MOLECULAR MANIPULATION OF SUCROSE METABOLISM

By

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SUCROSE plays a central role in plants as it is the primary product of photosynthesis and is the major form of carbohydrate translocated from the leaves and utilised as an energy source for growth or storage reserves (Ap Rees 1987). Sugarcane belongs to the group of plants which are characteristically the world’s most productive, utilising the C₄ mechanism of CO₂ fixation for the production of approximately 70% of the world’s sucrose.

Although lagging behind the rapid advances being made in model systems and crop species grown more extensively around the globe, sugarcane researchers are beginning to implement the exciting new molecular approaches available to dissect the biochemical processes underpinning sucrose accumulation in sugarcane, as well as applying our improved understanding to the generation of alternative products. There are a number of requisite tools which are at a significantly more advanced stage of development in model and other crop systems as compared with sugarcane. These tools include:

1. Transformation methods and tissue culture procedures to generate a sufficiently high number of regenerants.
2. Isolation of suitable genes from eukaryotic or prokaryotic sources.
3. Identification of suitable gene promoter elements to direct strong tissue/organelle and cell specific expression.
4. Identification of suitable targeting sequences to direct the gene product to the appropriate subcellular location (organelle or compartment).

To date, significant development of such tools has been made in sugarcane, the most recent of which will be described during these proceedings.

The first successful transformation of sugarcane callus by particle bombardment was reported in 1992 (Bower and Birch, 1992) whereas successful incorporation of foreign genes by Agrobacterium was reported in 1998 (Arencibia et al., 1998; Elliott et al., 1998). A significant issue which has arisen as a consequence of the transformation and/or tissue culture process is somaclonal variation. This has had a profound impact on our ability to successfully interpret the consequence of genetic manipulations, particularly of genes encoding enzymes involved in primary metabolism. Agronomic parameters, including yield and CCS (commercial cane sugar), measured during field trials in transgenic sugarcane aimed at down regulating polyphenol oxidase activity, were shown to be significantly reduced (Vickers et al., in preparation) Such results highlight the importance of improving the tissue culture/transformation process if transgenic sugarcane is to be successfully cultivated.

During the course of this meeting, presentation of transgenic experiments aimed at manipulating primary sucrose metabolism in planta will be made. The targets chosen for manipulation have been derived from biochemical measurements made over a number of decades from a number of laboratories in different countries and are directed towards key enzymes involved in the sucrose synthesis and cleavage pathways. A recent study from the Hawaiian laboratory long renowned for ground-breaking research, describes the manipulation of invertases in sugarcane cells and highlights the significance of these enzymes in the sucrose accumulation process (Ma et al., 2000).

As the primary focus of this workshop is molecular biology, I would like to provide a brief overview of the contribution of this remarkable tool to furthering the understanding of metabolism in general and carbohydrate metabolism in particular. Following the successful introduction of foreign genes into plants, the preliminary steps taken to influence metabolism were attempted through manipulation of single genes encoding potentially key enzymes.

Perhaps the single most comprehensive program has been directed at the manipulation of starch metabolism in the laboratory of Lothar Wilmitzer by altering the levels of a series of enzymes in source and sink pathways of potato. A principal outcome of this work has been the realisation of the subtlety, complexity and plasticity of plant metabolic mechanisms.

Notable successes however, have also been reported in model plant systems through the manipulation of single key enzymes believed to be major contributors to the control of flux through the sucrose synthesis pathway.

Successful manipulation of SPS in tomato resulting in greater dry weight, number of fruit, and higher sugar concentration was first reported 10 years ago (Worrell et al., 1991; Laporte et al., 1997). However, attempts to emulate this success in a range of plants, including crop species, have proven inconclusive at best. Such an outcome is unfortunate as SPS is also thought to play a key role in the process of sucrose accumulation in sugarcane.

The essence of the sucrose accumulation model in sugarcane, originally proposed by Glasziou and Gayler (1972) and based upon a dynamic cycle of sucrose synthesis and degradation, has been attributed to the change in the ratio of SPS : SAI activity (Zhu et al., 1997). The modulation of the activity of these enzymes, may dictate the timing of the sucrose

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accumulation process and indeed the final sucrose concentration.

Significant steps in unravelling the movement of photoassimilate from the source to the sink have also been made through the application of molecular techniques. Seminal work in model species illustrating the importance of sugar transporters (Lalonde et al., 1999; Williams et al., 2000) as well as the role of invertases (Sturm, 1999), have been highlighted by such techniques. The challenge in store for researchers working on sugarcane is to extend the knowledge derived from model species in order to strategically focus limited resources on the most appropriate targets for successful manipulation to achieve increased sucrose accumulation. Some attention should perhaps be directed towards more global mechanisms which control entire suites of genes impacting upon key processes influencing sucrose accumulation.

REFERENCES


