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SURVEY OF PLANT PIGMENTS: MOLECULAR AND ENVIRONMENTAL DETERMINANTS OF PLANT COLORS

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It is difficult to estimate the importance of plant pigments in plant biology. Chlorophylls are the most important pigments, as they are required for photosynthesis. Carotenoids are also necessary for their functions in photosynthesis. Other plant pigments such as flavonoids play a crucial role in the interaction between plants and animals as visual signals for pollination and seed scattering. Studies related to plant pigmentation are one of the oldest areas of work in plant science. The first publication about carotenoids appeared in the early nineteenth century, and the term "chlorophyll" was first used in 1818 (Davies, 2004). Since then, the biochemical structure of plant pigments has been revealed, as have the biosynthetic pathways for the major pigments that provide a useful variety of colors to blossoms and other plant organs. There is widespread interest in the application of molecular methods to improve our knowledge of gene regulation mechanisms and changes in plant pigment content. Genetic modification has been used to alter pigment production in transgenic plants. This review focuses on flower pigmentation, its biochemistry and biology. It presents a general overview of the major plant pigment groups as well as rarer plant dyes and their diversity and function in generating the range of colors observed in plants.

Key words: Flower and fruit colors, co-pigmentation, plant dyes, pigment groups.

ANTIBACTERIAL ACTIVITY OF PLUMBAGIN AND ROOT EXTRACTS OF PLUMBAGO ZEYLANICA L.

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This work assesses the antibacterial activity of plumbagin (5-hydroxy-2-methylnaphthalene-1,4-dione) and of methanol, chloroform and aqueous extracts of *Plumbago zeylanica* L. root against various pathogenic bacteria, and the minimum inhibitory concentrations (MICs). Plumbagin and chloroform extracts of *Plumbago zeylanica* L. root showed antibacterial activity against *Escherichia coli*, *Salmonella typhi* and *Staphylococcus aureus*. Inhibition against *Klebsiella pneumoniae*, *Serratia marcescens* and *Bacillus subtilis* was moderate, and lower against *Proteus vulgaris* and *Pseudomonas aeruginosa*. The methanolic extract exhibited moderate activity and the aqueous extract weak activity against the bacterial strains as assessed by disc diffusion assays. The bioactive compound plumbagin and extract of *Plumbago zeylanica* root show a wide spectrum of antibacterial activity. The compound shows promise as a new drug for various bacterial infectious diseases.

Key words: *Plumbago zeylanica*, Plumbaginaceae, antimicrobial screening, plumbagin, antibiotics, minimum inhibitory concentration (MIC).

ANTIOXIDANT RESPONSE SYSTEM AND CHLOROPHYLL FLUORESCENCE IN CHROMIUM (VI)- TREATED ZEA MAYS L. SEEDLINGS

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The effects of different concentrations of Cr(VI) (1 μ M, 10 μ M, 100 μ M) applied for 7, 14 or 21 days on initiation of high lipid peroxidation level (POL) and consequent changes in the enzymatic-antioxidant protective system and minimization of photosystem II (PSII) activity were studied in maize seedlings. Chromium(VI) caused an increase in the electrical conductivity of the cell membrane, and malondialdehyde (MDA) content (a peroxidation product) reflected peroxidation of membrane lipids leading to the loss of the membrane's selective permeability. It also induced distinct and significant changes in antioxidant enzyme activity. Versus the control, superoxide dismutase (SOD, EC 1.15.1.1.), catalase (CAT, EC 1.11.1.6.) and peroxidase (POD, EC 1.11.1.11.) activity in maize seedling roots and leaves was progressively enhanced by the different Cr(VI) doses and stress periods, except for decreases in SOD and POD activity in leaves exposed to 100 μ M Cr(VI) for 21 days. The different Cr(VI) concentrations changed chlorophyll (chl) content differently. The 10 μ M and 100 μ M doses of Cr(VI) decreased the chl *a/b* ratio and quenched the chl *a* fluorescence emission spectra. These effects reflect disturbance of the structure, composition and function of the photosynthetic apparatus as well as PSII activity.

Key words: Chromium(VI), lipid peroxidation, antioxidant protective system, chlorophyll fluorescence, maize.

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SEED GERMINATION OF SEVERAL INVASIVE SPECIES POTENTIALLY USEFUL FOR BIOMASS PRODUCTION OR REVEGETATION PURPOSES UNDER SEMIARID CONDITIONS

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The introduction of several plant species to areas beyond their natural distribution has been a global phenomenon that poses critical problems and challenges for the conservation and management of many agricultural and natural ecosystems. Shrub medick (*Medicago arborea* L.), Spanish broom (*Spartium junceum* L.) and chaste tree (*Vitex agnus castus* L.) are three of the most important native shrubs in arid and semiarid Mediterranean regions, being noxious invasive species for some areas but in some cases remarkably useful for several purposes. An understanding of their seed germination and seedling emergence should prove useful for their management. Laboratory and greenhouse experiments were done to examine the effects of high temperature on seed germination and seedling emergence. Dry heat benefitted (or at high temperatures damaged) seed germination and emergence. This implies potential effects of fire on the dynamics of populations of *M. arborea*, *S. junceum* and *V. agnus castus*, a factor which should be taken into account as fire is a frequent component of Mediterranean-type ecosystems.

Key words: *Medicago arborea, Spartium junceum, Vitex agnus castus*, temperature, invasive weeds, biomass, Vonitsa.

TOCOCHROMANOLS, PLASTOQUINONE AND POLYPRENOLS IN SELECTED PLANT SPECIES FROM CHILEAN PATAGONIA

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A collection of 14 species of Patagonian plants was evaluated for tocopherol content and composition, plastochromanol, plastoquinone content and polyprenol composition. Total tocopherols varied from 35.77 µg/g in *Mutisia decurrens* to 798 µg/g in *Philesia magellanica*. In most cases to copherol composition was dominated by α -to copherol, which accounted for more than 90% of total tocopherols. Of all the investigated species only Misodendrum *punctulatum* showed higher content of γ - than α -tocopherol, which is unusual for mature leaves. Plastochromanol, a homologue of γ -tocotrienol, was found in leaves of 10 of the 14 examined species, and was highest in Araucaria araucana leaves (132 µg/g). Total content of PO-9 (oxidized and reduced) was highest in Fuchsia magellanica (774.3 µg/g), Philesia magellanica (791 µg/g), Misodendrum linearifolium (569 µg/g) and Amomyrtus luma (518.5 $\mu g/g$). Analysis of polyprenol content in the leaves of investigated plant species revealed detectable amounts (>10 µg/g d.w.) of polyprenyl esters in six of them. Accumulation of free polyprenols was detected only in Chusquea quila leaves. Selected organs of Philesia *magellanica* and *Fuchsia magellanica* were further quantitatively analyzed for tocochromanol and polyprenol content. With the methods applied, different patterns of the analyzed compounds were identified in all the samples studied. Our results reveal some trends that may be of taxonomic interest. Some of these species can serve as a rich source of such bioactive compounds as tocochromanols or polyprenols.

Key words: Chromatography, HPLC, Patagonian plants, secondary metabolites, tocopherols, plastochromanol, plastoquinone, polyprenols.

MICROMORPHOLOGICAL STUDIES OF LALLEMANTIA L. (LAMIACEAE) SPECIES GROWING IN TURKEY

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Micromorphological features related to the pollen, nutlets and trichomes of Lallemantia species growing in Turkey were investigated mainly by scanning electron microscopy. Lallemantia pollen shares morphological features with subfamily Nepetoideae (tribe Mentheae, subtribe Nepetinae) pollen. However, the fine details are characteristic enough to differentiate the pollen between species. The exine is microreticulate in L. peltata and L. iberica, and reticulate-foveolate in L. canescens. Similarly, nutlet features are similar in general, but there are striking differences in surface details between species. Nutlets are black and oblong-triangular with V-shaped areoles. The surface is verrucate in L. iberica and L. canescens, and verrucate-rugulate in L. peltata. The warts are regular and separated in L. *peltata*, irregular and separated in *L. iberica*, and irregular and separated or sometimes associated in 2 to 4 groups in L. canescens. Two types of trichome, capitate and acicular, are present on the stems, leaves, calyx and bracts. The results suggest that although the distribution and micromorphology of trichomes has no taxonomic value, some pollen and nutlet micromorphological characters have the potential to serve as phylogenetic markers at the species level in the genus Lallemantia. However, pollen characteristics show no correlation with the nutlet characteristics.

Key words: Lamiaceae, *Lallemantia*, Mentheae, micromorphology, pollen, nutlet, trichomes, Nepetinae, taxonomy.

IN VIVO POLYEMBRYONY INDUCTION IN SPECIES OF CAPSICUM

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Haploid plants after doubling the chromosomes can give rise to diploid homozygote lines, which can be used as DH lines in breeding new varieties or as initial plant material in creating F1 hybrids. This work studied natural polyembryony and the effect of growth regulators on induction of polyembryonic seeds and haploid embryogenesis in five species of the genus Capsicum. Water solutions of the following growth regulators were used: 2,4-D (2,4dichlorophenoxyacetic acid) and BNOA (beta-naphthoxyacetic acid) at 0.001% used separately or combined with BAP (benzylamino-purine). Twin seed frequency was highest in C. chinense and lowest in C. baccatum var. pendulum. In C. annuum the share of twin embryos was highest in the 'Corno di toro' variety; 2,4-D clearly increased the number of twin plants only in 'Corno di toro' seeds. Treatment with combinations of 2,4-D or BNOA with BAP increased the frequency of polyembryonic seeds in the 'ATZ1' line. In C. frutescens the frequency of polyembryony increased following application of BNOA with BAP. Of all the seeds tested, seven haploid plants were obtained, representing C. frutescens, C. chacoense and C. baccatum var. pendulum. The differences in the frequency of polyembryony in the studied genotypes points to genetic control of this phenomenon. The presence of monoploid plants definitely depends on the genotype, as a high frequency of polyembryony is not always accompanied by a high share of haploid plants. For most genotypes evaluated, the effect of growth regulators was disadvantageous, resulting in a considerable decrease in the share of twin plants among germinated seeds.

Key words: *Capsicum* spp., growth regulators, haploid, pepper, polyembryony.

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MORPHOGENETIC RESPONSE TO PLANT GROWTH REGULATORS IN TRANSFORMED AND UNTRANSFORMED HYPERICUM PERFORATUM L. CLONES

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This study examined the effects of different exogenous auxins and cytokinins at 0.1-5.0 mg·l⁻¹ concentration on shoot cuttings of two *H. perforatum* clones transformed with a wild agropine strain of *A. rhizogenes* and one untransformed clone. Their sensitivity to the auxins varied and showed concentration-dependent behavior, and the response to auxins differed between the transgenic clones. The number of cuttings of transgenic clones capable of root formation, and the onset of rooting on most of the media with auxins lagged behind the control. The number of differentiated shoots of the transgenic clones on hormone-free medium was two to three times higher than that of the untransformed control. Regenerated shoots of the transgenic and control clones differed in their ability to form shoots on media supplemented with cytokinins. Increased cytokinins led to differentiation of shorter shoots with fewer leaf pairs. Because gene expression studies have shown integration of *rolABC* genes, their possible impact on the type of morphogenetic response is discussed.

Key words: Hairy root-regenerants, plant growth regulators, *rol* genes, shoot cuttings, St. John's wort.

GENETIC DIVERSITY OF MELICA TRANSSILVANICA SCHUR (POACEAE) AT ITS NORTHERN RANGE LIMIT

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Geographically marginal populations are expected to have low genetic variability, which potentially can affect their viability. In Poland Melica transsilvanica Schur reaches the northern limit of its continuous geographical range. Genetic diversity and population genetic structure were analyzed in 15 of its marginal and more central populations using AFLPs. Overall, genetic diversity parameters did not differ significantly, and comparable patterns of genetic variation were found in central and marginal populations. All AFLP phenotypes were unique to particular populations. Unique alleles were fixed in some central and some marginal populations. The percentage of polymorphic loci varied from 1.30 to 5.19 (3.24 average) in central populations and from 0.43 to 5.63 (2.36 average) in marginal ones. Hierarchical analyses of molecular variance (AMOVA) for each species/region combination revealed highly significant differentiation between populations and showed similar partitioning of molecular variance in marginal and central populations of *M. transsilvanica* (diversity between populations: 93.24% and 93.18%, p < 0.001, respectively). The scattered distribution of suitable species habitats and the predominant selfing breeding system of the species strengthen the effect of selection pressure on fixation of unique loci in individual populations. Marginal populations of *M. transsilvanica* with unique alleles considerably expand the genetic variation of the species and are therefore valuable for conservation of

genetic diversity.

Key words: AFLP, genetic diversity, marginal populations, *Melica transsilvanica*, range limit.

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ORGANOGENESIS AND SOMATIC EMBRYOGENESIS INDUCED IN PETAL CULTURES OF SEDUM SPECIES

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Plant regeneration was studied in petal cultures of three *Sedum* species: *S. aizoon, S. spectabile* and *S. gracile*. The course of morphogenesis was examined by light and electron microscopy. Histological examination revealed that morphogenesis took place as direct organogenesis, indirect organogenesis or somatic embryogenesis, depending on the species and the concentrations of growth regulators in the medium. Initial petals and explants from cultures were studied to determine the origin of organogenesis. Petal histology showed that all cells at the time of culture initiation were differentiated. Epidermal and parenchymatous cells were highly vacuolated and the parenchyma contained chloroplasts with starch grains. TEM revealed that cell dedifferentiation occurred in culture under the influence of BAP and IBA. In petal culture the first cell division started subepidermally on day 2 of culture initiation, as confirmed by histology and SEM. Direct formation of adventitious buds in petals was observed in meristematic cells dedifferentiated from the epidermis and parenchyma. In indirect organogenesis, callus tissue resulted from division of dedifferentiated parenchyma cells.

Key words: *Sedum*, adventitious buds, somatic embryos, callus, petal culture, light and electron microscopy.

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MORPHOLOGICAL VARIABILITY OF SEEDS AND FRUITS OF RUSCUS HYPOGLOSSUM IN CROATIA

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The paper reports a morphological study of fruits and seeds of *Ruscus hypoglossum* L., a species interesting as an ornamental plant. Initially, the length, width and weight of fruits as well as seeds were measured in 14 natural populations growing in Croatia. The length/width ratios of fruits and seeds were calculated, as well as the fruit/seed ratios of length, width and weight. Interannual differences in the analyzed traits over three years were compared in two populations. The most promising populations for further selection for ornamental use were the Bilogora population with the largest fruits (length 12.30 mm, width 11.56 mm, weight 0.75 g) and the Strahinšèica population with the roundest fruits (length/width 1.04). The average number of seeds per fruit for the populations was 1.40. Variability between the three years was minor for fruit traits but significant for seed traits.

Key words: Ornamental plant, Ruscus hypoglossum, fruit and seed traits, natural populations, interannual variability.

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MICROPROPAGATION OF CARLINA ACAULIS L.

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An efficient shoot propagation system for *Carlina acaulis* was developed in this study. The experimental material consisted of shoot tips and fragments of hypocotyls excised from 10day-old seedlings. The explants were transferred to proliferation medium supplemented with different types of cytokinins: 6-benzylaminopurine (BA, 4.4 or 13.3 μ M), kinetin (Kn, 4.7 or 13.9 μ M) and zeatin (Zea, 4.6 or 13.7 μ M) in combination with naphthaleneacetic acid (0.54 μ M NAA). The morphogenetic response was best in culture on medium supplemented with 13.3 μ M BA, and shoot organogenesis frequency was highest for shoot tips (100%). On average, 7.5 shoots were induced per explant of the initial material, and the multiplication rate in five subsequent subcultures was 6.1. Shoot length was lower in culture with BA in the medium than with Kn or Zea. Plantlets rooted with 60% frequency in vitro on full-strength MS medium and with 55.3% frequency ex vitro. Reduction of the mineral salt concentration (1/2MS) stimulated rhizogenesis. Addition of auxins stimulated both the frequency and number of roots per shoot, but only in combination with full-strength MS medium. Regenerated plants were able to flower and gave viable seeds.

Key words: Carlina, shoot tip, hypocotyl, benzylaminopurine, ex-vitro rooting.

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CALCIUM OXALATE CRYSTALS IN SOME SPECIES OF THE TRIBE INULEAE (ASTERACEAE)

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In this study, calcium oxalate (CaOx) crystals were investigated and their morphology and distribution determined by light microscopy in tissues and organs of *Inula graveolens* (L.) Desf., *Pulicaria dysenterica* (L.) Bernh., *Filago eriocephala* Guss., *Logfia arvensis* (L.) Holup and *Logfia gallica* (L.) Coss. & Germ., which belong to tribe Inuleae (Asteraceae). CaOx crystals were identified in cleared organs and tissues by a histochemical technique using silver nitrate and rubeanic acid. Druses were observed in stem pith cells, leaf mesophyll cells and style cells of *I. graveolens*. In anther tissues, crystals were found in petal and filament cells of *I. graveolens*. Druse crystals were present in the filament and style cells of *P. dysenterica*; styloids were found in the endothecial tissues of anthers, and prismatic crystals in the ovary cells of this species. No crystals were found in petal, stem and leaf tissues of *P. dysenterica*. *F. eriocephala* and *L. arvensis*, and *L. gallica* had small prismatic crystals only in their ovaries. No crystals were observed in the other tissues of these species. This study represents additional data on the presence of CaOx crystals in Asteraceae.

Key words: Asteraceae, calcium oxalate crystals, Inuleae, Inula, Pulicaria, Filago, Logfia.

SEED DEVELOPMENT IN ASTRAGALUS CEMERINUS AND A. RUSCIFOLIUS (FABACEAE), AND ITS SYSTEMATIC IMPLICATIONS

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This study focuses on seed development in Astragalus cemerinus and A. ruscifolius, two endemic species of Astragalus in Iran. In both species the ovules are campylotropous, bitegmic and crassinucellate. Two polar nuclei fuse before fertilization, forming the diploid secondary nucleus. Division of the primary endosperm nucleus gives rise to coenocytic endosperm; however, part of it becomes cellular at the late globular stage. The first division of the zygote is transverse and the embryo proper forms after several divisions of the terminal cell. The mature suspensor consists of a mass of cells equal in size to the globular embryo proper, with several inflated cells towards its base. This massive suspensor seems to be plesiomorphic, as compared with the biseriate suspensor known only in section Incani. Abnormalities in the embryo proper as well as in the suspensor are observed at the globular stage. In both A. cemerinus and A. ruscifolius, fusion of the polar nuclei occurs in the median regions of the central cell and before fertilization occurs, as is the rule in most of the Papilionoideae, but in species of section Incani as in a few other species of the family, the polar nuclei approach the egg apparatus before fertilization and do not fuse until fertilization. The embryological characters of A. cemerinus and A. ruscifolius are compared with those of other species of Astragalus, and the taxonomic application of these characters as well as their phylogenetic significance are discussed.

Key words: Abnormal embryology, *Astragalus cemerinus*, *Astragalus ruscifolius*, megagametophyte, embryo proper, suspensor, phylogeny.