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MICROPROPAGATION OF *ACONITUM UNCINATUM*: GROWTH REGULATOR AND ANTIOXIDANT SCREENING USING THE SURFACE-RESPONSE ANALYSIS METHOD

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Aconitum uncinatum L., a herbaceous native, is not widely available for the public because of difficulties in producing plants via conventional methods. The initial objective was to find a growth regulator combination and an antioxidant concentration that provided increased proliferation rates. Growth regulator combinations examined were BA (0, 11.1, 22.2, 44.4 μM), 2iP (0, 24.6, 49.2 μM), kinetin (0, 23.2, 46.5 μM), IAA (0, 28.5, 57.1 μM), and NAA (0, 26.9, 53.71 μM). Antioxidants examined were ascorbic acid (0, 150, 500 ppm), citric acid (0, 150, 500 ppm), PVP (0, 250, 500 ppm), PVPP (0, 250, 500 ppm), and MPP (0, 0.5, 1 ppm). Surface response analysis was used to select 30 (of more than 3000 possible) combinations of growth regulator or antioxidant concentrations. Experiments were subcultured every 4 weeks and terminated at 8 weeks. Total-shoot proliferation was significantly greater on media containing 44.4 μM BA + 46.5 μM kinetin. There were six antioxidant combinations that were no different from the control (150 ppm each ascorbic acid and citric acid). Further experiments looking at various BA + kinetin and antioxidant concentrations are needed to refine the media for maximal proliferation.

OPTIMAL CONDITIONS FOR CELL SUSPENSION CULTURES OF *THLASPI CAERULESCENS* AND *BRASSICA NAPUS*

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Thlaspi caerulescens (*Brassicaceae*), known as a Zn hyper accumulator, is able to accumulate and tolerate Zn, Ni, Cu, and Cd at high concentrations in its biomass. We are examining the feasibility of using cell suspensions of *T. caerulescens* and *B. napus* to study the effect of selected heavy metals on growth and nutrient uptake. Callus was initiated by culturing seedlings on basal medium containing MS salts supplemented with MS or B5 vitamins, 1, 2, 5, or 10 mg 2,4-D/liter, and 0.7% Phytagar. Cell suspensions were initiated by transferring calli to liquid basal medium containing MS or B5 vitamins, and 1 or 2 mg 2,4-D/liter, and were incubated on a gyratory shaker at 120 rpm. Growth of suspensions inoculated at 0.2, 0.4, or 0.6 g/25 ml was monitored for 13 days. Optimal conditions required to initiate and maintain suspension cultures of *T. caerulescens* and *B. napus* include MS medium supplemented with B5 vitamins and 1 mg 2,4-D/liter, an inoculation density of 0.4 g/25 ml, and a 2-week subculture schedule.

GIBBERELIC ACID DURING PRIMING OF PURPLE CONEFLOWER [*ECHINACEA PURPUREA* (L) MOENCH.] SEEDS IMPROVES GERMINATION AND SEEDLING EMERGENCE

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Purple coneflower seeds following priming (-0.04 MPa, 10 days,

15C, darkness) osmotically in polyethylene glycol 8000 (PEG) or matrically in expanded no. 5 vermiculite had greater germination rate and synchrony at continuous 20C or 30C than untreated seeds, but germination percentage was unaffected. Inclusion of 5.5×10^{-2} M gibberellic acid (GA₃, as ProGibb Plus 2X, Abbott Laboratories, N. Chicago, Ill.) further improved germination rate and synchrony at 20C, but not at 30C. In a greenhouse study (30C day/27C night, July-August natural light), seeds primed in PEG or vermiculite containing GA₃ compared to untreated seeds had 6 percentage points higher maximum emergence (ME), 3.3 fewer days to 50% ME, 1.9 fewer days between 10% and 90% ME, 116% greater shoot dry weight, and 125% longer leaves at 16 days after planting in peat-lite. Inclusion of ethephon (0.01 M, as Florel) either alone or with GA₃ during priming provided no benefit to seed germination or seedling emergence. Moistened vermiculite substituted for PEG solution as a priming medium for purple coneflower seeds, the priming benefit on seedling emergence and growth being enhanced by 5.5×10^{-2} M GA₃ inclusion in the priming media.

INTELLECTUAL PROPERTY RIGHTS UPDATE: PLANT VARIETY PROTECTION ACT AMENDMENTS

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The Plant Variety Protection (PVP) Act provides intellectual property rights to new varieties of seed-reproduced plants. Eligible varieties must demonstrate that they are uniform, stable, and distinct from all other varieties. In 1991 the International Union for the Protection of New Varieties of Plants (UPOV) adopted a new Convention. As a member of UPOV, the United States needed to amend the PVP Act to conform to the 1991 UPOV Convention. Amendments to the PVP Act were signed by President Clinton on 6 Oct. 1994, and will become effective on 4 Apr. 1995. Among other changes, these amendments will provide protection to tuber-propagated varieties and first-generation hybrids. An overview of the amendments and a comparison of rights granted under PVP and plant patents will be presented.

COMPARISON OF RAPD MARKERS BETWEEN HEAT-TOLERANT AND INTOLERANT CULTIVARS OF LIMA BEANS

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Lima beans are an important vegetable crop to the processing industry in Delaware, but yields in Delaware are below other areas due to heat. The objective was to correlate RAPD markers from heat-tolerant and intolerant cultivars with phenotypic data. Twenty-five primers were used, 10 of which generated 25 polymorphic bands among 11 cultivars. MDS analysis of genetic distance among the cultivars shows segregation into two major clusters, with Kingston as a distant outlier. Kingston's position can be correlated to published data reporting its consistently good yields even when temperatures are high. The results of this study indicate RAPD markers may be used to

screen for cultivars that have high yield potentials despite high temperatures. Further studies to screen F₂ and inbreds will determine the usefulness of these markers in breeding programs.

PRELIMINARY LOCALIZATION OF IPTASE IN TRANSGENIC *NICOTIANA*

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Nicotiana transformed with the *isopentenyl transferase (ipt)* gene from *Agrobacterium tumefaciens* was fixed in 1% glutaraldehyde and 4% formaldehyde for 1 h. Grids were treated with polygonal anti-IpTase antibody raised in rabbits and visualized with 10 nm protein-A-labeled colloidal gold. Initial localization was performed on *Nicotiana* transformed with the *ipt* gene under the control of the 35S promoter from cauliflower mosaic virus. Colloidal gold was found throughout the cell, including the cell wall, vacuole, and rough ER. Cell wall and vacuole labeling appears to be due to nonspecific binding and is greatly reduced by a BSA block. Colloidal gold label on rough ER provides preliminary evidence that translation occurs here rather than on free polysomes. General reaction throughout the cell indicates cytoplasmic activity of the enzyme. Future research will attempt to localize IPTase in wild-type *Nicotiana* and in plants transformed with the *ipt* gene under the control of the hsp 70 heat shock promoter.

THE DYNAMICS OF HYDRAULIC LIFT IN PEACH TREE

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The split root technique was used to study water afflux in peach [*Prunus persica* (L.) Batsch] from wet to dry soil through root systems that bridge wet and dry soil. Peach trees conduct hydraulic lift (HL) to ameliorate water deficits in dry soil layers, under conditions of low transpirational demand. The objectives of this study were to examine the magnitude of HL in peach and to determine its effect on nutrient uptake from dry soil. In addition, the split root system was used to measure peach water uptake from soil supporting 'Kentucky 31' tall fescue [*Festuca arundinaceae* (Schreb)] and determine the diurnal partitioning of water use from covered and bare soil treatments. A Scholander pressure bomb was used to record hourly measurements of water potentials (10 AM to 4 PM), daily for a total of 14 days in 3 replicates (1 tree/rep.). Leaf stomatal resistance was measured using a porometer, simultaneously with the water potential measurements. The CR 7 datalogger was used to record water transfer into the dry root section. ¹⁵N was applied in the 15-30 cm root zone, and the concentration in the leaves was determined using a mass spectrometer. Results obtained will be discussed in relation to objectives stated above.

MOSAIC: A MULTIMEDIA PLATFORM FOR TEACHING PLANT MATERIALS

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A Mosaic tour of the Univ. of Delaware Botanic Gardens is available on Internet using the Mosaic server and provides fact sheets about herbaceous and woody plants. Each sheet is a mosaic page with in-lined images that include hyper-links to a sound file of the pronunciation of the Latin plant name and images of the plant, including buds, flowers, fruits, leaves, and other characteristics. A map of the garden identifying the plant's location is provided on each page to orient visitors and students. There are currently over 1500 images in development. The tour uses map images and key words to help students identify and select plants. Composite views illustrating the same characteristic from several plants provide a visual method to aid students in identification of unknown specimens. Database searches provide a method to search for plant information in the tour, and in the future will provide lists of plants with specific characteristics. The Mosaic Tour of the Univ. of Delaware Botanic Gardens can be accessed via URLs:<http://bluehen.ags.udel.edu/ugarden.html>.

COMPARISON OF FISH HYDROLYSATE AND INORGANIC FERTILIZERS FOR LOWBUSH BLUEBERRY

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A commercial lowbush blueberry (*Vaccinium angustifolium* Ait.) field deficient in leaf N and P was used to compare organic and inorganic fertilizers. In a RCB design with eight replications of 12 treatments, experimental plots received 33.6 or 67.2 kg-ha⁻¹ rates of N (urea), P (23% phosphoric acid), N + P (DAP), N + P + K (5-10-5), or N + P + K (fish hydrolysate, 242). Fertilizer containing N alone was as effective in raising N leaf concentrations as those containing N and P. However, leaf P concentrations were raised more by fertilizer providing N and P than only P. Fish hydrolysate fertilizer was as effective as 5-10-5 in raising leaf N, P, and K concentrations in prune and crop year leaf samples. At the 67.2 kg-ha⁻¹ rate, fish hydrolysate, N, NP and NPK increased stem length, N and NP increased flower bud density and fish hydrolysate, N and NPK increased yield compared to the control.

COLD TOLERANCE OF CRANBERRY BUDS AND FRUIT: LABORATORY AND FIELD STUDIES

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Cold tolerance of cranberry (*Vaccinium macrocarpon* Ait.) flower buds (spring) and fruit had previously been investigated for the cultivars Early Black (EB) and Howes (H), leading to predictors of cold tolerance based on appearance of the buds (size and growth) and fruit (color). We studied these cultivars along with 'Ben Lear' (BL) (buds only) and 'Stevens' (S) using controlled temperatures to determine the accuracy of predicting cold damage. BL was the least tolerant cultivar in early spring, both BL and S were less tolerant than EB and H during budbreak (-2.8C vs. -3.9C) and elongation (-2.8C vs. -1.4C), and all survived any exposure to -1.4C. EB fruit were tolerant of -5C once maximum color was achieved and 2 weeks later would tolerate short exposures to -6.5C or less. H fruit developed deep tolerance (below 6.5C) by November in only 1 year out of 2. S fruit were least tolerant: -5C for short periods at M maturity. The phenological model used to predict cold tolerance of flower buds was 48% accurate in our trials, generally overestimating bud tolerance, particularly for BL and S. EB fruit showed more tolerance than predicted, H less.

UPDATE ON MICROPROPAGATION OF *PACHYSANDRA PROCUMBENS* MICHX.

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Pachysandra procumbens, the Allegheny spurge, is a native herbaceous perennial. Microcuttings were maintained in liquid MS medium in Magenta boxes with membrane rafts. A preliminary study looked at 1,2,5,5, and 10 mg BA/liter and 0 and 1 mg NAA/liter. Shootscultured with 5 or 10 mg BA/liter and no NAA produced significantly more axillary shoots. When shoots were cultured on media containing 5,10, 15, 20, or 25 mg BA/liter, trend analysis indicated significant linear and quadratic trends. Shoots proliferated similarly when cultured with 5 to 20 mg BA/liter. Non-fully expanded shoots proliferated significantly better compared to fully expanded shoots. After 6 weeks, microcuttings dipped in 500 or 1000 ppm IBA (50% ethanol) had produced significantly more roots of greater length compared to the control. Sixteen weeks after potting, rooted microcuttings potted in MetroMix 500, Promix BX, or a nursery mix (60% pine bark, 20% peat, 20% stone dust) had initiated significantly more shoots.

SURVIVAL OF *LISTERIA MONOCYTOGENES* ON LIGHTLY PROCESSED SPINACH

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The population of *Listeria monocytogenes* inoculated on lightly

processed spinach held at 5C or 10C remained constant for 9 days, whereas the populations of mesophilic aerobic flora, pseudomonas, and enterobacteria increased sharply. We studied the cause of the bacteriostatic activity of spinach leaves against *L. monocytogenes*. *Listeria monocytogenes* were inoculated at the concentration of 2×10^4 cells/ml in raw, autoclaved and vacuum filtration sterilized spinach macerates as well as in tryptic soy broth used as control. The concentrations after 24 hat 28C were 2×10^4 , 4×10^7 , and 4×10^8 cells/ml in respective spinach macerates and 4×10^8 cells/ml in tryptic soy broth. The anti-listeria activity was still present in spinach macerates sterilized by vacuum filtration but not in autoclaved macerates. In conclusion, the bacteriostatic activity against *L. monocytogenes* might be due to antimicrobial compounds present in spinach leaves or produced by the other microorganisms. Studies are being continued to identify the compounds involved in the anti-listeria activity.

THE EFFECTS OF HEAT STRESS ON RETENTION AND ABSCISSION OF LIMA BEAN REPRODUCTIVE STRUCTURES IN THE FIELD

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In an effort to increase lima bean yields in Delaware, the documentation of lima bean plant development and the comparison of Delaware and California lima bean production was conducted. Delaware lima bean yields have averaged 1905 kg-ha⁻¹ for the last 30 years. California averages 3923-4484 kg-ha⁻¹. Cultivar M-15 is used by both states for production. Plant population density, plant fresh weight, and final yield was greater in California than in Delaware. Although plant populations were the same in 1992, yields remained higher in California than in Delaware. High night temperatures have an adverse affect on lima bean yields. Minimum temperatures from both states were compared. Minimum temperatures from the California planting were greater than the minimum temperatures for the late planting in Delaware.

PLANT MORPHOLOGY AND FLOWERING PHENOLOGY OF LIMA BEANS GROWN AT HIGH TEMPERATURES IN THE GREENHOUSE

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Lima bean yields are lower in Delaware than in other lima-bean-producing states. One of the factors that contributes to the low production is the high temperatures that occur during production. Six commercial varieties of lima beans, both fordhook and baby lima bean types, were grown in a glass greenhouse at either 25C or 35C daytime temperatures to screen for heat tolerance. Plants grown at high temperature were typically shorter and more bushy than plants grown at 25C. Few, if any, buds, flowers, or early pods remained on plants at harvest if the plants were grown at 25C, while plants grown at 35C were still producing buds and flowers. Lima bean yields were generally reduced at 35C. The magnitude of the effect on yield ranged from 'F1072', which had a 100-fold decrease in yield, to 'Early Thorogreen', which demonstrated a slight increase in yield in response to increased temperatures.

THE EFFECTS OF MECHANICAL CONDITIONING ON FIELD PERFORMANCE OF TOMATO TRANSPLANTS

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Stretching is a problem in high-density transplant production. Mechanical conditioning provides good height control for many crops, but there may be adverse effects on field performance. Mechanical conditioning was applied to processing tomatoes (Ohio 8245) grown in #288-deep flats (=2000 plants/m²) using two methods, brushing and impendance. Brushing was applied by gently stroking the plant canopy with a Styrofoam planter flat 20 times back and forth every morning. The impeded plant canopy was compressed slightly by apiece of Plexiglas suspended overnight. The treatments were applied from canopy closure until transplanting to the field. At transplanting,

brushed plants were 31% (1993) and 12% (1994) shorter than control plants, and impeded plants were 25% (1993) and 24% (1994) shorter than control plants. In both years, the caliper of impeded transplants was significantly larger than that of both the control and brushed plants. There was also no reduction in dry weight and no noticeable difference in plant quality between treatments. The treatments did not affect the speed at which the plants recovered from transplant shock or the rate at which they grew in the field. Within 5 weeks after transplanting, there were no significant differences between treatments in biomass, leaf area estimates, stem caliper, flowering, early set, or field yield, despite differences in size at transplanting. Therefore, both brushing and impendance result in sturdy, high-quality transplants without adversely affecting establishment or yield.

EFFECTS OF AN ANTI-GIBBERELLIN GROWTH RETARDANT ON GROWTH RESPONSES OF POTATO NODAL STEM SEGMENTS CULTURED IN VITRO

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Foliage of non-induced 'Katahdin' potato plants was treated with BAS-111. Other plants were sprayed with GA₃ solution and placed in an inducing chamber. All treatments were repeated the following week. After final treatment, apical, sub-apical, medial, and basal nodal stem segments were taken from each plant, surface-sterilized, and placed on MS culture media. After 3 weeks in a darkened incubator, cultures were examined. Induced plants produced 5.5 times more tubers than did non-induced segments. BAS-111 applied to non-induced plants was associated with 63% reduction in rhizome length and 3.2-fold increase in tuber number. GA treatment to induce plants resulted in improved rhizome elongation, delayed and reduced tuberization when compared with control explants. Lower nodes produced more and larger tubers than did younger tissues. Results will be discussed in light of current literature.

SUPERFICIAL SCALD OF APPLES: A CHILLING INJURY THAT ALSO CAN BE INDUCED UNDER NON-CHILLING CONDITIONS?

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On Granny Smith apples, scald development exhibits characteristics that are typical of chilling injury. Yet, when 'Cortland' and 'Delicious' apples were placed in loosely closed polyethylene bags and kept continuously at 20C, scald-like injuries began to occur after 2 weeks. Lesions were predominantly expressed as lenticel spotting and as bronzing in the calyx cavity, which are not typical of superficial scald, but some typical scald lesions did develop on the shaded sides of the fruit. Lesion development decreased with later harvest of fruit. Fruit enclosed in poly bags accumulated high concentrations of α -farnesene and conjugated trienes in their peel. Whether or not scald should be considered to be a chilling injury will be examined in light of these contrasting results.

INDUCTION OF CHILLING-TOLERANCE IN CUCUMBER (*CUCUMIS SATIVUS* L.) SEEDLINGS BY ENDOGENOUS AND APPLIED ETHANOL

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Five-day-old etiolated cucumber (*Cucumis sativus* L. cv. Marketmore) seedlings held at 2C for 72 hours develop chilling injury resulting in desiccation and collapse of the hypocotyl tissues and eventual plant death. Hypoxia-induced accumulation of ethanol and acetaldehyde led to tolerance to subsequent chilling as evidenced by continued hypocotyl growth and freedom from injury. Arrest of volatile accumulation by applied bisulfite negated the development of hypoxia-induced chilling tolerance in seedlings. In seedlings held in normoxia, cold tolerance was induced by applied ethanol vapors, whereas acetaldehyde had a marginal effect, suggesting that hypoxia-

induced cold tolerance may arise from the accumulation and activity of ethanol. Cold tolerance was induced by exposure to gaseous n-propanol and n-butanol vapors and other volatile anesthetics, including chloroform and halothane, indicating that ethanol activity may stem in part from an anesthetic effect that causes disorder of membrane lipids. However, development of cold tolerance in ethanol-enriched tissues was time-dependent, suggesting an association with biosynthetic event(s). Ethanol did not change the fatty acid composition in cucumber hypocotyl membranes.

PHYSIOLOGICAL CHANGES ASSOCIATED WITH LEAF SENESCENCE IN EASTER LILIES

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Senescence of lower leaves of Easter lilies (*Lilium longiflorum* Thunb.) was previously shown to be delayed with application of the growth regulators, gibberellic acid (GA₃) and benzyladenine (BA). This study was done to determine the physiological effects of GA₃ and BA in relation to the delay of leaf senescence. Foliar application with 500 ppm BA or GA₃ delayed chlorosis and lowered respiration rate in Easter lily leaves. A combination of 500 ppm BA and 500 ppm GA₃ was more effective than the individual application of each. Gibberellic acid, BA, or their combination before cold storage resulted in delayed chlorosis and lowered respiration following removal from cold storage. Treatment with growth regulators after cold storage was less effective. Senescence of leaves was not associated with ethylene since ethylene production by leaves was undetectable by gas chromatograph. In addition, pulsing or continuous treatment with silver thiosulphate (STS), an inhibitor of ethylene synthesis, did not delay foliar chlorosis. Analysis of carbohydrate levels in Easter lily leaves treated with GA₃, BA, or their combination may contribute to the understanding of the physiological effects of these two growth regulators.

POPULATION VARIATION IN LOW-TEMPERATURE TOLERANCE OF *ACER SACCHARINUM*

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A single clone of *Acer saccharinum* was selected and propagated from each of 15 provenances across the plant native range. The clones were field grown in Carbondale, Ill., during the study period. Plants were sampled during Winter 1992-93 and 1993-94 and assayed for low-temperature tolerance. During both winters, plants exhibited greatest variation in tolerance around the November and April sampling dates. In midwinter, there was little variation observed and 13 of 15 clones were tolerant to at least -40C. The relationship among *Acer saccharinum* provenance and cold tolerance curves will be discussed.

CONTROL OF GREENHOUSE WHITEFLY (*TRIALEURODES VAPORARIORUM*) USING *DELPHASTUS PUSILLUS* AND AZATIN, A BOTANICAL INSECTICIDE

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Fifty greenhouse whitefly (*Trialeurodes*) were placed in each of 10, one-meter square nylon chiffon cages containing nine regal geraniums in 6-inch pots. After allowing the whitefly to reproduce for 2 weeks, the treatments tested were *Delphastus pusillus* alone, Azatin sprays alone, *D. pusillus* and Azatin together, and no control method. Two repetitions for each treatment were conducted. The Azatin, 14 oz/100 gallons, and a spreader/sticker were applied weekly with a mist sprayer. *Delphastus pusillus*, nine per cage, were released every 2 weeks. Sampling was conducted weekly by placing yellow sticky traps into each box for a 24-hour period, then counting the number of adults caught. All treatments gave statistically significant fewer whitefly than the cages with no control method. The cages with Azatin and/or *D. pusillus* were not statistically different from each other. Results indicate that *D. pusillus* can control whitefly as well as a growth regulator/botanical insecticide.

PERFORMANCE OF 'NELLIE WHITE' EASTER LILIES IN ROOT MEDIA CONTAINING COAL BOTTOM ASH

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Easter lilies (*Lilium longiflorum* Thunb. 'Nellie White') were forced in root media composed of 1 peat : 1 vermiculite (v/v) mixed with coal bottom ash (CBA) at rates of 0%, 25%, 50%, 75%, or 100% CBA. Lilies in all levels of CBA were equal in mean per plant flower bud numbers, fresh and dry weights, and numbers of yellow or brown lower stem leaves. Lilies in 100% CBA were significantly lower in mean stem length than plants in 0% or 50% CBA. Plants in 100% CBA required more frequent irrigation than plants in all other media. Media pH and solution electrical conductivity increased with increase in percent CBA. Analysis of leaf tissue showed no difference in nutrient levels between plants in 0% or 100% CBA.

DETERMINING THE EXTINCTION POINT OF SPINACH CULTIVARS FOR STORAGE IN LOW-OXYGEN ATMOSPHERE

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The extinction point (EP) of spinach cultivars was determined to identify the minimum O₂ concentration that can be used for modified-atmosphere storage of spinach. EP was based on respiratory quotient (RQ) and appearance quality during storage. Oxygen consumption decreased as O₂ concentration was lowered from 2.0% to 0.1%; whereas CO₂ production decreased as O₂ concentration was lowered from 2.0% to 0.5%, but not below 0.5%. The RQ was close to 1 in oxygen atmospheres of 2.0% to 0.4% and exceeded 1 at 0.2% or less. No alcohol production was noted at 0.2% or less O₂, but deterioration of leaves occurred at these low-O₂ atmospheres. Since the EP is slightly below 0.4% O₂, the concentration of O₂ should not be allowed to go below 0.5% for successful modified-atmosphere storage of spinach.

EFFECTS OF LOW OXYGEN CONCENTRATIONS ON GLYCOLYSIS IN LIGHTLY PROCESSED CARROTS

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Carrot (*Daucus carota* L.) shreds were stored under a continuous flow of 0.5% O₂ (balance N₂), 2% O₂ (balance N₂), or air for 9 days at 5C and 15C. The resulting changes in respiration and levels of the glycolytic intermediates were monitored. Low-oxygen atmosphere reduced respiration rate, but the RQ increased. The RQ was higher at 0.5% O₂ than at 2% O₂ atmosphere at both 5C and 15C. The most significant change in the levels of glycolytic intermediates was the accumulation of fructose 1,6-biphosphate. The level of fructose 1,6-biphosphate at 0.5% O₂ was about 2-fold greater than at 2% O₂ atmosphere at both 5C and 15C. The level of the other glycolytic intermediates at low-oxygen atmosphere was similar to that held in air atmosphere. These results suggest that phosphofructokinase activity in the tissue of carrots may increase under low-oxygen condition and it may be one of the controlling points in the glycolytic pathway affected by low oxygen concentrations.

GROWTH AND YIELD OF GREENHOUSE TOMATO WITH CONSTANT OR DIURNAL HEATING OF ROOT AND SHOOT

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What is the effect of constant compared to diurnal heating of the shoot and root on growth and yield of greenhouse tomato (*Lycopersicon esculentum* L.)? Seedlings were transplanted on 4 or 25 Mar. 1994 into troughs that were not heated or heated to 21C by buried tubing, either constantly or for 12 h during the day or the night. The greenhouses had

either 14/14C or 26/6C day/night minimum air temperatures. After 2 weeks, leaves of the 4 Mar. transplants weighed most with constant root heat and least with no heat. Roots weighed more with 14/14C than 26/6C air heat. With 14/14C air heat, only no root heat reduced leaf weight, whereas with 26/6C air heat, leaf weight was in the order: constant > day > night - no heat. After 2 weeks, leaves of the 25 Mar. transplants weighed least with no heat, and other treatments did not differ. Root heating affected yield. By 1 July, the number of fruit and the number and weight of marketable fruit produced from 4 Mar. transplants was in the order: constant heat > day > night > no heat. The 22/6C air heat increased marketable yield because fewer fruit were small, irregular, or had blossom-end rot. Root heat had no effect on yield of 25 Mar. transplants.

DAY AND NIGHT CONCENTRATIONS OF N AND C METABOLITES IN TOMATO SEEDLINGS GROWN IN SOLUTION WITH VARIOUS NITRATE CONCENTRATIONS

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Nitrate (NO₃) was supplied at 100, 200, 400, and 800 µM in nutrient solution to 3-week-old tomato seedlings grown hydroponically, and day and night concentrations of N and C metabolites were measured. Tissue [NO₃] at the end of the night was greater than at the end of the day, especially for leaves. Leaf tissue [NO₃] was about 350 µmol·g⁻¹ dry weight at night, 3 times as high as that during the day. Generally, root and stem tissue [NO₃] was similar and higher than that of leaves and increased as medium [NO₃] increased. The difference was greater at night than during the day. During the day, total free amino acid concentration was the greatest for roots and the least for stem. Generally, root tissue had higher total free sugar concentration than leaf and stem tissues during the day. Fructose concentration was lower at night than during the day for all parts of the plants, especially for roots. At night, NO₃ accumulated in plant tissues, especially in leaves, and was not incorporated into amino acids, perhaps due to the lack of energy and reductant.

ALLEVIATION OF CHILLING INJURY IN CUCUMBERS AND ZUCCHINI SQUASH BY METHYL JASMONATE

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Cucumbers (*Cucumis sativus* L.) and zucchini squash (*Cucurbita pepo* L.) were pressure-infiltrated (82.7 kPa for 3 min) with methyl jasmonate (MJ) in aqueous suspension and then stored at a chilling temperature of 5C. Control fruit were infiltrated with distilled water and handled in a similar manner. Treatment with MJ delayed the onset and reduced the severity of chilling injury symptoms in both cucumbers and zucchini squash. Analysis of polyamines in zucchini squash showed that putrescine increased with time in storage at 5C, while spermidine and spermine decreased during the same period. MJ treatment did not have an appreciable effect on putrescine, but the treated fruit maintained higher levels of spermidine and spermine than the control fruit throughout storage at 5C.

STRUCTURAL DETERMINANTS REQUIRED FOR IDENTIFICATION OF ACC TRANSPORT-RELATED MEMBRANE PROTEINS

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The ethylene precursor, 1-aminocyclopropane-1-carboxylic acid (ACC), is actively transported across the tonoplast of plant cells, impacting cellular compartmentation of ACC and ethylene biosynthesis. To identify potential photoaffinity probes for identifying ACC transport-related membrane proteins, the effects of over 70 ACC and other amino acid analogs on ACC uptake into isolated maize vacuoles were investigated. Only relatively nonpolar, neutral amino acid stereoisomers of L-configuration were strong inhibitors of ACC transport. Group additions, substitutions, or deletions at the carboxyl, (x-amino

and the Pro-(R) methylene, or hydrogen moieties essentially eliminated transport inhibition, whereas side-chain substitutions remained antagonistic. The kinetics of ACC and neutral L-amino acid analogs tested were competitive. The results indicate that the ACC transport system can be classified as a neutral L-amino acid carrier having a relatively high affinity for ACC and other nonpolar amino acids. The results also suggest that the carrier interacts with the carboxyl, alpha-amino, and Pro-(R) groups and the side chain of substrate amino acids. Based on these findings, potential photoaffinity probes of the ACC transport system have been identified.

FACTORS AFFECTING ELLAGIC ACID CONTENT IN SMALL FRUITS

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Ellagic acid, a putative anticarcinogenic compound, was detected in plants of mayhaw (*Crataegus* spp.), false strawberry (*Duchesnea indica*), strawberry (*Fragaria* spp.), black currant (*Ribes nigrum*), thornless blackberry (*Rubus* subgenus *Eubatus*), red raspberry (*Rubus* subgenus *Idaeobatus*), and cranberry (*Vaccinium macrocarpon*). Large differences in ellagic acid contents have been found among species and cultivars and also among tissues. Ellagic acid content in plant tissues is also affected by environmental factors and shows a seasonal variation in strawberry leaves. A decrease in ellagic acid content of leaves was associated with seasonal decreases in photoperiod and temperature from September to December. Ellagic acid content in the leaves of red raspberry infected with orange rust showed more than a 3-fold increase compared to healthy leaves.

EFFECT OF NAA OR ACCEL SPRAYS ON YIELD AND FRUIT SIZE OF 'EMPIRE' APPLE ON THREE ROOTSTOCKS

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'Empire' is a popular new apple with fruit growers in the northeastern United States, noted for producing small-sized fruit. To test the efficacy of chemical thinners and rootstocks for increasing fruit size of 'Empire', three-tree plots containing trees on M.7 EMLA, MM. 111, and seedling rootstocks were chemically thinned at petal fall with 10 ppm NAA or 85 ppm 6 BA, applied as Accel. Both NAA and Accel reduced fruit set. Trees on M.7 EMLA had higher set than trees on seedling. Yield was highest on M.7 EMLA and lowest on seedling. Fruit diameter after final set in July was increased by both chemical thinners and was greater for both clonal rootstocks than for seedling. Fruit on seedling trees were delayed in maturity relative to the two clonal rootstocks. Accel increased the number of fruit 70 mm or greater in diameter, while NAA increased the number of fruit in the 64- to 69-mm-diameter class. Analysis of covariance with crop load suggested that the increase in fruit size associated with Accel was a direct effect rather than a secondary effect from thinning.

COLD ACCLIMATION IN THE LEAVES OF SIBLING DECIDUOUS AND EVERGREEN PEACH: ALTERATIONS IN DEHYDRIN AND BARK STORAGE PROTEINS

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Seasonal pattern of cold tolerance and proteins were studied in the leaves of sibling deciduous and evergreen peach (*Prunus persica*). In contrast to deciduous peach that undergoes endodormancy in fall, evergreen peach does not (leaves are retained and shoot tips elongate under favorable conditions) (Arora et al., Plant Physiol. 99:1562-1568). Cold tolerance (LT₅₀) was assessed using electrolyte leakage method. Proteins were separated by SDS-PAGE. Electroblobs were probed with anti-dehydrin (Dr. T. Close) and anti-19 kD, peach bark storage protein (BSP) antibodies. LT₅₀ of leaves successively increased from about -7C (18 Aug.) to -15C and -11.5C (23 Oct.) in deciduous and evergreen genotypes, respectively. The most apparent

change in the protein profiles was the accumulation of a 60-kD protein during cold acclimation in the leaves of deciduous trees; however, it did not change significantly in evergreen peach. Immunoblots indicate that 60-kD protein is a dehydrin protein. PAGE and immunoblots indicated that 19-kD BSP disappeared progressively during summer through fall in the leaves of deciduous peach, but accumulated to large amounts in bark tissues. Similar inverse relationship for its accumulation in leaf vs. bark tissue was not evident in evergreen peach. Results indicate that BSP expression may be regulated by altered source/sink relationship.

ROLE OF CALCIUM IN REDUCING POSTHARVEST CELL WALL DEGRADATION IN 'GOLDEN DELICIOUS' APPLE FRUIT

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Calcium is an important constituent of the cell wall and plays roles in maintaining firmness of fruit and reducing postharvest decay. The modification of the cell wall is believed to be influenced by calcium that interacts with acidic pectic polymers to form cross-bridges. Infiltrating apples with CaCl₂ has been suggested as an effective postharvest treatment for increasing the calcium content. Three different methodologies were used to analyze the effects of calcium on the cell walls: 1) nickel staining of polygalacturonate on free-hand sections, 2) cationic gold labeling of anionic binding sites in the cell walls, and 3) analytical detection of calcium ions (40Ca, 44Ca) using a secondary ion mass spectrometry. The combination of these methods allowed us to directly visualize the cellular features associated with the infiltration of calcium. Treatment resulted in significant enrichment in the cell wall of the pericarp, transformed the acidic pectins in calcium pectates, and resulted in new calcium cross-bridges. Evidence now suggests that exogenously applied calcium affects the cell wall by enhancing its strength and reinforcing adhesion between neighbor cells; therefore, calcium infiltration delays fruit degradation.

LIPIDS IN SUBEPIDERMAL CORTICAL TISSUE OF 'GOLDEN DELICIOUS' APPLE FRUIT

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Altered metabolism of membrane lipids has been proposed as a mechanism for the beneficial effects of postharvest calcium treatment on apple quality. A previous study showed that after transfer of apples stored 6 months at 0C to 20C, calcium-treated fruit exhibited slower loss of galactolipid and altered levels of sterol conjugates. The present study of lipids in "control" fruit was conducted as a prelude to further in-depth analyses of the effects of postharvest calcium and heat treatments on lipid metabolism in apples during and after cold storage. Neutral lipid, glycolipid (GL), and phospholipid (PL) fractions were obtained by column chromatography followed by TLC separation of GL and PL classes. The major GL were steryl glycosides (SG), acylated steryl glycosides (ASG), cerebrosides (CB), and mono- and digalactosyl diacylglycerols. Phosphatidylcholine (PC) > P-ethanolamine (PE) > P-inositol (PI) were the major PL. The fatty acids of PC and PE were quite similar, whereas those of PI were more saturated. CB included only 2-hydroxy fatty acids. Among the steryl lipids, free sterols > SG > ASG, with beta-sitosterol >90% of the total sterol in each.

STRAWBERRY VARIETY YIELDS AND SUSCEPTIBILITY TO TARNISHED PLANT BUG AND STRAWBERRY BUD WEEVIL

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Twelve strawberry varieties established in matted row plots in 1993 were treated with insecticides for tarnished plant bug and strawberry bud weevil or left untreated during 1994. Honeoye and Cavendish had the highest yields of marketable fruit. Oka, Jewel,

Chambly, and Kent also had relatively high yields. Lateglow, Blomidon, Seneca, NY 1424, Settler, and Governor Simcoe had lower yields than the other varieties. Tarnished plant bug populations were very small during the 1994 season and the injury levels observed were relatively low. Feeding pressure on the plants may have been too low for all differences in susceptibility between varieties to be expressed. Governor Simcoe, Cavendish, and Oka had the lowest injury levels. Kent, Lateglow, and Seneca had the highest levels of injury. Insecticide sprays significantly reduced injury for all varieties except Cavendish and Governor Simcoe. The number of flower buds killed by strawberry bud weevil differed very little between varieties and spray treatments. No obvious differences in susceptibility to this injury were observed in this trial.

THE IMPORTANCE OF α - AND β -GALACTOSIDASE IN TOMATO FRUIT RIPENING

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Antisense technology has shown that neither polygalacturonase nor pectin methyltransferase alone are responsible for tomato fruit softening, leading to the likelihood that other enzymes or factors are important. Our laboratory recently found that α - and β -galactosidase from avocado fruit solubilized tomato fruit pectin in vitro. Previously, Pressey (Plant Physiol. 1983,71:132) found that the activity of one of three α -galactosidase isozymes from tomato fruit increased during ripening and was capable of degrading cell wall galactan, suggesting a role for the enzyme in fruit softening. Increased β -galactosidase activity was observed in a number of other fruit during ripening. In the present study, NaCl extraction of tomato pericarp yielded relatively high levels of α - and β -galactosidase activity. At least two isozymes of each were resolved during Mono-Q HPLC α -Galactosidase was further purified by additional Mono Q and Superose 12 gel filtration HPLC. Gel filtration and SDS-PAGE yielded an apparent molecular weight of 44 kD. The partially pure α -galactosidase had a specific activity of 294 μ mol product/min per mg protein, a K_m of 317 μ M, a pI of 5.0, and a pH optimum of 5.5. Activity was inhibited 67% by α -D-galactose. Preliminary results show that β -galactosidase can also be purified by the same techniques. Following further purification, the isozymes will be sequenced and cloned. A second approach being used in an attempt to identify cDNA clones for the α - and β -galactosidase genes from tomato fruit involves using heterologous cDNA clones from guar (Overbeek et al., 1989; Plant Molecular Biology 13:541-550) and carnation (Raghothama et al., 1991; Plant Molecular Biology 17:61-71), respectively, to screen a ripening tomato fruit cDNA library. Basic molecularbiological techniques will be used to elucidate the role of these enzymes in tomato fruit ripening.

INFLUENCE OF PLANTING DATE, PLANTING SYSTEM, PLANT TYPE, AND MULCH COLOR ON 'ALLSTAR' AND 'CHANDLER' PRODUCTIVITY IN STRAWBERRY PLASTICULTURE

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High-density, annual, strawberry production systems ("plasticulture") have shown high productivity under New Jersey conditions; however, cultural practice and variety research is needed to increase profitability. The system includes raised beds, plastic mulch, trickle irrigation, and double-row 12 x 12-inch plant spacing. Polypropylene floating rowcovers were applied in December and removed in early April when flowers were visible under the cover. Treatments included comparisons of plugs and dormant crowns of the cultivars Chandler and Allstar, planted at multiple planting dates, on white or plastic mulch, in "matted-row" (single row at 18-inch spacing; peg runners through plastic) or high-density production systems. The plug plants were superior to dormant crowns. Black plastic was best all planting dates with plugs; 'Allstar' performed best on black on the early planting dates, while 'Chandler' preferred the white for the early planting dates. Both 'Allstar' and 'Chandler' had commercially prof-

itable yield, fruit weight, and quality. "Matted-row" system on plastic is high-yielding but labor-intensive. Late-summer plugs on black plastic is best overall.

PERCEPTION AND ADOPTION OF IPM TACTICS BY LANDSCAPE CONTRACTORS IN CENTRAL NEW JERSEY

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The landscape/nursery/turfgrass industry is the largest agricultural industry in New Jersey, as well as one of the highest users of pesticides. In the lawn-care industry alone, more than 906,000 lbs of active ingredient of pesticides was used in 1990. Landscape Integrated Pest Management (LIPM) tactics have been commercially proven to reduce pesticide usage; however, adoption of LIPM has been slow. In 1993-94, a survey of 425 landscape contractors, arborists, groundskeepers, nurserymen, and turfgrass professionals was taken to determine attitudes toward adoption of LIPM tactics. Business changes, marketing, customer perceptions, educational needs, and attitudes toward alternative control tactics were assessed. Results show that the majority of landscapers are interested in LIPM for personal reasons, to reduce their own contact with pesticides. Contractors favor pesticide products that are cost effective and proven as opposed to environmentally "safe." Concerns inhibiting LIPM adoption include potential customer dissatisfaction, recovering monitoring costs, and inadequate control. Challenges lie ahead in pest identification and control education, marketing programs, delays in profits, and writing bids.

NUCLEAR MAGNETIC RESONANCE IMAGING OF FUNGAL INFECTIONS IN STRAWBERRY FRUIT

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We report the use of nuclear magnetic resonance (NMR) imaging to detect differences in invasion and colonization of fruit by pathogens (*Botrytis cinerea*, *Colletotrichum acutatum*, and *Phytophthora cactorum*), and bruise wounds are sharply distinguishable from healthy fruit tissue by their T1 times. Digitized images from T1 images clearly show two or more zones of pathogen activity in fruit tissue. The innermost zone corresponds to the area of greatest invasive activity at the leading margin of the infection. A second zone corresponds to the area of tissue that has been killed and is being degraded by the pathogen. Sometimes, a third zone is present at the outer border of the lesion and this correspond to where aerial sporulation may occur. Images of bruises, however, are uniform with no apparent gradations in T1 characteristics. Detection of fruit deterioration and decay is important in understanding and controlling postharvest loss of fruit crops. The nondestructive nature of MRI provides a means to quantify the process of decay development and control measures applied to fruits.

EVALUATION OF ORGANIC RESIDUES FOR THE PRODUCTION OF FLOWERING POT PLANTS

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During Winter 1994, seedlings of *Impatiens wallerana* 'Accent Coral' and *Pelargonium x hortorum* 'Orbit Hot Pink' were grown in commercial substrate (Pro-Mix BX) or in one of the 24 substrates composed of six organic residues (composted water-treated sludge, forestry compost, fresh or composted used peat extracted by a biofilter during treatment of municipal water, and fresh or composted paper sludge). These residues were incorporated with peatmoss and perlite at 5%, 10%, 25%, and 40% per volume to obtain the 24 substrate combinations. Plants were watered and fertilized by flooding of ebb-and-flow benches. Growth measurements (growth index, top and root dry weight, number of flowers and buds, visual quality) varied considerably depending on the percentage of residue incorporated into the substrates. Moreover, substrates containing 40% of organic residues are not recommended for production of impatiens or geraniums.

FERTILIZATION VALUE OF THREE ORGANIC RESIDUES AS AN AMENDMENT TO MEDIA USED TO PRODUCE CONTAINER-GROWN WOODY ORNAMENTAL PLANTS

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The agricultural valorization of waste, rich in organic matter and minerals, is one of the best economical and ecological methods of disposal. This study was carried out to evaluate the release of mineral elements restrained in fresh bio-filters, composted sewage sludges, and composted de-inking sludges, and their effects on growth of *Physocarpus opulifolius* 'Nanus' produced in containers. The physical and chemical analysis of the organic residues proved that the fertilization value of composted sewage sludges was greater than the other residues. Moreover, the granular characteristics of fresh bio-filters and composted de-inking sludges were finer than composted sewage sludges. Each organic residue was combined, in proportion of 10%, with peatmoss, composted conifer bark, and fine crushed gravel. The regular leaching of container medium showed that the composted sewage sludges release a higher quantity of major mineral elements. *Physocarpus opulifolius* 'Nanus' plants were larger than those plants grown in the control substrate (without residue). The results obtained in media containing fresh bio-filters or composted de-inking sludges were similar to those obtained in the control substrate.

GENETIC RELATIONSHIP AMONG STRAINS OF *XYLELLA FASTIDIOSA* BASED ON RAPD-PCR DATA

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Xylella fastidiosa is a fastidious gram-negative, xylem-limited, leafhopper-transmitted bacterium that has proven to be the casual agent of many economically important diseases, including Pierce's disease of grapevine and citrus variegated chlorosis. Genetic relationships among 11 *Xylella fastidiosa* strains isolated from mulberry, almond, ragweed, grape, plum, elm, and citrus were determined using random amplified polymorphic DNA (RAPD). Twenty-two 10 base primers amplified a total of 77 discrete polymorphic bands. Phenetic analysis based on a similarity matrix corresponded well with previous reports on *X. fastidiosa* RFLP-based similarity relationships, indicating that RAPD-PCR amplification products can be used as a reliable indicator of genetic distance in *X. fastidiosa*. Cladistic analysis suggests the existence of five groups of *X. fastidiosa*: the citrus group, the plum-elm group, the grape-ragweed group, the almond group, and the mulberry group.

USE OF ¹H-NMR TO DETERMINE GRAPE BUD WATER STATUS DURING THE PHOTOPERIODIC INDUCTION OF DORMANCY

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Changes in tissue water content have been correlated, with varying success, with changes in freezing tolerance and dormancy in woody perennials. Recent studies indicate that changes in the state of water are more strongly correlated with dormancy than are changes in bulk water content. In this study, traditional destructive methods of monitoring tissue water content and dormancy were compared with measurements using nondestructive in situ proton nuclear magnetic resonance ¹H NMR to determine plant water status. These studies were designed to determine whether changes in bud water status are correlated with dormancy and can be used as a reliable indicator of the onset of dormancy. Two-year-old *Vitis riparia* plants were subjected to short-day (SD, 8 h daylight) or long-day (LD, 15 h daylight), dormancy-inductive or noninductive treatments, respectively. Bud water was monitored at 2, 4, and 6 weeks of photoperiod treatments. SD treatments promoted a rapid onset in bud dormancy. Water content was not different in SD or LD treatments after 2 weeks. However, it did decrease over 6 weeks in both treatments, but SD treatments promoted a more rapid decrease in water content. The nondestructive ¹H NMR methods give comparable measures of water content and provide a

measure of bud water status. There were shorter T1 relaxation times in the 2-, 4-, and 6-week SD treatments. The SD treatment T2 relaxation times were shorter in the 4- and 6-week SD treatments only. Changes in the T1 and T2 relaxation times indicated changes in bud water status are correlated with the onset of dormancy.

THE INFLUENCE OF C : P RATIO ON THE BIOLOGICAL ACTIVITY OF MSW

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Optimum conditions for composting encourage and maintain the growth of microorganisms. Aerobic conditions must be maintained along with a 30 C : 1 N ratio and appropriate moisture levels. Our research found that P along with C and N are primary nutrients required by the microorganisms involved in composting. Phosphorus is a very important component of ATP and ADP, which drive most biochemical processes and are therefore necessary to all energy-driven processes. Results of this experiment show that MSW treatments with a minimum of 120 C : 1 P result in significantly higher temperatures during the composting process; lower final C : N ratios; greater volume reduction; and more available N in the final product. Emphasis of ongoing research is to determine appropriate C : P levels.

PERICLINAL CHIMERAS COMPOSED OF *LYCOPERSICON PERUVIANUM* AND *L. ESCULENTUM* DEMONSTRATE THAT S-LOCUS ASSOCIATED PROTEINS AND SELF-INCOMPATIBILITY CAN BE UNCOUPLED

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Lycopersicon peruvianum is a wild species of tomato that exhibits gametophytic self-incompatibility (S), wherein the SI response is controlled by the genotype of the pollen. Cultivated tomato (*L. esculentum*) is a self-compatible species. Assisted by phenotypic markers, periclinal graft chimeras between these two species have

been obtained. Fruit set analysis following breeding demonstrated that the available five chimeras (PPE, PEE, PEP, EPP, and EEP) are able to accept pollen from *L. peruvianum*, suggesting that there is a failure of the SI response. SI response is known to be dependent on S-locus associated proteins. These proteins are present in the style, which is mainly derived from the L1 and L2 layers of meristem. RNA analysis of the style tissue using a cloned S-locus cDNA as a probe showed that, except for EEP, all chimeras expressed the S-allele. This was also confirmed by SDS-PAGE analysis of stylar proteins that were present in variable amounts depending on the periclinal combination. Thus, the breakdown of SI is not associated with the lack of expression of the S-locus. Further work is being conducted to understand the nature of this breakdown.

INITIAL GENETIC LINKAGE MAP AND EXPERIMENTS TOWARD TAGGING GENES WHICH CONTROL CHILLING REQUIREMENT IN BLUEBERRY (*VACCINIUM* SPP.)

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Efforts are underway to develop genetic linkage maps for two interspecific blueberry populations (*Vaccinium darrowi* × *V. elliotii* and *V. caesariense*-derived populations). To date, 72 RAPD markers have been mapped, and another 200 markers have been identified as suitable for mapping in the *V. darrowi* × *V. elliotii*-derived population. Inheritance of 40 RAPD markers has been followed, and additional 40 RAPD markers have been identified as suitable for mapping in the *V. darrowi* × *V. caesariense* population. These two populations are comprised of individual plants that should have a wide range of chilling requirements. At a later date, plants will be classified according to their chilling requirements to identify RAPD markers that cosegregate with chilling requirements. Presently, a bulked-segregant analysis is being performed on a tetraploid breeding population (primarily *V. corymbosum*) to identify RAPD markers linked to chilling requirement genes.