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Dendranthema species and F_1 hybrids from the different habitats in Korea were collected to investigate the variation of their somatic chromosomes. The results in this study revealed series of polyploidy in *Dendranthema* from diploids ($2n=2x=18$) up to octoploids ($2n=8x=72$) with the basic chromosome number $x=9$. Some of the F_1 hybrids have also intraspecific polyploids as well as aneuploids. Due to *Dendranthema* has shown to comprise series of polyploidy ($2x, 4x, 6x, 8x$), it was possible that polyploidy ($2x, 4x, 5x, 6x$) individuals also produced in F_1 hybrid progeny. Mostly the range of variation was found as diploid ($2n=2x=18$) individuals with *D. boreale*, *D. makinoi* and *D. lineare*, irrespective of whether they occurred together with tetraploid ($2n=4x=36$) of *D. indicum*, *D. zawadskii* ssp. *lucidum*, *D. zawadskii* ssp. *latilobum* var. *leiophyllum*, *D. japonicum*, and hexaploid ($2n=6x=54$) of *D. indicum* for. *albescens*, *D. zawadskii* ssp. *acutilobum*, *D. zawadskii* ssp. *acutilobum* var. *tenuisectum*, *D. zawadskii* ssp. *acutilobum* var. *alpinum*, *D. zawadskii* ssp. *naktongense*, *D. zawadskii* ssp. *yezoense*, *D. zawadskii* ssp. *latilobum* and octoploids ($2n=8x=72$) of *D. zawadskii* ssp. *coreanum*. Number of chromosomes in different species suggests that the hybridization between individuals had repeatedly occurred not only in *D. zawadskii* complex, but also in *D. boreale* and *D. indicum*. As morphological characteristic, intermediate individuals between *D. zawadskii* complex and *D. indicum* suggests that the hybridization occurred with different species respectively. Artificial reciprocal crosses between *D. japonicum* ($2n=36$, tetraploid) and *D. indicum* for *albescens* ($2n=54$, hexaploid) were made and produced F_1 hybrid ($2n=45$, pentaploid). The cytogenetic investigation in *Dendranthema* has been largely directed to ascertain chromosome numbers and the descriptions of chromosome morphology.

2:45–3:00 pm

In Vitro Propagation of *Colutea gifana*, an Endangered Species in Iran

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In vitro methods provide a variety of tools to supplement traditional methods for collecting, propagating and preserving endangered plant species. In this study, an efficient protocol was developed for in vitro propagation of *Colutea gifana*, a rare and endangered plant species with limited reproductive capacity that grows in a very narrow area of Iran. Single nodes explants were used for a series of experiments to select appropriate disinfection method and growth regulators for establishment, proliferation and rooting stages. Explants showed the highest establishment percent after 15 min treatment with 2% sodium hypochlorite (NaOCl) cultured in MS medium plus $2.2 \mu\text{M}$ 6-benzylaminopurine (BAP) and $1 \mu\text{M}$ indole-3-butyric acid (IBA). BA was more effective cytokinin in comparison to Thidiazuron (TDZ) and Kinitin (Kn) in proliferation stage and the best result was obtained with $8.8 \mu\text{M}$ BA. In vitro rooting of proliferated shoots were induced in half-strength MS medium with both tested auxins i.e. IBA and α -naphthaleneacetic acid (NAA). Eighty percent of the plantlets were successfully acclimatized to ex vitro conditions, exhibiting normal development. These plantlets can be used to replenish declining populations in the wild to conserve *C. gifana* from extinction and also for further studies about this species.

Thursday, August 5, 2010

Springs H & I

Oral Session 40:

Crop Physiology: Fruit Crops

1:00–1:15 pm

Cold Hardiness of Apple Rootstock Trunk and Root Tissues

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Ungrafted clonal apple rootstocks were stored at 1°C until controlled exposure of the trunk and shoots to $-12, -15, -18, -21, -24$ and -27°C in Dec. 2009. To protect roots from cold temperature injury, the roots were insulated by peat-based media inside a Styrofoam container. Temperature of the media remained above -1°C during exposure to subzero temperatures. Internal browning of phloem, cambium and xylem was rated on a scale of

1 to 5 in the lower and upper sections of the tree trunk. Browning of Bud.9 xylem was not affected by any of the temperatures, but purple coloring made visual rating difficult. P.2 was also unaffected. Browning of the xylem occurred at a temperature of -24°C in G.11 and G.30, and at -21°C in M.26. Xylem browning was most severe in M.26, G.30 and G.11 which had a rating of 4.0, 3.3 and 2.8, respectively at the coldest temperature, -27°C . Browning of the phloem and cambium was less severe. Trees were grown in a greenhouse for two months after which shoot growth was measured as an assessment of survival. In Feb. 2010, the same rootstock cultivars were subjected to -10 , -13 , -15 , -18 , -21 and -24°C with the roots exposed to these temperatures. Roots were placed inside plastic bags to protect them from desiccation. Relative electrical conductivity of fine root segments increased with decrease in temperature, but did not vary between the different rootstocks. Trees were grown in a greenhouse to assess tree survival.

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1:15–1:30 pm

Responses of Transgenic Apple Trees with Decreased Sorbitol Synthesis to Nitrogen Deficiency

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Sorbitol is the main photosynthetic end-product and the primary translocated form of carbohydrates in apple. When the expression of aldose-6-phosphate (A6PR), the key enzyme in sorbitol synthesis, was decreased via antisense inhibition in 'Greensleaves' apple, both A6PR activity and sorbitol concentration were significantly decreased. As a result, hexose phosphates accumulated in the cytosol at the expense of inorganic phosphate, leading to up-regulation of starch synthesis, organic acid metabolism and amino acid metabolism in the antisense plants. To determine the responses of the transgenic plants to N deficiency, a transgenic line (A10) and the untransformed control (CK) were supplied with 0.5, 2.5, 7.5 or 15 mM nitrogen in Hoagland's solution via fertigation for 7 weeks from mid-May. A10 leaves had significantly higher photosynthesis and chlorophyll concentrations than CK at both 0.5 and 2.5 mM N treatments. 2-oxoglutarate concentration in A10 leaves remained unchanged whereas that in CK increased with decreasing N supply. Oxaloacetate concentration increased with decreasing N supply in both A10 and CK, but it increased to a lesser extent in A10 than in CK. Both soluble proteins and total free amino acids in A10 and CK decreased with decreasing N supply, but A10 had significantly higher levels of soluble proteins and higher levels of aspartate

and glutamate than CK under N deficiency. In addition, A10 leaves had much higher levels of trehalose and maltose than CK under N deficiency. These results indicate that more organic acids are used in amino acid and protein synthesis to maintain higher photosynthesis in the transgenic plants under N deficiency and trehalose signaling might be involved in the responses of transgenic plants to nitrogen deficiency.

1:30–1:45 pm

Metabolism of Organic Acids, Nitrogen and Amino Acids in Chlorotic Leaves of 'Honeycrisp' Apple with Excessive Accumulation of Carbohydrates

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Metabolite profiles and activities of key enzymes in the metabolism of organic acids, nitrogen and amino acids were compared between chlorotic leaves and normal leaves of 'Honeycrisp' apple to understand how accumulation of non-structural carbohydrates affects the metabolism of organic acids, nitrogen and amino acids. Higher concentrations of non-structural carbohydrates, particularly starch, fructose, sucrose, maltose, maltitol, and trehalose were accumulated in chlorotic leaves, and correspondingly much lower CO_2 assimilation was observed in chlorotic leaves than in normal leaves, confirming feedback inhibition of photosynthesis in chlorotic leaves. Dark respiration and activities of several key enzymes in glycolysis and tricarboxylic acid (TCA) cycle, ATP-phosphofructokinase, pyruvate kinase, citrate synthase, aconitase and isocitrate dehydrogenase, were significantly higher in chlorotic leaves than in normal leaves. However, concentrations of most organic acids including pyruvate, phosphoenolpyruvate (PEP), oxaloacetate, 2-oxoglutarate, malate and fumarate, and activities of key enzymes involved in the anapleurotic pathway including PEP carboxylase, NAD-malate dehydrogenase and NAD-malic enzyme were significantly lower in chlorotic leaves than in normal leaves. Concentrations of total soluble proteins and most free amino acids were significantly lower in chlorotic leaves than in normal leaves. Activities of key enzymes in nitrogen assimilation and amino acid synthesis, including nitrate reductase, glutamine synthetase, ferredoxin and NADH-dependent glutamate synthase, and glutamate pyruvate transaminase were significantly lower whereas activities of glutamate dehydrogenase and glutamate oxaloacetate transaminase were significantly higher in chlorotic leaves than in normal leaves. It was concluded that, in response to excessive accumulation of non-structural carbohydrates, glycolysis and TCA cycle are up-regulated to "consume" the excess carbon available whereas the anapleurotic pathway, nitrogen assimilation and amino acid synthesis are down-regulated to reduce the overall rate of amino acid and protein synthesis.