki and Ishikawa, 1986). Because the fluctuation pattern of NAD-SDH activity during apple fruit development corresponded to that of mRNA expression, NAD-SDH activity is controlled by transcriptional regulation (Yamada et al., 1999). Nosarzewski et al. (2004) found high activity and high amount of NAD-SDH protein during fruit setting and in the early development stages by ELISA. *SDH1* and *SDH3* localized in flesh and seeds, *SDH2* in flesh and *SDH6* and *SDH9* only in seeds, and NAD-SDH activity in seeds was clearly higher than in flesh for 2 to 5 weeks after fruit setting in apple (Nosarszewski and Archbold, 2007). Suzuki et al. (2001) suggested that accumulation of fructose in apples fruit is not produced only by an increase in NAD-SDH activity, but also by a decrease in fructokinase activity.

In Japanese pear fruit, the fluctuation pattern of NAD-SDH activity with fruit development was similar to that in apple fruit (Yamaki and Moriguchi, 1989). The fluctuation pattern of pear fruit corresponded to that of the mRNA level, that is, it was controlled by transcriptional regulation (Yamada et al., 2006). Ito et al. (2002) examined activities of NAD-SDH and NADP-SDH on bud growth; NAD-SDH activity clearly increased, but NADP-SDH activity did not change. Both activities of NAD-SDH and NADP-SDH increased slightly to stimulate sorbitol metabolism by shading buds (Ito et al., 2003). Both activities in lateral buds increased with horizontal stem growth, but NAD-SDH activity in shoot internodes of horizontal stems decreased with growth. Consequently, this change may increase the sink capacity of buds relative to shoot tissue, and so stimulate bud growth (Ito et al., 2004).

In the peach, high NAD-SDH activity was detected in the shoot tip (Lo Bianco and Rieger, 1999). Changes in NAD-SDH activity with peach fruit development were more or less similar to that in apple fruit and Japanese pear fruit, but the level of mRNA did not correspond partially with fluctuations in the activity, suggesting post-transcriptional modification (Yamada et al., 2006). NAD-SDH activity was high in flesh tissue before the stone hardening stage of peach fruit and was high in seeds during the stone hardening stage (Lo Bianco and Rieger, 2002). In loquat fruit, the activity and mRNA amount of NAD-SDH increased simultaneously with fruit maturation. S6PDH activity in fruit clearly increased, as well as NAD-SDH activity (Bantog et al., 2000). However, generation of sorbitol from hexose in plum fruit itself was not found by tracer experiments using ^{14}C-hexose (Hansen and Ryugo, 1979). If this S6PDH functions in the catabolism of sorbitol that accumulates in loquat fruit, sorbitol kinase will be required to produce sorbitol-6-P from sorbitol as a substrate. However, sorbitol kinase activity has not been detected. The role of S6PDH in fruit remains unclear.

(4) Regulation of sorbitol-metabolizing enzymes by phytohormones and other factors

There are few reports on activation and regulation by phytohormones and other factors. The activity and mRNA level of S6PDH increased in sliced tissue of apple fruit treated by abscisic acid (Kanayama et al., 2006). The activity and mRNA level of NAD-SDH also increased in sliced tissue of strawberry fruit treated with IAA (Duangsrisai et al., 2008). Regulation of NAD-SDH by sugar was examined. Decreasing the sugar concentration in apple fruit by girdling the stem reduced NAD-SDH activity (Berüter and Feusi, 1997). NAD-SDH activity clearly increased by supplying sorbitol or glucose to discs of fruit on girdled stems (Archbold, 1999). Discs of apple fruit incubated with sorbitol, glucose, or sucrose enhanced both the activity and mRNA level of NAD-SDH (Iida et al., 2004). The activity and mRNA level of S6PDH in apple leaves increased with a low temperature or salt stress (Kanayama et al., 2006). The activity of NAD-SDH and S6PDH in peach leaves decreased during drought (Lo Bianco et al., 2000).

3) Other important enzymes for sugar metabolism and accumulation

Other glycolytic enzymes are also important to accumulate sugar. Especially, fructokinase is important to regulate the level of fructose that is the primary product of sucrose and sorbitol. Kanayama et al. (1997, 1998) purified isozymes FrK1 and FrK2, isolated the cDNA of each isozyme, and showed the expression of FrK1 in

young fruit and FrK2 in mature fruit. Fructokinase was regulated by sugar (Kanayama et al., 1997), and controlled the fructose level in apples and Japanese pear fruit (Suzuki et al., 2001) and peach fruit (Kanayama et al., 2005). ADPglucose pyrophosphorylase and starch synthase for starch generation, and phosphorylase and amylase for starch breakdown are also important for sugar accumulation and sink activity in fruits accumulating a lot of starch. However, this review does not touch starch metabolism.

3. Sucrose-accumulating type and hexose-accumulating type of fruit

1) Relation between fruit growth and sweetness

Sweetness differs depending on the variety of sugar accumulating in fruit. Generally, the sweetness levels of fructose, glucose and sorbitol are roughly 130, 70, and 60, respectively, if the sweetness of sucrose is 100. Therefore, which sugar accumulates is important for sweetness as a quality. If fruit accumulates the same amounts of sugar, accumulation of fructose is good for sweetness. However, transgenic tomatoes introduced VAIV antisense gene in hexose-accumulating tomato accumulated a lot of sucrose and were sweeter (Klann et al., 1996; Ohyama et al., 1995). This can be explained by transgenic tomatoes containing the AIV antisense gene accumulating more sugar by suppressing the increase in osmotic pressure by reducing the breakdown of sucrose to hexose. Fruits accumulated mainly hexose during the early stages inducing active cell division and during the middle stages showing the most RGR by active cell enlargement, but accumulated mainly sucrose during the mature stages showing weak cell enlargement (Lo Bianco and Rieger, 2002; Yamada et al., 2006). In development stages that have active enlargement, producing great turgor pressure by high osmotic pressure made by converting sucrose to hexose is good for fruit growth. However, suppressing the increase in turgor pressure by converting hexose to sucrose is good for protecting cellular degradation in the mature stages, resulting in the accumulation of a lot of sugar in mature fruit. For example, transgenic tomato fruit containing the introduced AIV antisense gene accumulated more sucrose, but the fruit size was smaller (Klann et al., 1996). The specific expression of the isoform of AIV genes in the middle stages of development that show active enlargement may supply a lot of hexose for cell enlargement (Yamada et al., 2007). Fruit seems able to change dramatically the metabolic pathway to control the ratio of sucrose to hexose during fruit growth.

2) Sucrose-metabolizing enzyme activity in sucrose- and hexose-accumulating types of fruit

Some mature fruit accumulates mainly sucrose (sucrose-accumulating type) and other mature fruit accumulates mainly hexose (hexose-accumulting type). For example, many commercial varieties of tomato fruit belong to the hexose-accumulating type, but some wild varieties belong to the sucrose accumulating type. In mature fruits of melon, persimmon, strawberry, Asian pear, and apple, both types and their intermediates are present. In Rosaceae fruit, the sugar composition is complicated because sorbitol is present in addition to both types.

The VAIV activity of the hexose-accumulating type of tomato was high in mature fruit and broke down actively translocated sucrose to hexose, resulting in the tomato accumulating a lot of hexose. However, the VAIV activity of the sucrose-accumulating type of tomato was low in mature fruit and did not break down translocated sucrose, resulting in the tomato accumulating a lot of sucrose despite low SPS activity (Klann et al., 1993; Miron and Schaffer, 1991; Ohyama et al., 1995; Stommel, 1992). Thus, the ratio of sucrose to hexose in tomato fruit is controlled by the strength of VAIV activity in mature fruit.

Sucrose accumulation in melon fruit differs from that in tomato fruit. By comparing sucrose contents with the capacity for sucrose synthesis, that is, the value that SPS activity was subtracted by SuSy activity and invertase activity, among many varieties of mature and immature fruits, the sucrose content was parallel to the capacity for sucrose synthesis (Hubbard et al., 1989; Lester et al., 2001). In a comparison of sugar metabolism between seeded and seedless fruits, the sucrose content in seeded fruit was higher than in seedless fruit due to the higher activity of SPS in seeded fruit (Hayata et al., 2001a). Thus, sucrose accumulation in melon fruit depends on the strength of SPS activity. Other fruits accumulating sucrose by mainly SPS activity are kiwifruit (Hubbard et al., 1991), banana (Hubbard et al., 1990), and citrus fruits (Lowell et al., 1989).

Many mature peach fruits accumulate more than 80% of sugars as sucrose. This accumulation of sucrose was produced by an increase in SuSy activity with fruit maturation, but SPS activity did not increase (Moriguchi et al., 1990). In the sucrose-accumulating type of Asian pear fruit, sucrose accumulated because of an increase in both SuSy and SPS activities, especially SuSy, with fruit maturation (Moriguchi et al., 1992). Generally, SuSy catalyzes the reaction of sucrose degradation, rather than of sucrose generation, to have ADPglucose as a precursor of starch synthesis. However, the SuSy isozyme, whose reaction inclines toward sucrose synthesis, was present in pear fruit (Tanase and Yamaki, 2000b). Thus, sucrose accumulates in fruit by using various sucrose-metabolic pathways.

4. Sugar accumulation in vacuoles

1) Sugar concentration in vacuoles and turgor pressure production

It is well known that vacuoles sequester or accumulate various substances related to fruit quality, such as sugars,

organic acids, phenolic compounds, anthocyanine pigments, alkaloids, and minerals. Especially, a lot of sugar accumulates in vacuoles and can produce large turgor pressure. However, there are only a few reports about sugar concentration in large storage cells, such as fruit flesh, due to the difficulty of isolating intact vacuoles. Vacuoles isolated intact from immature apple fruit flesh contained 706 mM of total sugars composed of 396, 295, 1, and 14 mM of fructose, glucose, sucrose, and sorbitol, respectively. This sugar accumulation in vacuoles formed about 14 atm of turgor pressure because 67 mM of total sugar accumulated in the apoplast (Yamaki, 1984). According to compartment analysis, vacuoles and extra-cellular free space in sugar beet root contained 514 and 63 mM sucrose, respectively, and produced 10 atm of turgor pressure (Saftner et al., 1983). Mature apple fruit contained 937 mM total sugars composed of 122, 149, and 613 mM of sucrose, fructose, and glucose, respectively, and produced about 11 atm of turgor pressure as total sugar of 440 mM in apoplasts (Yamaki and Ino, 1992). Sugar concentrations in vacuoles and apoplasts were assayed and the turgor pressure was estimated by using compartment analysis in strawberry fruit (Ofosu-Anim and Yamaki, 1994a) and melon fruit (Ofosu-Anim and Yamaki, 1994b).

2) Transporter and endocytosis of sugars

A lot of sugar accumulates in vacuoles and it produces high osmotic pressure. Therefore, a sugar transporter coupled with energy on the tonoplast, i.e., an H^+/sugar antiporter, is needed. The presence of this transporter was found by using isolated vacuoles (Yamaki, 1987) or tonoplast vesicles (Getz, 1991; Getz and Klein, 1995; Greuter and Keller, 1993). Recently, the gene of this sugar transporter localized on the tonoplast has been cloned (Endler et al., 2006; Wormit et al., 2006), although many genes of H^+/sugar co-transporters localized on the plasma membrane have already been cloned from various plants, and their properties and functions have been clarified.

Etxeberria et al. (2005a) and Baroja-Fernandez et al. (2006) reported the uptake system of sucrose, in which vesicles containing sucrose are generated in apoplasts and are taken up into the cytoplasm by endocytosis. Vesicles containing sucrose were carried directly into vacuoles by endocytosis in sycamore suspension cells, and the endocytosis of sucrose was induced by sucrose. In citrus fruit, sucrose in apoplasts is transported directly into vacuoles by the same endocytosis as for sycamore cells (Etxeberria et al., 2005b). Although this kind of sugar uptake system is very efficient for accumulation of a lot of sugar in vacuoles, further study is needed.

5. Sugar-metabolizing enzymes and sink activity

The attractive force that takes up assimilate into sink organs is called the sink capacity. The sink capacity is expressed as the degree of sink strength composed of sink size and sink activity.

$$\text{Sink strength} = \text{Sink size} \times \text{Sink activity}$$

Sink activity is the rate of uptake of assimilates per unit weight of sink tissue. Sink size is the total weight of the sink tissue. When the sink tissue size is unchangeable, the rising of sink activity is indispensable for an increase in sink strength. If translocation sugar is sufficient in leaves, sink activity will occur mainly by four elements of unloading, membrane transport, metabolic conversion and compartmentation. Especially, the role of AIV, SuSy, and NAD-SDH related unloading and metabolic conversion is important. RGR which was reviewed by Opara (2000) is influenced by the degree of sink strength. Sink strength can regulate photosynthesis. That is, if sink strength was reduced in the sink-limited condition of apple trees by girdling, Rubisco and photosynthesis activities in leaves decreased (Chen et al., 2008). Thus, comparing sink activity with RGR is important.

In peach fruit, changes in NAD-SDH and AIV activities correlated positively with changes in RGR by increasing the sink activity, and in the stone hardening stage SuSy contributed to sink activity (Lo Bianco and Rieger, 2002). The activities of NAD-SDH, SuSy and VAIV in pear fruit participated actively in accumulating sugar, that is, changes in NAD-SDH and SuSy activities correlated to that of RGR. Both NAD-SDH and SuSy contributed a lot to sink activity, as well as to peach fruit (Yamada et al., 2006). The fruit size of apples with a high crop load depended mainly on sink activity, and that of apples with a low crop load depended mainly on partitioning from leaves rather than sink activity (Klages et al., 2001). Transgenic apples introduced antisense S6PDH gene produced a lot of accumulation of sucrose and less accumulation of sorbitol in leaves, with a decrease in NAD-SDH activity and an increase in SuSy activity in shoots (Zhou et al., 2006). Thus, SuSy and NAD-SDH contribute mainly to sink activity.

In grape berries, unloading is mainly by the symplasmic system until the veraison stage and changes to mainly the apoplastic system with maturation. Simultaneously with maturation, BAIV activity increased and strengthened the sink activity (Zhang et al., 2006). Conversely, sugar beet root used the apoplastic unloading system during the early growth stages and retained higher BAIV activity, but it used the symplasmic unloading system and retained higher SuSy activity during the late stages of growth (Godt and Roitsch, 2006). Changes in SuSy, SPS, and AIV activities were also investigated in relation to defoliation and thinning of papaya (Zhou and Paull, 2001; Zhou et al., 2000). SuSy and AIV activities influenced sink activity in the early growth stages, and AIV activities influenced a lot of sink activity in the later growth stages, but SPS activity did not influence sink activity throughout the whole season.

In transgenic tomato fruit introduced antisense BAIV

gene, BAIV activity was effective for the sink activity of fruit (Ohyama et al., 1995, 1998). SPS also influenced sink activity because over-expression of the SPS gene increased partitioning and unloading of photoassimilates into fruit (Laporte et al., 2001; Nguyen-Quoc et al., 1999). This contradicts the view that sink activity is produced mainly by unloading, membrane transport, metabolic conversion and compartmentation. However, SPS may indirectly influence sink activity when generation of translocation sugar is insufficient to partition it in fruit and it becomes a limiting factor of partitioning.

6. Phenotypic expression by regulating expression of sucrose- and sorbitol-metabolizing enzymes

Clarifying how the expression control of sucrose-and sorbitol-metabolizing enzymes influences the function of various organs and other metabolic systems is very important.

i) Invertase: Suppression of AIV activity by the antisense gene of BAIV in tomatoes led to an increase in sucrose and a decrease in hexose (Ohyama, 1995). Over-expression of the yeast invertase gene in each compartment of apoplasts, vacuoles or cytosol of potato leaves decreased assimilation of CO_2, retarded growth and they accumulated more hexose and proline (Bussis et al., 1997). Carrots had thick leaves when VAIV or BAIV activity was suppressed by introducing their antisense gene. However, roots became small with VAIV suppression, and root growth was suppressed and carbohydrate content was reduced by BAIV suppression (Tang et al., 1999). Introducing the AIV antisense gene into broccoli suppressed the expression of *BoINV2*, but not *BoIVN1*, and simultaneously suppressed the expression of cystein protease (*BoCP5*) that led to a delay of floret senescence (Eason et al., 2007).

ii) SuSy: Transgenic tomatoes containing an introduced SuSy antisense gene linked to a fruit-specific promoter inhibited 99% of SuSy activity, but did not change the accumulation of starch and sugars (Chengappa et al., 1999). Therefore, SuSy seems not to participate in the main sink activity. A similar result was shown by another transgenic tomato containing an introduced SuSy antisense gene (D'Aoust et al., 1999). Suppression of SuSy activity by introducing the SuSy antisense gene into potatoes did not much influence the production of the glycolysis intermediate (Biemelt et al., 1999). In transgenic potato plants with over-expression due to the SuSy gene or with suppressed expression due to the antisense gene, transgenic potatoes did not influence the expression of ADPglucose pyrophosphorylase, which is a key enzyme to accumulate photosynthate for starch synthesis (Kim et al., 2000a). Thus, SuSy does not seem to be a key enzyme needed to accumulate photosynthate.

iii) SPS: Transgenic tomatoes containing the over-expressed maize SPS gene increased SPS activity in leaves 6-fold, increased sucrose content, but decreased starch content (Worrell et al., 1991). SPS in leaves regulates partitioning of assimilate. In transgenic tomatoes containing the over-expressed SPS gene, SPS activity increased 3-fold in leaves, but not in fruit, while the sugar contents in fruit increased (Laporte et al., 1997). In other transgenic tomatoes containing the over-expressed SPS gene, the SPS activity increased 2.4-fold, but other enzyme activities did not change, and unloading of sucrose was more active (Nguyen-Quoc et al., 1999). That is, sink activity in fruit is higher. Transgenic tomatoes containing the over-expressed SPS gene linked to a cauliflower mosaic virus 35S promoter increased yield by more than 80% because more assimilate is partitioned in fruit, but photosynthesis did not change (Laporte et al., 2001). SPS seems to have a major role in partitioning assimilate in leaves for transport to fruit.

iv) S6PDH and NAD-SDH: Transgenic tobacco introduced S6PDH gene produced a little sorbitol and did not accumulate much sorbiotol (Tao et al., 1995). This transgenic tobacco was resistant to boron deficiency by stimulating the uptake and transport of boron (Bellaloui et al., 1999; Brown et al., 1999). Rice introduced S6PDH gene was resistant to boron deficiency similar to transgenic tobacco (Bellaloui et al., 2003). Transgenic persimmons introduced S6PDH gene accumulated a lot of sorbitol and was resistant to salt stress (Gao et al., 2001), but became dwarves with no change in IAA or GA contents (Deguchi et al., 2004). Transgenic apples introduced S6PDH gene had increased S6PDH activity that increased the total amount of sorbitol and sucrose to 1.5-fold, and reduced S6PDH activity by co-suppression that increased sucrose in place of a prominent decrease in sorbitol (Kanamaru et al., 2004). Transgenic apples containing the introduced antisense S6PDH gene decreased S6PDH activity, but did not influence SPS activity, but increased fructose 1,6-bisphosphatase activity (Cheng et al., 2005). Similar transgenic apples reduced SDH activity in fruit, but did not influence the activities of AIV, fructokinase or hexokinase (Teo et al., 2006). Such transgenic apples reduced sorbitol synthesis and SDH activity in the shoot tip, but did not influence AIV activity, and increased SuSy activity (Zhou et al., 2006). Recently Deguchi et al. (2006) produced transgenic tobacco introduced genes of S6PDH and NAD-SDH together, and showed that this transgenic tobacco had a higher survival rate than the transgenic tobacco introduced only S6PDH gene and accumulated a lot of sorbitol.

7. Conclusion

Enzymes and transporters relating to synthesis, loading, unloading, membrane transport, metabolic conversion and compartmentation of translocation sugars, sucrose and sorbitol, discussed above are summed up in Fig. 2.

Fruit can enlarge by producing turgor pressure by

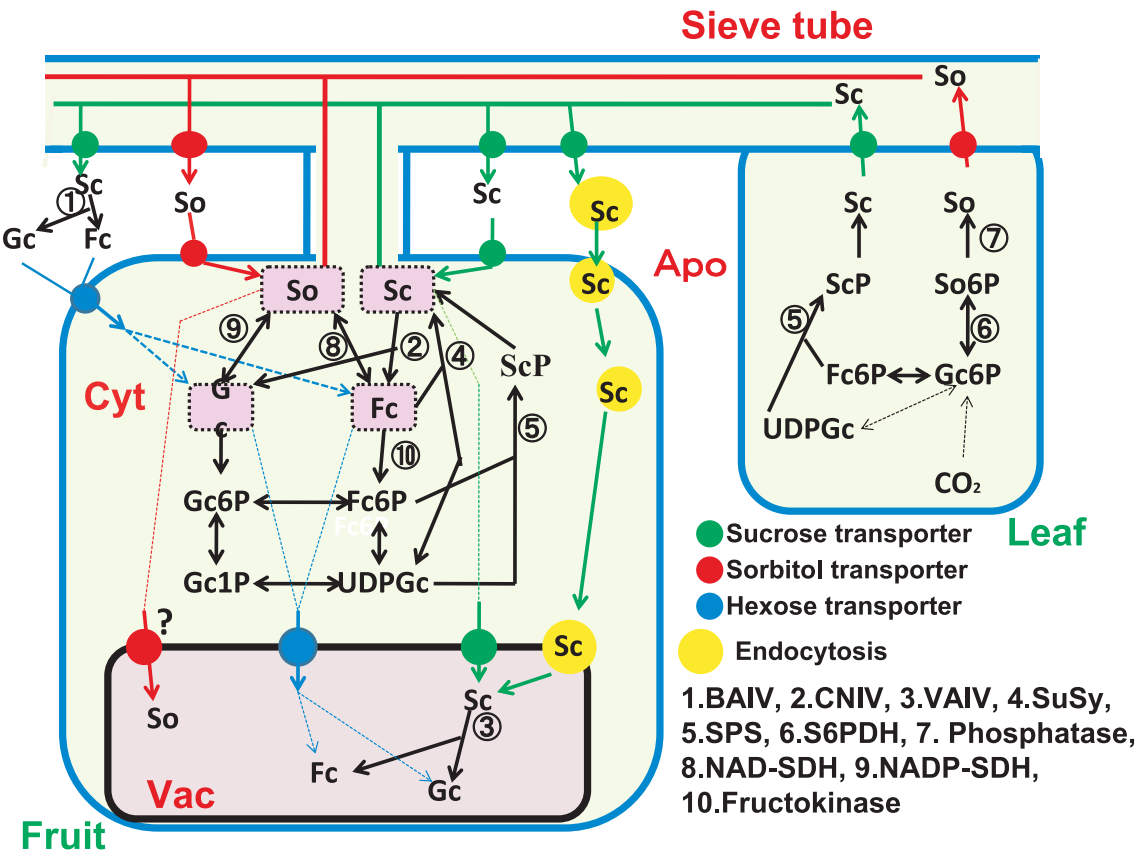

Fig. 2. Enzymes and transporters relating to synthesis, loading, unloading, membrane transport, metabolic conversion, and compartmentation of translocation sugars, sucrose and sorbitol. BAIV; cell wall-bound acid invertase, CNIV; cytoplasmic neutral invertase; Fc; fructose, Gc; glucose, NAD-SDH; NAD-dependent sorbitol dehydrogenase, NADP-SDH; NADP-dependent sorbitol dehydrogenase, Sc; sucrose, So; sorbitol, S6PDH; sorbitol-6-P dehydrogenase, SPS; sucrose phosphate synthase, SuSy: sucrose synthase, VAIV; vacuolar acid invertase.

accumulating sugars and by changing the ratio of hexose to sucrose. The force accumulating sugar is mainly formed by sink activity. The expression of SuSy, invertase and NAD-SDH regulated by various conditions are very active to produce sink activity. Therefore, expression of the genes of these enzymes and the activities in various fruits should be clarified and the different regulations by various conditions should be investigated. These enzymes do not always contribute to sink activity as shown in studies using transgenic fruit containing these genes. The use of promoters that are expressed specifically in fruit is useful to understand their influence on sink activity. The other important thing in the production of sink activity is that almost all sugars in fruit accumulate in vacuoles. However, how the sugars accumulate in vacuoles is not so clear, although recently genes of the sugar transporter localized on the tonoplast were cloned. It might predict other systems of sugar accumulation such as direct uptake of sugars into vacuoles from apoplasts by endocytosis. To freely convert translocation sugars quantitatively or qualitatively to other sugars will have a great impact on stimulating fruit development and improving fruit quality because sugars are the primary substrate of various fruit components.

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Literature Cited

Archbold, D. D. 1999. Carbohydrate availability modifies sorbitol dehydrogenase activity of apple fruit. Physiol. Plant. 105: 391–395.

Bantog, N. A., K. Yamada, N. Niwa, K. Shiratake and S. Yamaki. 2000. Gene expression of NAD^+-dependent sorbitol dehydrogenase and $NADP^+$-dependent sorbitol-6-phosphate dehydrogenase during development of loquat (*Eriobotrya japonica* Lindl.) fruit. J. Japan. Soc. Hort. Sci. 69: 231–236.

Baroja-Fernandez, E., E. Etxeberria, F. J. Munoz, M. T. Moran-Zorzano, N. Alonso-Casajus, P. Gonzalez and J. Pozueta-Romero. 2006. An important pool of sucrose linked to starch biosynthesis is taken up by endocytosis in heterotrophic cells. Plant Cell Physiol. 47: 447–456.

Bellalouri, N., P. H. Brown and A. M. Dandekar. 1999. Manipulation of in vivo sorbitol production alters boron uptake and transport in tobacco. Plant Physiol. 119: 735–741.

Bellaloui, N., R. C. Yadavc, M. S. Chern, H. Hu, A. N. Gillen, C. Greve, A. M. Dandekar, P. C. Ronald and P. H. Brown. 2003. Transgenically enhanced sorbitol synthesis facilitates phloem-boron mobility in rice. Physiol. Plant. 117: 79–84.

Berüter, J. and M. E. S. Feusi. 1997. The effect of girdling on

carbohydrate partitioning in the growing apple fruit. J. Plant Physiol. 151: 277–285.

Bhatia, S. and R. Singh. 2002. Phytohormone-mediated transformation of sugars to starch in relation to the activities of amylases, sucrose-metabolizing enzymes in sorghum grain. Plant Growth Reg. 36: 97–104.

Biemelt, S., M. R. Hajirezaei, M. Melzer, G. Albrecht and U. Sonnewald. 1999. Sucrose synthase activity does not restrict glycolysis in roots of transgenic potato plants under hypoxia conditions. Planta 210: 41–49.

Brown, P. H., N. Bellaloui, H. Hu and A. M. Dandekar. 1999. Transgenically enhanced sorbitol synthesis phloem boron transport and increases tolerance of tobacco to boron deficiency. Plant Physiol. 119: 17–20.

Bussis, D., D. Heineke, U. Sonnewald, L. Willmitzer, K. Raschke and H-W. Heldt. 1997. Solute accumulation and decreased photosynthesis in leaves of potato plants expressing yeast-derived invertase either in the apoplast, vacuole or cytosol. Planta 202: 126–136.

Carvajal, M., A. Cerda and V. Martinez. 2000. Modification of the reponse of saline stressed tomato plants by the correction of cation disorders. Plant Growth Reg. 30: 37–47.

Chavez-Barcenas, A. T., J. J. Valdez-Alarcon, M. Marinez-Trujillo, L. Chen, B. Xoconostle-Cazares, W. J. Lucas and L. Herrera-Estrella. 2000. Tissue-specific and developmental pattern of expression of the rice *sps1* gene. Plant Physiol. 124: 641–653.

Chen, C. C. and R. E. Paull. 2000. Sugar metabolism and pineapple flesh translucency. J. Amer. Soc. Hort. Sci. 125: 558–562.

Chen, Y., O. Arakawa, M. Kasai and S. Sawada. 2008. Analysis of reduced photosynthesis in the apple leaf under sink-limited conditions due to girdling. J. Japan. Soc. Hort. Sci. 77: 115–121.

Cheng, L., R. Zhou, E. J. Reidel, T. D. Sharkev and A. M. Dandekar. 2005. Antisense inhibition of sorbitol synthesis leads to up-regulation of starch synthesis without altering CO_2 assimilation in apple leaves. Planta 220: 765–776.

Chengappa, S., M. Guilleroux, W. Phillips and R. Shields. 1999. Transgenic tomato plants with decreased sucrose synthase are unaltered in starch and sugar accumulation in the fruit. Plant Mol. Biol. 40: 213–221.

Chengappa, S., N. Loader and R. Shields. 1998. Cloning, expression, and mapping of a second tomato sucrose synthase gene, *Sus3* (Accession Nos. AJ011319 and AJ011534). Plant Physiol. 118: 1533.

Comparot, S., G. Lingiah and T. Martin. 2003. Function and specificity of 14-3-3 proteins in the regulation of carbohydrate and nitrogen metabolism. J. Exp. Bot. 54: 595–604.

D'Aoust, M. A., S. Yelle and B. Nguyen-Quoc. 1999. Antisense inhibition of tomato fruit sucrose synthase decreases fruit setting and the sucrose unloading capacity of young fruit. Plant Cell 11: 2407–2418.

Davies, C. and S. P. Robinson. 1996. Sugar accumulation in grape berries. Cloning of two putative vacuolar invertase cDNAs and their expression in grapevine tissues. Plant Physiol. 111: 275–283.

Deguchi, M., A. B. Bennett, S. Yamaki, K. Yamada, K. Kanahama and Y. Kanayama. 2006. An engineered sorbitol cycle alters sugar composition, not growth, in transformed tobacco. Plant Cell Environ. 29: 1980–1988.

Deguchi, M., Y. Koshita, M. Gao, R. Tao, T. Tetsumura, S. Yamaki and Y. Kanayama. 2004. Engineered sorbitol accumulation induces dwarfism in Japanese persimmon. J. Plant Physiol. 161: 1177–1184.

Doehlert, D. C. 1987. Ketose reductase activity in developing maize endosperm. Plant Physiol. 84: 830–834.

do Nascimento, J. R. O., B. R. Cordenunsi and F. M. Lajolo. 2000. Sucrose synthase activity and expression during development and ripening in bananas. J. Plant Physiol. 156: 605–611.

Duangsrisai, S., K. Yamada, N. A. Bantog, K. Shiratake, Y. Kanayama and S. Yamaki. 2007. Presence and expression of NAD^+-dependent sorbitol dehydrogenase and sorbitol-6-phosphate dehydrogenase genes in strawberry. J. Hort. Sci. Biotech. 82: 191–198.

Duangsrisai, S., K. Yamada, K. Shiratake, Y. Kanayama and S. Yamaki. 2008. Properties of sorbitol dehydrogenase in strawberry fruit and enhancement of the activity by fructose and auxin. J. Japan. Soc. Hort. Sci. 77: 318–323.

Eason, J. R., D. J. Ryan, L. M. Watson, T. Pinkney, D. Hedderley, M. C. Christey, R. H. Braun and S. A. Coupe. 2007. Suppressing expression of a soluble acid invertase (*BoINV2*) in broccoli (*Brassica oleracea*) delays postharvest floret senescence and down regulates cystein protease (*BoCP5*) transcription. Physiol. Plant. 130. 46–57.

Endler, A., S. Meyer, S. Schelbert, T. Schneider, W. Weschke, S. W. Peters, F. Keller, S. Baginsky, E. Martinoia and U. G. Schmidt. 2006. Identification of a vacuolar sucrose transporter in barley and Arabidopsis mesophyll cells by a tonoplast proteomic approach. Plant Physiol. 141: 196–207.

Estruch, J. J. and J. P. Beltran. 1991. Changes in invertase activities precede ovary growth induced by gibberellic acid in *Pisum sativum*. Physiol. Plant. 81: 319–326.

Etxeberria, E., E. Baroja-Fernandez, F. J. Munoz and J. Pozueta-Romero. 2005a. Sucrose-inducible endocytosis as a mechanism for nutrient uptake in heterotrophic plant cells. Plant Cell Physiol. 46: 474–481.

Etxeberria, E., P. Gonzalez and J. Pozueta-Romero. 2005b. Sucrose transport into citrus juice cells: Evidence for an endocytic transport system. J. Amer. Soc. Hort. Sci. 130: 269–274.

Gao, M., R. Tao, K. Miura, A. M. Dandekar and A. Sugiura. 2001. Transformation of Japanese persimmon (*Diospyros kaki* Thunb.) with apple cDNA encoding NADP-dependent sorbitol-6-phosphate dehydrogenase. Plant Sci. 160: 837–845.

Gao, Z., L. Maurousset, R. Lemoine, S-D. Yoo, S. van Nocker and W. Loescher. 2003. Cloning, expression, and characterization of sorbitol transporters from developing sour cherry fruit and leaf sink tissues. Plant Physiol. 131: 1566–1575.

Germain, V., B. Ricard, P. Raymond and P. H. Saglio. 1997. The role of sugars, hexokinase, and sucrose synthase in the determination of hypoxically induced tolerance to anoxia in tomato roots. Plant Physiol. 114: 167–175.

Geromel, C., L. P. Ferreira, S. M. C. Guerreiro, A. A. Cavalari, D. Pot, L. F. P. Pereira, T. Leroy, L. G. E. Vieira, P. Mazzafera and P. Marraccini. 2006. Biochemical and genomic analysis of sucrose metabolism during coffee (*Coffea arabica*) fruit development. J. Exp. Bot. 57: 3243–3258.

Getz, H. P. 1991. Sucrose transport in tonoplast vesicles of red beet roots is linked to ATP hydrolysis. Planta 185: 261–268.

Getz, H. P. and M. Klein. 1995. Characteristics of sucrose transport and sucrose-induced H^+ transport on the tonoplast of red beet (*Beta vulgaris* L.) storage tissue. Plant Physiol. 107: 459–467.

Giaquinta, R. 1977. Possible role of pH gradient and membrane ATPase in the loading of sucrose into the sieve tubes. Nature 267: 369–370.

Godt, D. E. and T. Roitsch. 1997. Regulation and tissue-specific distribution of mRNAs for three extracellular invertase isoenzymes of tomato suggests an important function in establishing and maintaining sink metabolism. Plant Physiol. 115: 273–282.

Godt, D. and T. Roitsch. 2006. The developmental and organ specific expression of sucrose cleaving enzymes in sugar beet suggests a transition between apoplasmic and symplasmic phloem unloading in the tap roots. Plant Physiol. Biochem. 44: 656–665.

Greuter, H. and F. Keller. 1993. Further evidence for stachyose and sucrose/H^+ antiporters on the tonoplast of Japanese artichoke (*Stachys sieblodii*) tubers. Plant Physiol. 101: 1317–1322.

Hansen, P. and K. Ryugo. 1979. Translocation and metabolism of carbohydrated fraction of ^{14}C-photosynathates in 'French' prune, *Prunus domestica* L. J. Amer. Soc. Hort. Sci. 104: 622–625.

Harada, T., S. Saitoh, T. Yoshiola and K. Ishizawa. 2005. Expression of sucrose synthase genes involved in enhanced elongation of pondweed (*Potamogeton distinctus*) turions under anoxia. Ann. Bot. 96: 683–692.

Hashizume, H., K. Tanase, K. Shiratake, H. Mori and S. Yamaki. 2003. Purification and characterization of two soluble acid invertase isozymes from Japanese pear fruit. Phytochemistry 63: 125–129.

Hayata, Y., X. X. Li and Y. Osajima. 2001a. Scurose accumulation and related metabolizing enzyme activities in seeded and induced parthenocarpic muskmelons. J. Amer. Soc. Hort. Sci. 126: 676–680.

Hayata, Y., X. X. Li and Y. Osajima. 2001b. CPPU promotes growth and invertase activity in seeded and seedless muskmelons during early growth stage. J. Japan. Soc. Hort. Sci. 70: 299–303.

Hirai, M. 1979. Sorbitol-6-phosphate dehydrogenase from loquat fruit. Plant Physiol. 63: 715–717.

Hubbard, N. L., S. C. Huber and D. M. Pharr. 1989. Sucrose phosphate synthase and acid invertase as determinants of sucrose concentration in developing muskmelon (*Cucumis melo* L.) fruits. Plant Physiol. 91: 1527–1534.

Hubbard, N. L., D. M. Pharr and S. C. Huber. 1990. Role of sucrose phosphate synthase in sucrose biosynthesis in ripening bananas and its relationship to the respiratory climacteric. Plant Physiol. 94: 201–208.

Hubbard, N. L., D. M. Pharr and S. C. Huber. 1991. Sucrose phosphate synthase and other sucrose metabolizing enzymes in fruits of various species. Physiol Plant. 82: 191–196.

Huber, S. C. and J. L. Huber. 1996. Role and regulation of sucrose-phosphate synthase in higher plants. Annu. Rev. Plant Physiol. Plant Mol. Biol. 47: 431–444.

Huber, S. C., J. L. Huber, P. C. Liao, D. A. Gage, R. W. McMichael, Jr., P. S. Chourey, L. C. Hannah and K. Koch. 1996. Phosphorylation of serine-15 of maize leaf sucrose synthase (Occurrence in vivo and possible regulatory significance). Plant Physiol. 112: 793–802.

Husain, S., H. N'Tchobo, B. Nguyen-Quoc, A. H. Kingston-Smith, B. J. Thomas and C. H. Foyer. 2003. Changes in activities of acid invertase isoforms affect sugar accumulation and composition during ripening of tomato fruit. J. Hort. Sci. Biotech. 78: 182–192.

Iida, M., N. A. Bantog, K. Yamada, K. Shiratake and S. Yamaki. 2004. Sorbitol- and other sugar induced expressions of the NAD^+-dependent sorbitol dehydrogenase gene in Japanese pear fruit. J. Amer. Soc. Hort. Sci. 129: 870–875.

Islam, S. 2001. Sucrose metabolism in domesticated cherry tomato, *Lycopersicon esculentum* var. cerasiforme Alef., and purification of sucrose synthase. J. Hort. Sci. Biotech. 76: 40–47.

Islam, S. and S. Khan. 2001. Seasonal fluctuations of carbohydrate accumulation and metabolism of three tomato (*Lycopersicon esculentum* Mill.) cultivars grown at seven sowing times. J. Hort. Sci. Biotech. 76: 764–770.

Itai, A. and T. Tanahashi. 2008. Inhibition of sucrose loss during cold storage in Japanese pear (*Pyrus pyrifolia* Nakai) by 1-MCP. Postharvest Biol. Technol. 48: 355–363.

Ito, A., H. Hayama and Y. Kashimura. 2002. Sugar metabolism in bud during flower bud formation: A comparison of two Japanese pear (*Pyrus pyrifolia* (Burm.) Nak.) cultivars possessing different flowering habits. Sci. Hortic. 96: 163–175.

Ito, A., H. Hayama and Y. Kashimura. 2003. Sugar metabolism in spur bud during flower bud formation: A comparison between exposed and shaded buds of Japanese pear (*Pyrus pyrifolia* (Burm.) Nak.) 'Kosui'. J. Japan. Soc. Hort. Sci. 72: 253–261.

Ito, A., H. Hayama and Y. Kashimura. 2005. Partial cloning and expression analysis of genes encoding NAD^+-dependent sorbitol dehydrogenase in pear bud during flower bud formation. Sci. Hortic. 103: 413–420.

Ito, A., H. Yoshioka, H. Hayama and Y. Kashimura. 2004. Reorientation of shoots to the horizontal position influences the sugar metabolism of lateral buds and shoot internodes in Japanese pear (*Pyrus pyrifolia* (Burm.) Nak.). J. Hort. Sci. Biotech. 79: 416–422.

Kanamaru, N., Y. Ito., S. Komori, M. Saito, H. Kato, S. Takahashi, M. Omura, J. Soejima, K. Shiratake, K. Yamada and S. Yamaki. 2004. Transgenic apple transformed by sorbitol-6-phosphate dehydrogenase cDNA. Switch between sorbitol and sucrose supply due to its gene expression. Plant Sci. 167: 55–61.

Kanayama, Y. 2009. Physiological roles of polyols in horticultural crops. J. Japan. Soc. Hort. Sci. 78: 158–168.

Kanayama, Y., N. Dai, D. Granot, M. Petreikov, A. Schaffer and A. B. Bennett. 1997. Divergent fructokinase genes are differentially expressed in tomato. Plant Physiol. 113: 1379–1384.

Kanayama, Y., D. Granot, N. Dai, M. Petreikov, A. Schaffer, A. Powell and A. B. Bennett. 1998. Tomato fructokinases exhibit differential expression and substrate regulation. Plant Physiol. 117: 85–90.

Kanayama, Y., M. Kogawa, M. Yamaguchi and K. Kanahama. 2005. Fructose content and the activity of fructose-related enzymes in the fruit of eating-quality peach cultivars and native-type peach cultivars. J. Japan. Soc. Hort. Sci. 74: 431–436.

Kanayama, Y., H. Mori, H. Imaseki and S. Yamaki. 1992. Nucleotide sequence of cDNA encoding NADP-sorbitol-6-phosphate dehydrogenase from apple. Plant Physiol. 100: 1607–1608.

Kanayama, Y., M. Watanabe, R. Moriguchi, M. Deguchi, K. Kanahama and S. Yamaki. 2006. Effects of low temperature and abscisic acid on the expression of the sorbitol-6-phosphate dehydrogenase gene in apple leaves. J. Japan. Soc. Hort. Sci. 75: 20–25.

Kaur, S., A. K. Gupta and N. Kaur. 2000. Effect of GA_3, kinetin and indole acetic acid on carbohydrate metabolism in chickpea seedlings germinating under water stress. Plant Growth Regul. 30: 61–70.

Kaur, S., A. K. Gupta and N. Kaur. 2002. Effect of osmo- and hydropriming of chickpea seeds on seeding growth and carbohydrate metabolism under water deficit stress. Plant Growth Regul. 37: 17–22.

Kaur, S., A. K. Gupta and N. Kaur. 2003. Indole acetic acid mimics the effect of salt stress in relation to enzymes of carbohydrate metabolism in chickpea seedlings. Plant Growth Regul. 39:

91–98.

Keller, F. and D. M. Pharr. 1996. Metabolism of carbohydrates in sinks and sources: Galactosyl-sucrose oligosaccharides. p. 157–183 In: E. Zamski and A. A. Schaffer (eds.). Photoassimilate distribution in plants and crops. Source-sink relationship. Marcel Dekker Inc, New York.

Kikuchi, K., S. Ishii, S. Fujimaki, N. Suzui, S. Matsuhashi, I. Honda, Y. Shishido and N. Kawachi. 2008. Real-time analysis of photoassimilate translocation in intact eggplant fruit using $^{11}CO_2$ and a positron-emitting tracer imaging system. J. Japan. Soc. Hort. Sci. 77: 199–205.

Kim, H-Y., J. C. Ahn, J-H. Choi, B. Hwang and D-W. Choi. 2007. Expression and cloning of the full-length cDNA for sorbitol-6-phosphate dehydrogenase and NAD-dependent sorbitol dehydrogenase from pear (*Pyrus pyrilolia* N.). Sci. Hortic. 112: 406–412.

Kim, H. S., J. H. Jeon, K. H. Choi, Y. H. Joung, B. I. Lee and H. Joung. 2000a. Regulation of starch contents in potato (*Solanum tuberosum* L.) by manipulation of sucrose synthase gene. J. Japan. Soc. Hort. Sci. 69: 243–249.

Kim, J., S. H. Jun, H. G. Kang, J. Lee and G. An. 2002. Molecular characterization of a GA-inducible gene, *Cvsus1*, in developing watermelon seeds. Mol. Cells 14: 255–260.

Kim, J. Y., A. Mahe, J. Brangeon and J. L. Prioul. 2000b. A maize vacuolar invertase, *IVR2*, is induced by water stress. Organ/tissue specificity and diurnal modulation of expression. Plant Physiol. 124: 71–84.

Klages, K., H. Donnison, J. Wunsche and H. Boldingh. 2001. Diurnal changes in non-structural carbohydrates in leaves, phloem exudate and fruit in 'Braeburn' apple. Aust. J. Plant Physiol. 28: 131–139.

Klann, E. M., R. T. Chetelat and A. B. Bennett. 1993. Expression of acid invertase gene controls sugar composition in tomato (*Lycopersicon*) friut. Plant Physiol. 103: 863–870.

Klann, E. M., B. Hall and A. B. Bennett. 1996. Antisense acid invertase (*T1V1*) gene alters soluble sugar composition and size in transgenic tomato fruit. Plant Physiol. 112: 1321–1330.

Koch, K. 2004. Sucrose metabolism: regulatory mechanisms and pivotal roles in sugar sensing and plant development. Curr. Opini. Plant Biol. 7: 235–246.

Koch, K. E. 1996. Carbohydrate-modulated gene expression in plants. Annu. Rev. Plant Physiol. Plant Mol. Biol. 47: 509–540.

Koch, K. E. and W. T. Avigne. 1990. Post-phloem, nonvascular transfer in citrus: kinetics, metabolism, and sugar gradients. Plant Physiol. 93: 1405–1416.

Komatsu, A., T. Moriguchi, K. Koyama, M. Omura and T. Akihama. 2002. Analysis of sucrose synthase genes in citrus suggests different roles and phylogenetic relationship. J. Exp. Bot. 53: 61–71.

Komatsu, A., Y. Takanokura, T. Moriguchi, M. Omura and T. Akihama. 1999. Differential expression of three sucrose-phosphate synthase isoforms during sucrose accumulation in citrus fruits (*Citrus unshiu* Marc.). Plant Sci. 140: 169–178.

Komatsu, A., Y. Takanokura, M. Omura and T. Akihama. 1996. Cloning and molecular analysis of cDNAs encoding three sucrose phosphate synthase isoforms from a citrus fruit (*Citrus unshiu* Marc.). Mol. Gen. Genet. 252: 346–351.

Konno, Y., T. Vedvick, L. Fitzmaurice and T. E. Mirkov. 1993. Purification, characterization and subcellular localization of soluble invertase from tomtao fruit. J. Plant Physiol. 141: 385–392.

Kubo, T., I. Hohjo and S. Hiratsuka. 2001. Sucrose accumulation and its related enzyme activities in the juice sacs of satsuma mandarin fruit from trees with different crop loads. Sci. Hortic. 91: 215–225.

Langenkämper, G., R. W. Fung, R. D. Newcomb, R. G. Atkinson, R. C. Gardner and E. A. MacRae. 2002. Sucrose-phosphate synthase genes in plants belong to three different families. J. Mol. Evol. 54: 322–332.

Langenkämper, G., R. McHale, R. C. Gardner and E. MacRae. 1998. Sucrose-phosphate synthase steady-state mRNA increases in ripening kiwifuit. Plant Mol. Biol. 36: 857–869.

Laporte, M. M., J. A. Galagan, A. L. Prasch, P. J. Vandervee, D. T. Hanson, C. K. Shewmaker and T. D. Sharkey. 2001. Promoter strength and tissue specificity effects on growth of tomato plants transformed with maize sucrose-phosphate synthase. Planta 212: 817–822.

Laporte, M. M., J. A. Galagan, J. A. Shapiro, M. R. Boersig, C. K. Shewmaker and T. D. Sharkey. 1997. Sucrose-phosphate synthase activity and yield analysis of tomato plants transformed with maize sucrose-phosphate synthase. Planta 203: 253–259.

Lester, G. E., L. S. Arias and M. Gomez-Lim. 2001. Muskmelon fruit soluble acid invertase and sucrose phosphate synthase activity and polypeptide profiles during growth and maturation. J. Amer. Soc. Hort. Sci. 126: 33–36.

Li, X. X., Y. Hayata, J. Yasukawa and Y. Osajima. 2002. Response of sucrose metabolizing enzyme activity to CPPU and p-CPA treatments in excised discs of muskmelon. Plant Growth Reg. 36: 237–240.

Lo Bianco, R. and M. Rieger. 2002. Roles of sorbitol and sucrose in growth and respiration of 'Encore' peaches at the tree developmental stages. J. Amer. Soc. Hort. Sci. 127: 297–302.

Lo Bianco, R., M. Rieger and S. J. S. Sung. 1999. Activities of sucrose and sorbitol metabolizing enzymes in vegetative sinks of peach and correlation with sink growth rate. J. Amer. Soc. Hort. Sci. 124: 381–388.

Lo Bianco, R., M. Rieger and S. J. S. Sung. 2000. Effect of drought on sorbitol and sucrose metabolism in sinks and sources of peach. Physiol. Plant. 108: 71–78.

Loescher, W. H. and J. D. Everard. 1996. Sugar alcohol metabolism in sink and sources. p. 185–207 In: E. Zamski and A. A. Schaffer (eds.). Photoassimilate distribution in plants and crops. Source-sink relationship. Marcel Dekker Inc, New York.

Lowell, C. A., P. T. Tomlinson and K. E. Koch. 1989. Sucrose-metabolizing enzymes in transport tissues and adjacent sink structures in developing citrus fruit. Plant Physiol. 90: 1394–1402.

Miranda Rossetto, M. R., E. Purgatoo, J. R. Oliveria do Nascimento, F. M. Lajolo and B. R. Cordenunsi. 2003. Effects of gibberellic acid on sucrose accumulation and sucrose biosynthesizing enzymes activity during banana ripening. Plant Growth Regul. 41: 207–214.

Miron, D. and A. A. Schaffer. 1991. Sucrose phosphate synthase, sucrose synthase, and invertase activities in developing fruit of *Lycopersicon esculentum* Mill. and the sucrose accumulating *Lycopersicon hirsutum* Humb. and Bonpl. Plant Physiol. 95: 623–627.

Miyamaoto, K., E. Ito, H. Yamamoto, J. Ueda and S. Kamisaka. 2000. Gibberellin-enhanced growth and sugar accumulation in growing subhooks of etiolated *Pisum sativum* seedlings: Effects of actinomycin D on invertase activity, soluble sugars and stem elongation. J. Plant Physiol. 156: 449–453.

Moriguchi, T. and S. Yamaki. 1988. Purification and characterization of sucrose synthase from peach (*Prunus persica*) fruit. Plant Cell Physiol. 29: 1361–1366.

Moriguchi, T., K. Abe, T. Sanada and S. Yamaki. 1992. Levels and role of sucrose synthase, sucrose-phosphate synthase, and

acid invertase in sucrose accumulation in fruit of Asian pear. J. Amer. Soc. Hort. Sci. 117: 274–278.

Moriguchi, T., Y. Ishizawa, T. Sanada, S. Teramoto and S. Yamaki. 1991. Role of sucrose synthase and other related enzymes in sucrose accumulation in peach fruit. J. Japan. Soc. Hort. Sci. 60: 531–538.

Moriguchi, T., T. Sanada and S. Yamaki. 1990. Seasonal fluctuations of some enzymes relating to sucrose and sorbitol metabolism in peach fruit. J. Amer. Soc. Hort. Sci. 115: 278–281.

Nakagawa, H., Y. Kawasaki, N. Ogura and H. Takehana. 1971. Purification and some properties of two types α-fructofuranosidase from tomato fruit. Agric. Biol. Chem. 36: 18–26.

Negm, F. B. and W. H. Loescher. 1979. Detection and characterization of sorbitol dehydrogenase from apple callus tissue. Plant Physiol. 64: 69–73.

Nguyen-Quoc, B., H. N'Tchobo, C. H. Foyer and S. Yelle. 1999. Overexpression of sucrose phosphate synthase increases sucrose unloading in transformed tomato fruit. J. Exp. Bot. 50: 785–791.

Nosarzewski, M. and D. D. Archbold. 2007. Tissue-specific expression of sorbitol dehydrogenase in apple fruit during early development. J. Exp. Bot. 58: 1863–1872.

Nosarszewski, M., A. M. Clements, A. B. Downie and D. D. Archbold. 2004. Sorbitol dehydrogenase expression and activity during apple fruit set and early development. Physiol. Plant. 121: 391–398.

Ofosu-Anim, J. and S. Yamaki. 1994a. Sugar content, compartmentation and efflux in strawberry tissue. J. Amer. Soc. Hort. Sci. 119: 1024–1028.

Ofosu-Anim, J. and S. Yamaki. 1994b. Sugar content and compartmentation in melon fruit and the restriction of sugar efflux from flesh tissue. J. Japan. Soc. Hort. Sci. 63: 685–692.

Ohta, K., R. Moriguchi, K. Kanahama, S. Yamaki and Y. Kanayama. 2005. Molecular evidence of sorbitol dehydrogenase in tomato, a non-Rosaceae plant. Phytochemistry 66: 2822–2828.

Ohyama, A., H. Ito, T. Sato, S. Nishimura, T. Imai and M. Hirai. 1995. Suppression of acid invertase activity by antisense RNA modifies the sugar composition of tomato fruit. Plant Cell Physiol. 36: 369–376.

Ohyama, A., S. Nishimura and M. Hirai. 1998. Cloning of cDNA for a cell wall-bound acid invertase from tomato (*Lycopersicon esculentum*) and expression of soluble and cell wall-bound invertases in plants and wounded leaves of *L. esculentum* and *L. peruvianum*. Genes Genet. Syst. 73: 149–157.

Opara, L. U. 2000. Fruit growth measurement and analysis. Hort. Rev. 24: 373–431.

Oura, Y., K. Yamada, K. Shiratake and S. Yamaki. 2000. Purification and characterization of a NAD^+-dependent sorbitol dehydrogenase from Japanese pear fruit. Phytochemistry 54: 567–572.

Pan, Q. H., M. J. Li, C. C. Peng, N. Zhang, X. Zou, K. Q. Zou, X. L. Wang, X. C. Yu, X. F. Wang and D. P. Zhang. 2005a. Abscisic acid activates acid invertases in developing grape berry. Physiol. Plant. 125: 157–170.

Pan, Q. H., X. C. Yu, N. Zhang, X. Zou, C. C. Peng, X. L. Wang, K. Q. Zou and D. P. Zhang. 2006. Activity, but not expression, of soluble and cell wall-bound acid invertases is induced by abscisic acid in developing apple fruit. J. Integrative Plant Biol. 48: 536–549.

Pan, Q. H., K. Q. Zou, C. C. Peng, X. L. Wang and D. P. Zhang. 2005b. Purification, biochemical and immunological characterization of acid invertases from apple fruit. J. Integrative Plant Biol. 47: 50–59.

Park, S. W., K. J. Song, M. Y. Kim, J-H. Hwang, Y. U. Shin, W-C. Kim and W-I. I. Chung. 2002. Molecular cloning and characterization of four cDNAs encoding the isoforms of NAD-dependent sorbitol dehydrogenase from the Fuji apple. Plant Sci. 162: 513–519.

Privat, I., S. Foucrier, A. Prins, T. Epalle, M. Eychenne, L. Kandalaft, V. Caillet, C. Lin, S. Tanksley, C. Foyer and J. McCarthy. 2008. Differential regulation of grain sucrose accumulation and metabolism in *Coffea Arabic* (Arabica) and *Coffea canephora* (Robusta) revealed through gene expression and enzyme activity analysis. New Phytol. 178: 781–797.

Ranwala, A. P., C. Suematsu and H. Masuda. 1992. Soluble and wall-bound invertase in strawberry fruit. Plant Sci. 84: 59–64.

Riesmeier, J. W., L. Willmitzer and W. B. Frommer. 1994. Evidence of an essential role of the sucrose transporter in phloem loading and assimilate partitioning. EMBO J. 13: 1–7.

Ruan, Y. L. and J. W. Patrick. 1995. The cellular pathway of postphloem sugar transport in developing tomato fruit. Planta 196: 434–444.

Saftner, R. A., J. Dai and R. E. Wyse. 1983. Sucrose uptake and compartmentation in sugar beet taproot tissue. Plant Physiol. 72: 1–6.

Saito, Y., N. Banatog, R. Morimoto, T. Horibe, K. Yamada and S. Yamaki. 2009. Stimulation of rooting from cuttings of strawberry runner plants by abscisic acid under high temperature condition. J. Japan. Soc. Hort. Sci. 78: 314–318.

Schaffer, A. A. and M. Petreikov. 1997. Inhibition of fructokinase and sucrose synthase by cytological levels of fructose in young tomato fruit undergoing transient accumulation of starch. Physiol. Plant. 110: 800–806.

Stommel, J. R. 1992. Enzymic components of sucrose accumulation in the wild tomato species *Lycopersicon peruvianum*. Plant Physiol. 99: 324–328.

Suzuki, Y., S. Odanaka and Y. Kanayama. 2001. Fructose content and fructose-related enzyme activity during the fruit development of apple and Japanese pear. J. Japan. Soc. Hort. Sci. 70: 16–20.

Tanase, K. and S. Yamaki. 2000a. Purification and charcterization of two sucrose synthase isoforms from Japanese pear fruit. Plant Cell Physiol. 41: 408–414.

Tanase, K. and S. Yamaki. 2000b. Sucrose synthase isozymes related to sucrose accumulation during fruit development of Japanese pear (*Pyrus pyrifolia* Nakai). J. Japan. Soc. Hort. Sci. 69: 671–676.

Tanase, K., K. Shiratake, H. Mori and S. Yamaki. 2002. Changes in the phosphorylation state of sucrose synthase during development of Japanese pear fruit. Physiol. Plant. 114: 21–26.

Tang, G. Q., M. Lüscher and A. Sturm. 1999. Antisense repression of vacuolar and cell wall invertase in transgenic carrot alters early plant development and sucrose partitioning. Plant Cell 11: 177–189.

Tao, R., S. L. Uratsu and A. M. Dandekar. 1995. Sorbitol synthesis in transgenic tobacco with apple cDNA encoding NADP-dependent sorbitol-6-phosphate dehydrogenase. Plant Cell Physiol. 36: 525–532.

Tateishi, A., T. Nakayama, K. Isobe, K. Nomura, K. Watanabe and H. Inoue. 2004. Changes in sugar metabolism enzyme activities in cultivars of two pumpkin species (*Cucurbita maxima* and *C. moschata*) during fruit development. J. Japan. Soc. Hort. Sci. 73: 57–59.

Teo, G., Y. Suzuki, S. L. Uratsu, B. Lampinen, N. Ormonde, W. K. Hu, T. M. DeJong and A. M. Dandekar. 2006. Silencing

leaf sorbtiol synthesis alters long-distance partitioning and apple fruit quality. Proc. Natl. Acad. Sci. USA 103: 18842–18847.

Wang, F., A. G. Smith and M. L. Brenner. 1993. Isolation and sequencing of tomato fruit sucrose synthase cDNA. Plant Physiol. 103: 1463–1464.

Wang, H. L., P. D. Lee, W. L. Chen, D. J. Huang and J. C. Su. 2000. Osmotic stress-induced changes of sucrose metabolism in cultures sweet potato cells. J. Exp. Bot. 51: 1991–1999.

Watari, J., Y. Kobae, S. Yamaki, K. Yamada, K. Toyofuku, T. Tabuchi and K. Shiratake. 2004. Identification of sorbitol trasporters expressed in the phloem of apple source leaves. Plant Cell Physiol. 45: 1032–1041.

Wormit, A., O. Trentmann, I. Feifer, C. Lohr, J. Tjaden, S. Meyer, U. Schmidt, E. Martinoia and H. E. Neuhaus. 2006. Molecular identification and physiological characterization of a novel monosaccharide transporter from Arabidopsis involved in vacuolar sugar transport. Plant Cell 18: 3476–3490.

Worrell, A. C., J.-M. Bruneau, K. Summerfelt, M. Boersig and T. A. Voelker. 1991. Expression of a maize sucrose phosphate synthase in tomato alters leaf carbohydrate patitioning. Plant Cell 3: 1121–1130.

Wu, L-L., J. P. Mitchell, N. S. Cohn and P. B. Kaufman. 1993. Gibberellin (GA_3) enhances cell wall invertase activity and mRNA levels in elongating dwarf pea (*Pisum sativum*) shoots. Int. J. Plant Sci. 154: 280–289.

Wu, G. L., X. Y. Zhang, L. Y. Zhang, Q. H. Pan, Y. Y. Shen and D. P. Zhang. 2004. Phloem unloading in developing walnut fruit is symplasmic in the seed pericarp, and apoplasmic in the fleshy pericarp. Plant Cell Physiol. 45: 1461–1470.

Yamada, K., T. Kojima, N. Bantog, T. Shimoda, H. Mori, K. Shiratake and S. Yamaki. 2007. Cloning of two isoforms of soluble acid invertase of Japanese pear and their expression during fruit development. J. Plant Physiol. 164: 746–755.

Yamada, K., H. Mori and S. Yamaki. 1999. Gene expression of NAD-dependent sorbitol dehydrogenase during fruit development of apple (*Malus pumila* Mill. var. *domestica* Schneid.). J. Japan. Soc. Hort. Sci. 68: 1099–1103.

Yamada, K., N. Niwa, K. Shiratake and S. Yamaki. 2001. cDNA cloning of NAD-dependent sorbitol dehydrogenase from peach fruit and its expression during fruit development. J. Hort. Sci. Biotech. 76: 581–587.

Yamada, K., Y. Oura, H. Mori and S. Yamaki. 1998. Cloning of NAD-dependent sorbitol dehydrogenase from apple fruit and gene expression. Plant Cell Physiol. 39: 1375–1379.

Yamada, K., Y. Suzue, S. Hatano, M. Tsukuda, Y. Kanayama, K. Shiratake and S. Yamaki. 2006. Changes in the activity and gene expression of sorbitol- and sucrose-related enzymes associated with development of 'La France' pear fruit. J. Japan. Soc. Hort. Sci. 75: 38–44.

Yamaki, S. 1984. Isolation of vacuoles from immature apple fruit flesh and compartmentaion of sugars, organic acids, phenolic compounds and amino acids. Plant Cell Physiol. 25: 151–166.

Yamaki, S. 1987. ATP-promoted sorbitol transport into vacuoles isolated from apple fruit. Plant Cell Physiol. 28: 557–564.

Yamaki, S. and M. Ino. 1992. Alteration of cellular compartmentation and membrane permeability to sugars in immature and mature apple fruit. J. Amer. Soc. Hort. Sci. 117: 951–954.

Yamaki, S. and K. Ishikawa. 1986. Roles of four sorbitol related enzymes and invertase in the seasonal alteration of sugar metabolism in apple tissue. J. Amer. Soc. Hort. Sci. 111: 1099–1103.

Yamaki, S. and T. Moriguchi. 1989. Seasonal fluctuation of sorbitol related enzymes and invertase activities accompanying maturation of Japanese pear (*Pyrus serotina* Rehder var. *culta* Rehder) fruit. J. Japan. Soc. Hort. Sci. 57: 602–607.

Yu, J. Q., L. F. Huang, W. H. Hu, Y. H. Zhou, W. H. Mao, S. F. Ye and S. Nogues. 2004. A role of brassinosteroids in the regulation of photosynthesis in *Cucumis sativus*. J. Exp. Bot. 55: 1135–1143.

Yu, X., X. Wang, J. Fan, H. Tian and C. Zheng. 2007. Cloning and characterization of a sucrose phosphate synthase-encoding gene from muskmelon. J. Amer. Soc. Hort. Sci. 132: 557–562.

Zeng, Y., Y. Wu, W. T. Avigne and K. E. Koch. 1998. Differential regulation of sugar-sensitive sucrose synthases by hypoxia and anoxia indicate complementary transcriptional and posttranscriptional responses. Plant Physiol. 116: 1573–1583.

Zeng, Y., Y. Wu, W. T. Avigne and K. E. Koch. 1999. Rapid repression of maize invertases by low oxygen. Invertase/sucrose synthase balance, sugar signaling potential, and seedling survival. Plant Physiol. 121: 599–608.

Zhang, L. Y., Y. B. Peng, S. Pelleschi-Travier, Y. Fan, Y. F. Lu, Y. M. Lu, X. P. Gao, Y. Y. Shen, S. Delrot and D. P. Zhang. 2004. Evidence for apoplasmic phloem unloading in developing apple fruit. Plant Physiol. 135: 574–586.

Zhang, X. Y., X. L. Wang, X. F. Wang, G. H. Xia, Q. H. Pan, R. C. Fan, F. Q. Wu, X. C. Yu and D. P. Zhang. 2006. A shift of phloem unloading from symplasmic to apoplasmic pathway is involved in developmental onset of ripening in grape berry. Plant Physiol. 142: 220–232.

Zhou, L. and R. E. Paull. 2001. Sucrose metabolism during papaya (*Carica papaya*) fruit growth and ripening. J. Amer. Soc. Hort. Sci. 126: 351–357.

Zhou, L. and B. Quebedeaux. 2003. Changes in photosynthesis and carbohydrate metabolism in mature apple leaves in response to whole plant source-sink manipulation. J. Amer. Soc. Hort. Sci. 128: 113–119.

Zhou, L., C. C. Chen, R. Ming, D. A. Christopher and R. E. Paull. 2003a. Apoplastic invertase and its enhanced expression and post-translation control during fruit maturation and ripening. J. Amer. Soc. Hort. Sci. 128: 628–635.

Zhou, L., D. A. Christopher and R. E. Paull. 2000. Defoliation and fruit removal effects on papaya fruit production, sugar accumulation, and sucrose metabolism. J. Amer. Soc. Hort. Sci. 125: 644–652.

Zhou, R., L. Cheng and R. Wayne. 2003b. Purification and characterization of sorbitol-6-phosphate phosphatase from apple leaves. Plant Sci. 165: 227–232.

Zhou, R., L. Cheng and A. M. Dandekar. 2006. Down-regulation of sorbitol dehydrogenase and up-regulation of sucrose synthase in shoot tips of the transgenic apple trees with decreased sorbitol synthesis. J. Exp. Bot. 57: 3647–3657.

Zhou, R., R. C. Sicher and B. Quebedeaux. 2002. Apple leaf sucrose-phosphate synthase is inhibited by sorbitol-6-phosphate. Funct. Plant Biol. 29: 569–574.

Zimmermann, M. H. and H. Ziegler. 1975. List of sugars and sugar alcohols in sieve-tube exudates. p. 480–503. In: M. H. Zimmermann and J. A. Milburn (eds.). Transport in plants I. Phloem transport. Springer-Verlag, Berlin.