XXVII INTERNATIONAL EUCARPIA SYMPOSIUM SECTION ORNAMENTALS.

FROM NATURE TO CULTURE: BREEDING ORNAMENTALS FOR SUSTAINABILITY

Genoa (Italy). July 2-5, 2023

ABSTRACTS

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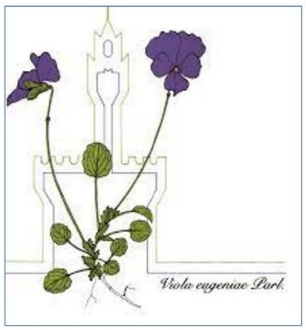

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INTRODUCTION

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Ornamentals, e.g., cut flowers, ornamental plants, trees, and bulbs, is undoubtedly the most diversified and fast-changing agriculture sector. The global floral industry is expanding in both production and trade. The world cultivated surface for ornamentals is about 745,000 ha (Hübner, 2020) and the sector is estimated to grow at a compound annual growth rate (CAGR) of 4.3% from 2023 to 2031 reaching USD 271.3 Mn by the end of 2031 (Rabobank, 2022). Changing in life style of people, increase in population and rising the spending capacity of consumers have addressed new trends in flowers consumption. The purchase relates to the all-year consumption of flowers and ornamental plants and the customers have a more pronounced attention to the well-being and the ecological aspects. In this light, innovation and sustainability are key factors for satisfying the nowadays flowers consumption evolution.

The ornamental plant breeding is an important step to meet the several issues arisen in this framework *viz.* modified consumption trends showing an increasing consciousness about biophilia and "neo-ecology" sentiments; evolution and spreading of information and communications technology (ICT) and internet; alterations arisen from climate changes; necessity to reduce the energy-input and to reach a production flow sustainable under the environmental, social and economic point of view; pest management performed with alternative strategies and tools to reduce the agrochemicals; need to provide end-consumers with an increased product quality at an acceptable price.

From July 2 to 5, 2023, the **XXVII International EUCARPIA Symposium Section Ornamentals - From Nature to Culture: Breeding Ornamentals for Sustainability** is held under the aegis of the European Association for Research on Plant Breeding (EUCARPIA; https://www.eucarpia.eu/) and the International Society for Horticultural Science (ISHS; https://www.ishs.org/). The symposium is organized by the University of Genoa and the Botanical Hanbury Gardens (http://www.unige.it; https://gbh.eucarpia27.unige.it/it) and supported by Ligurian Region (https://www.regione.liguria.it/), the Floriculture District, the Italian Society for Horticulture Science (SOI; https://www.soihs.it/) and the Italian Botanical Society (SBI; https://www.societabotanicaitaliana.it/).

The theme of the symposium is explored in four sessions addressed to (1) the use of wild plants as sources for the breeding programs. (2) the strategies which could be adopted during the breeding programs to get new products and to enhance a sustainable production, (3) the aid of new genetic tools in the breeding programs and (4) the new uses of ornamentals for landscaping and to create an added value to the territory. Nine key note speakers, renowned at international level, will

introduce the four sections by highlighting the scientific approaches and giving case-studies which could elucidated the important fallout in the sector.

During each ISHS Symposium two Young Mind Awards for junior scientists are given; 15 young scientists applied for this competition in this event and a special committee will declare the winners.

The last day of the symposium, the 5th July, is dedicated to the technical tours. Two optional tours are offered, the first one is addressed to visit the floriculture reality in the Western part of Liguria (Albenga and Sanremo surrounding); we would like to express our gratitude to **Azienda Agricola Biologica RaveraBio** (www.raverabio.com) and to **Diemme Fiori** (www.diemmeexport.com) for their availability. The second technical tour is dedicated to visit Villa Pallavicini in Genoa Pegli (https://www.villadurazzopallavicini.it) and other gardens and parks in Genoa included the Botanical Garden of the University of Genoa.

The 6th July, a post-symposium tour is scheduled to discover the Ligurian Riviera and the Nature Park of Portofino from the seaside, sailing along the coast. We are grateful to the **Authority of Portofino Park** (http://www.parcoportofino.it/parcodiportofino) for the support.

It is expected that about one hundred people involved in the field at different extent and from more than 17 countries join the event; 65 presentations (orals and posters) were submitted and in this issue the abstracts are published. The full-text of the articles will be available in the **Acta Horticulturae** (*ISSN 0567-7572 print and ISSN 2406-6168 electronic*) which is a peer reviewed series published by ISHS.

KEY NOTES AND COMMUNICATIONS

SESSION I. BIODIVERSITY AND ACCESS TO GENETIC RESOURCES

FROM WILD SPECIES TO ORNAMENTAL CROPS. A NEVER-ENDING STORY

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Plant ornamental industry is characterized by the enormous amounts of products, it is even difficult to try to classify them, everything is subject of commercialization, stems, leaves flowers, fruits and whole plants. Products are breed for every purpose and for every region of the world. However, every product has started in the same way, from a wild species. Ornamental plant breeding does not differ much from other crops, mainly for one particular thing, diversity, which is the most valuable characteristic while breeding ornamental crops. There is a general path for the generation of new ornamental varieties, which includes several steps that must be considered. These steps are:

i) Parental selection,
ii) Chromosome analysis,
iii) Crosses,
iv) Pollen tube growth,
v) Embryo growth,
vi) Fertile plants,
vii) New variety.

However, pre- or post- fertilization barriers might be present in between some steps, and biotechnological tools are often needed, some of these tools include: somatic hybridization, cut-style pollination, in vitro pollination, mentor pollen, embryo rescue, chromosome doubling and backcrossing. Which and how many tools should be used will vary regarding the species involved in the generation of a new variety. One of most important factors for breeding is a wide genetic base, the wider this is, the higher amount of variation that could be achieved at the beginning of a breeding program, which will ease the selection of new characteristics in novel cultivars. Nowadays, species from tropical and subtropical countries are becoming more important in terms of cultivated hectares and research involved in its generation. In this work some examples of breeding new ornamental varieties from wild species will be given, and the methods to obtain the final products will be depicted.

KEY WORDS: Breeding, Hybridization, Ornamental crops.

ETHICAL PLANT ACCESS – EXPERIENCES IN NEW ZEALAND

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Māori are the indigenous people of Aotearoa – New Zealand (NZ). In 1840 the Treaty of Waitangi was established between Māori and the Crown, thereby guaranteeing undisturbed possession of taonga (treasured) species. There were many breaches of the Treaty of Waitangi which resulted in the establishment of the Waitangi Tribunal to rectify the breaches of the past, and the resulting Settlements with Iwi – tribal sovereign nations of Māori. One such report, WAI 262, known as the flora and fauna claim, asserts Māori sovereignty over taonga species – those endemic to NZ and of significance to Māori in both traditional and contemporary contexts.
The WAI 262 report, alongside international agreements such as the Nagoya Protocol, create a moral and ethical context for authentic partnerships and benefit sharing between indigenous people and industry/science, including breeding and use of taonga plant species.
This paper will be co-presented with both an indigenous perspective and a science perspective, providing examples of how indigenous science partnerships can create innovative opportunities for the international ornamentals industry, within the moral and ethical frameworks of international policies and commitments when based on best practice principles. The examples include projects spanning several genera (e.g. *Leptospermum, Gentianella, Syzygium*, and *Corynocarpus*), illustrating increasing success for authentic partnerships between Māori and industry/science including; governance of data, benefit sharing agreements, informed consent, and facilitated access.

KEY WORDS: indigenous rights, breeding, plant genetic resources, governance of data, benefit sharing, informed consent.

IRANIAN *DIONYSIA*, DOMESTICATION TO COMMERCIALIZATION

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The genus *Dionysia* Fenzl, belonging to Primulaceae family, is a typically Irano-Turanian genus that is almost restricted to the rather dry mountains of the *Flora Iranica* area, from South East (SE) of Anatolia and West (W) of Iran to Tadzhikistan and Afghanistan.

There are more than 65 species of *Dionysia* in the world that most of them (around 49 species) are belonging to Iran. Since 2021 until now, 12 new *Dionysia* species have been introduced by Magnus Liden and others. Considering the mountainous climate of Iran, there are still more undescribed species in Iran.

The most serious threat to the long-term survival of at least the rarer species of *Dionysia*, is without doubt temperature increase and erratic precipitation correlated with current climate change. Other threatening factors are roads, illegal harvesting in some species, and mining activities. Furthermore, only a very limited number of botanical gardens and collectors propagated this precious native plant, and this genus has not yet been fully known in the floriculture industry.

This genus is important due to its ornamental and medicinal perspectives (local use) and could be used as a unique flowering pot plant. Unfortunately, the scientific studies on this species are limited and almost no scientific research has been done on their propagation and domestication. The establishment of protected areas, colonization, ex situ conservation, efficient propagation methods through seed cultivation and tissue culture techniques, collection and establishment of endangered species in Botanical Gardens and Research Centers are some strategies suggested for better conservation. This report, as the first in the field of domestication to commercialization of the *Dionysia* species, can be the first step to the world floriculture industry to introduce this species as a new plant with a good potential for landscaping design.

KEY WORDS: Conservation, Primulaceae, Climate change, Alpine plants, Ornamental plants.

SHORT NOTES ON BREEDING USING BOTANICAL SPECIES TO PRODUCE INNOVATIVE ORNAMENTAL PRODUCTS

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In nature each species had to solve problems to be successful in its ecosystem. Minding the possible ornamental value and the environment in which a species must survive, through the use of wild species it is sometimes possible to improve an existing ornamental product or create a new one.

In this short note, we will disclose the story of some botanical species used in the breeding programs of our company. For potted *Dianthus*, the use of *Dianthus alpinus* in the breeding program produced new varieties with good shooting, faster growth, and resistance to water stress and frost.

In the genus *Iberis*, the breeding between *Iberis sempervirens* and *Iberis saxatilis* generated a pot plant suitable for winter flowering with good resistance to pests and cold.

Both these species are common in the Alps between 1200 and 1800 mt of altitude; to be competitive in their ecosystem these species must grow at low temperatures and have a huge flowering to get enough seeds for reproduction. The flower size and the number of flowers per stem were improved through breeding to have an improved ornamental impact and to elongate the flowering period.

As last we will consider a genus that is typical of several regions of Italy and other seaside Mediterranean countries, the *Helichrysum*. Its silvery leaves, fast growth, and drought resistance were used in a breeding program to develop garden varieties suitable for landscaping and borders.

KEY WORDS: breeding, interspecific, *Dianthus, Iberis, Helichrysum*.

TOWARDS UNDERSTANDING THE GENOME COMPLEXITY OF HEXAPLOID *CHRYSANTHEMUM*

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Chrysanthemum morifolium Ramat. (2n=6x=54) is an hexaploid ornamental crop that is thought to be originating from up to 10 wild species in the domestication process. Although it has this expected allo-polyploid origin, segregation tested in populations mainly showed a random segregation.

To make a start in understanding the complex nature of the hexaploid genome and the use of genomics for breeding, diploid wild species genomes provide a valuable resource. The completion of the *de novo* genome assemblies of both *C. makinoi* Matsum. & Nakai and *C. seticuspe* (Maxim.) Hand.-Mazz. [accepted name: *C. lavandulifolium* (Fisch. ex Trautv.) Makino var. *lavandulifolium*] allowed us to look at the synteny between the two species and compared it also to genomes of other Asteraceae. Using resequencing data of other wild species and a number of hexaploid cultivars also provided a first glimpse in the presence of signals from the wild species in the hexaploid genepool.

KEY WORDS: Genome sequencing, *Chrysanthemum*, Asteraceae, hexaploid genepool.

CHRYSANTHEMUM ARCTICUM SUBSP. *POLARÉ* IS GENETICALLY DISTINCT FROM OTHER TAXA IN THE *CHRYSANTHEMUM ARCTICUM* SPECIES COMPLEX

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The salt-tolerant "*Chrysanthemum arcticum* species complex" consists of three taxonomically distinct species/subspecies: *C. arcticum* L., Arctic daisy [=*Arctanthemum arcticum* (L.) Tzvelev; =*Dendranthema arcticum* (L.) Tzvelev) and its two subspecies (*C.a*. subsp. *arcticum*, *C.a*. subsp. *polare* Hultén). All are native to N. America with remnant populations also occurring in the Kamchatka Peninsula (Russian Federation) and Hokkaido, Japan. Each taxon and most populations are genetically distinct with little to no gene exchange, due to a lack of sympatry among species.

Previous research found that single nucleotide polymorphisms (SNPs) clearly differentiated *C. arcticum* and *C.a*. subsp. *arcticum* as being taxonomically and genetically distinct. However, the genetic structure of *C.a*. subsp. *polare* populations and its relationship within the *C. arcticum* species complex is unknown.

Our objectives were to determine the genetic structure of wild, geographically distinct *C.a*. subsp. *polare* sites collected in Nome or Sitŋasuaq (Inupiaq), Alaska (1 population; n=81 genotypes) and Churchill, Manitoba, Canada (20 populations; n=375 genotypes). Population genetic diversity was analyzed using 971 SNP markers developed using low density DArTseq technology. Two distinct genetic clusters (delta K) within *C.a*. subsp. *polare* populations were detected by STRUCTURE 2.3.4, principal coordinate analysis (PCoA), unweighted pair group method with arithmetic mean (UPGMA) and SplitsTree. SNP data showed a clear taxonomic distinction between the two geographic locations of *C.a*. subsp. *polare*; all were also genetically distinct from *C. arcticum* and *C.a*. subsp. *arcticum*, based on 2,438 SNPs (n=989 genotypes). Principal component analyses (PCoA) had 31% of the genetic variation explained by PCoA1 (27%) and PCoA2 (4%). Ploidy levels and phenotypic traits will be integrated with the molecular data for future GWAS, QTL development and genome sequencing.

KEY WORDS: Arctic daisy, *Chrysanthemum*, salt tolerance, polyploidy, SNPs, genetic structure.

IRIS FERDOWSII, A THREATENED AND ENDANGERED IRANIAN *IRIS*

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Iran is one of the diversity centres of the *Iris* genus and more than 20 species and subspecies, with different colors and shapes, have been reported.
Iris ferdowsii Joharchi & Memariani is the last species of *Iris* discovered in Iran, which is named after the famous Iranian poet, the creator of the Shahnameh, Hakim Abolqasem Ferdowsi. The distribution of this species is limited to the gentle rocky slopes and mountain steppes of small parts of northeast Iran (Hezar Masjed Mountains). Unfortunately, due to the very small populations of this species (less than 300 in the studied area), and the factors such as land use change, overgrazing, road construction and climate change, the survival of this newly discovered species is in danger.
This species is known as one of the most beautiful *Iris* in Iran and the world due to its blue-leaning purple flowers that appear in spring; Hence, it has a high potential to be introduced as a new ornamental plant. The study of phenological growth stages of *Iris ferdowsii* in habitat was investigated based on the BBCH scale during several consecutive years and the results showed that changes in temperature and precipitation directly affect the growth indicators of the plant so that the plant height, the quality of flowers and the number of flowers are affected. Different propagation methods such as seed germination and tissue culture has been investigated and promising results have been observed so far. Undoubtedly, in-situ and ex-situ conservation is necessary to preserve the remaining plant population. Identifying and introducing new plant species that have ornamental value, in addition to preservation, can increase researchers' access to new genetic resources for plant breeding programs. *I. ferdowsii* plant is in a worrying condition and if it is not protected, there is a possibility of its destruction. It is hoped that the introduction of this new plant species will be effective in preserving it.

KEY WORDS: Biodiversity, Endemic plants, Geophyte, New plants, *Iris*.

INTERSPECIFIC HYBRIDIZATION OF *DAHLIA* (ASTERACEAE) FOR THE DEVELOPMENT OF VARIETIES WITH ORNAMENTAL VALUE

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The dahlia flower is renowned for global importance, since it is the flower with the largest number of varieties registered worldwide.

The objective of this work is to explore the interspecific compatibility of different species of *Dahlia* null or scarcely worked in obtaining fertile hybrids; hybridizations with two widely known taxa in the field of varietal generation (*D. coccinea* Cav. and *D. pinnata* Cav.) were also carried out.

Nine different species were used to carry out manual hybridization work, including 2 cases of special interest: *D. rupicola* P.D. Sørensen x *D. dissecta* S. Watson and *D. dissecta* x *D. merckii* Lehm. The manual fertilization work was carried out in a reciprocal and iterative way, isolating each flower head until the formation of seeds, then these were planted. The hybrid descendants obtained show an intermediate morphology between both parents, dominating the maternal inheritance in the growth habit of the plants while the paternal inheritance stood out primarily in the floral characteristics.

Four hybrids of the *D. rupicola* x *D. dissecta* cross were selected, showing characteristics of commercial interest such as abundant and recurrent flowering, ligules kkkù a broader secondary yellow basal color, as well as their early development and resistance to adverse growing cultivation conditions in addition to its lack of vegetative dormancy. Four more specimens of the *D. merckii* x *D. dissecta* cross were selected due to their abundant flowering, more accentuated lilac color of the ligules and their lack of vegetative dormancy. The selected specimens were described based on the UPOV technical guide for *Dahlia*.

KEY WORDS: compatibility, offspring, flowering, selection, dormancy, *Dahlia*.

SESSION II- STRATEGIES FOR BREEDING AND SELECTION OF NEW SUSTAINABLE ORNAMENTALS

SUSTAINABLE PRODUCTION OF GREENHOUSE ORNAMENTALS USING PLANT GROWTH PROMOTING BACTERIA

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Phosphorus (P) is an essential macronutrient with low availability for plant uptake. The bioavailability of P is reduced by the formation of insoluble complexes with calcium (Ca) and iron (Fe). Phosphorus solubilizing bacteria (PSB) can enhance P uptake by producing organic acids that acidify the rhizosphere and break down the insoluble P compounds.

The goal of this research is to identify PSB that can increase P uptake efficiency in soilless production systems. A collection of bacteria isolated from the rhizosphere of greenhouse-grown ornamentals was used to identify PSB using both in vitro and in planta evaluations. A malachite green assay optimized for 96-well plates was used to screen 1,056 bacterial isolates for the ability to solubilize phosphorus from both calcium phosphate and iron phosphate.

This in vitro assay identified 18 and 24 PSB that solubilized 25% or more of the P from $Ca_2(PO_4)_3$ and $FePO_4 \cdot 2H_20$, respectively. There was no overlap between the PSB that solubilized $Ca_2(PO_4)_3$, and those that solubilized $FePO_4$. Digital phenotyping with the TraitFinder (Phenospex) is being used for in planta evaluations of growth promotion in *Tagetes patula* L. (french marigolds) and *Solanum lycopersicum* L. (tomato) inoculated with individual PSB as a media drench. Digital biomass is used to quantify growth promotion, and hue index, green leaf index, and plant senescence reflectance index quantify the severity of nutrient deficiency symptoms. The malachite-green assay and digital phenotyping are suitable tools for the rapid identification of PSB that can be used to improve the sustainability of greenhouse ornamental production by reducing fertilizer inputs.

KEY WORDS: Plant Growth Promoting Bacteria, Phenotyping, Phosphorus, Greenhouse, Ornamentals.

THE MYCORRHIZA RESPONSIVENESS OF A *PETUNIA* RIL POPULATION IS INFLUENCED BY ENVIRONMENTAL AND GENETIC FACTORS

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Petunia is a valuable annual bedding crop known for its diverse colors and morphologies. Its small size, simple growth habit, and ease of cultivation make it an ideal model plant for studying the interaction between plants and mycorrhizal fungi. This mutualistic relationship, in which the plant trades sugars and lipids in exchange for mineral nutrients from the fungus, in particular phosphate (Pi) leads to an increased nutrient and water use efficiency of the plant, and to enhanced resistance and tolerance to pathogens and abiotic stress factors. This interaction could be of great importance for sustainable plant production systems. However, the unpredictable outcome of this symbiosis limits its use in agriculture and horticulture. Many studies have shown that the variation is at least partially due to the genotype of the plant, which implies that mycorrhiza-responsiveness could be a breeding target.

This study used a subset 19 recombinant inbred lines, derived from a cross between *Petunia axillaris* (Lam.) Britton, Sterns & Poggenb. and *Petunia exserta* Stehmann, to assess their responsiveness to the mycorrhizal fungi *Rhizophagus irregularis* (Błaszk., Wubet, Renker & Buscot) C. Walker & A. Schüßler and the impact on plant growth at three different light and temperature regimes.

The result showed a genotype-dependent gradient in AM-responsiveness, which was overlayed by a condition-dependent component. A subsequent analysis identified an optimal light regime for AM-responsiveness, independent of the plant genotype. These findings have significant implications for petunia cultivation and for a better understanding of the petunia-mycorrhiza interaction.

KEY WORDS: petunia, recombinant inbred lines, arbuscular mycorrhiza, plant-microbe interactions.

BREEDING OPPORTUNITIES IN GERANIACEAE: ENHANCING GENETIC DIVERSITY AND ORNAMENTAL TRAITS

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Hardy geranium is a popular perennial ornamental plant with high demand in the market.
In this study, we aim to create novel genetic diversity in the genus through three approaches: hybridization, polyploidisation, and hairy root induction. Interspecific and intergeneric hybridizations were performed using 42 genotypes of a collection. Data were collected on crossing barriers and a model was developed to predict success rates of specific cross combinations. Polyploidisation experiments using mitotic inhibitors (colchicine, oryzalin and trifluralin) were done in three genotypes. The efficiency of the different mitotic inhibitors was determined. Finally, protocols for hairy root induction by co-cultivating *Agrobacterium rhizogenes* (Riker et al.) Conn (= *Rhizobium r.* Riker et al.) and subsequent plantlet regeneration were developed for hardy geranium.
With this technology, we aim to introduce rol-genes in the genus, in order to create naturally compact growing phenotypes. The success of the different approaches was evaluated by examining the morphological, cytological, and molecular characteristics of the resulting genotypes.
This study provides insights into the potential of hybridization, polyploidisation, and hairy root induction techniques for creating novel genetic variations in hardy geranium.

KEY WORDS: Hybridization, polyploidisation, hairy root, diversity, *Geranium*.

A BIO-ASSAY TO SCREEN *ILEX CRENATA* FOR TOLERANCE TO HIGH SOIL PH AND BLACK ROOT ROT RESISTANCE

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Ilex crenata Thunb. or box-leaved holly has become a popular alternative to boxwood (*Buxus* spp.) as a result of the plants' similar appearance. However, box-leaved holly prefers acidic soils whereas boxwood prefers alkaline soils, which are more prevalent in private gardens. Placed in alkaline soils, box-leaved holly experiences problems in nutrient uptake and black root rot caused by *Berkeleyomyces basicola* (Berk. & Broome) Ferraris [previously *Thielaviopsis basicola* (Berk. & Broome) W.J. Nel, Z.W. de Beer, T.A. Duong & M.J. Wingf.], a soil-borne fungus that also prefers alkaline conditions. Breeding box-leaved holly for tolerance to alkaline soils and resistance to *B. basicola* can solve these issues. So far, cultivars mostly are the result of lucky findings. Recently, targeted breeding has gained interest, which would include improving (a)biotic stress tolerance.

To screen potential breeding parents and seedlings for tolerance to high soil pH and black root rot, a bio-assay is needed. To develop a protocol for this bio-assay, rooted cuttings in soil of pH 4, pH 5.5 or pH 7 were inoculated with *B. basicola* or treated with water (negative control). Three *B. basicola* genotypes were tested after a selection based on a genetic diversity analysis. Disease symptoms and plant biomass were evaluated after 12 weeks. Especially the combination of a high soil pH and inoculation with B. basicola caused severe discoloration of the roots and growth inhibition of the shoots. Application of only one of these stress factors led to less severe symptoms.

KEY WORDS: *Ilex crenata*, *Thievaliopsis. basicola*, Bio-assay, soil pH.

GENOMIC ANALYSIS OF INFLORESCENCE DEVELOPMENT AND DOUBLE FLOWERING IN BIGLEAF HYDRANGEA

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Hydrangeas are one of the most valuable woody plants in the nursery trade. They are popular worldwide for their large, showy inflorescences. Novel inflorescence shapes and flower forms are highly desired by breeders, producers, and retail consumers, yet little is known about the genetic regulation of inflorescence or flower development in hydrangea.

The objectives of this research are to 1) generate a high-quality, chromosome level genome assembly for bigleaf hydrangea, and 2) integrate this genome assembly with other genomic tools to identify candidate genes for inflorescence type and double-flowering in bigleaf hydrangea.

Two high-quality reference genomes of hydrangea cultivars Veitchii and Endless Summer were assembled using PacBio long read sequencing. The 'Veitchii' and 'Endless Summer' genomes are 2.21 Gb and 2.22 Gb, respectively, each scaffolded into 18 pseudochromosomes. An F1 population between 'Veitchii' and 'Endless Summer' was used for linkage mapping of the inflorescence shape trait using genotyping by sequencing.

By mapping highly selective markers associated with the inflorescence trait to both reference genomes, CYP78A5 located on chromosome 4 was identified as the candidate gene for inflorescence shape. A novel gene, BAM3 located on chromosome 17, was identified as a candidate gene responsible for double flowering. The genome resources and the candidate genes presented here will help researchers and breeders determine genetic mechanisms driving flowering characteristics and floral development to develop high-value hydrangea cultivars for the nursery industry and the public.

KEY WORDS: Hydrangea, inflorescence shape, double flowering, floral regulation.

EVALUATION OF FROST TOLERANCE SCREENING METHODS TO SUPPORT HARDY LAVENDER BREEDING

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Lavender (*Lavandula* spp.) is a shrub with origins in the Mediterranean, with *L. angustifolia* Mill. being the most widely used as a perennial ornamental plant. Other lavender species are also becoming more popular as annual garden plants because of their good drought tolerance. In temperate regions, however, their lack of frost tolerance limits their use as a perennial garden plant. Enhancing frost tolerance of *Lavandula* via breeding and selection could further exploit the ornamental potential of these species. Accurate assessment techniques and knowledge of the physiological mechanisms of frost tolerance can facilitate the breeding process.

In this study we used both field observation of frost damage and the measurement of electrolyte leakage after a controlled freezing treatment to evaluate the frost tolerance of our lavender collection of 38 genotypes.

Differences in frost tolerance were observed among the genotypes using these methods. Both methods were compared to other assessment techniques, i.e., measurements of chlorophyll fluorescence, water content, and proline accumulation. Hydraulic conductivity measurements were used to assess frost-induced embolism formation and the resulting possible disturbances in water transport. The conductivity data were correlated to xylem vessel diameter to generate insight into the physiological mechanisms involved in frost tolerance in different lavender species. Differences in accuracy, effectiveness and feasibility of the evaluated methods to screen large numbers of plants for their frost tolerance were noted in relation to the various techniques and plant parameters. We anticipate that the use of a standardized method under controlled conditions could make it possible to select for lavender genotypes with improved frost tolerance regardless of yearly variations in winter conditions, which would accelerate the total breeding process.

KEY WORDS: chlorophyll fluorescence, electrolyte leakage, embolism, freezing tolerance, hydraulic conductivity, *Lavandula*, xylem.

IN VITRO BIOTECHNOLOGY TO SUPPORT ANEMONE BREEDING (*ANEMONE CORONARIA* L.)

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Anemone coronaria L. (poppy anemone) is a winter-flowering allogamous species marketed as a cut flower or garden ornament. To produce cut flowers, the breeders aim to obtain new improved varieties with early flowering, flowers with larger and wider petals and sepals, long thick stems resistant to storage and transport and a uniform product. The cultivars are subclassified as sub-cultivars based on their flower color, and these represent a population of hybrids from crosses between a small number of selected parents. As these parental lines are typically highly heterozygous, the resulting sub-cultivar is genetically non-uniform. F1 hybrid strategy based on homozygous parents is widely used for breeding programs and two strategies are available to produce appropriate parental material: selfing (but anemone suffers inbreeding depression) or the induction of doubled haploids.

At the CREA of Sanremo the protocol to obtain *A. coronaria* plants from anthers (microspores) was developed and widely described (Copetta et al 2018; Copetta e Laura 2021). As anemones are recalcitrant to in vitro culture, even when many embryos have been obtained, their conversion to seedlings is not obvious and regeneration rates are low despite a high induction rate.

Applying this protocol, corms deriving from another culture were obtained for 8 cultivars (Biancheri Creazioni): 2 diploids (Magenta and Rosa) and 6 tetraploids. Part of the corms obtained in vitro were acclimatised and androgenic plants were obtained. The plants were grown and the colour, the number and size of the petals, the length and the thickness of the stems were evaluated. Furthermore, mature leaves of acclimatised plants were collected and used for evaluation of ploidy degree by cytofluorimetric analysis to identify di-haploid, double-haploid, haploid and chimera plants.

Many morphological differences are visible between androgen plants and mother plants such as the number, shape and colour of the petals, the length and thickness of the stem and these differences have also been detected among regenerants deriving from the same mother plant.

KEY WORDS: *in vitro* culture, androgenic plants, flower morphologies, cytofluorimetric analysis.

NITROUS OXIDE TREATMENT INCREASES THE PROPORTION OF VIABLE POLLEN AND THE POLLEN SIZE IN *LIMONIUM PEREZII*

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Interspecific crosses between *Limonium sinuatum* (L.) Mill. and *L. perezii* (Stapf) F.T.Hubb. have produced hybrids (*Limonium* siNZii^{TM}) utilized by the ornamental industry. The inclusion of genotypes as parents in the breeding program depends, among other factors, on their fecundity.
Two *L. perezii* genotypes 'per11' and 'per275' with comparatively low male fertility (zygote success <40% ± 16% for both genotypes when used as pollen donors in intraspecific crosses), were selected for this study. We had observed that pollen performance of the two genotypes improved after nitrous oxide (N_2O) treatment.
To understand what was contributing to this improvement, the aim of this study was to evaluate the effect of N_2O on pollen viability and size on these two genotypes following treatment of inflorescences in early development.
The proportion of stained pollen increased up to 1.7 times in 'per11' and up to three times in 'per275'. The size of pollen grains also increased between 1.2 and 1.5 times in both genotypes. These findings showed that N_2O treatment not only affects pollen viability but also pollen size. It is proposed that changes in pollen staining and size could be used to detect the optimum stage of inflorescence development to apply N_2O treatment for best pollen viability.

KEY WORDS: Ornamental plants, sea lavender, modified Alexander's stain, non-aborted pollen, aborted pollen.

SESSION III: NEW GENOMIC TECHNIQUES AND ORNAMENTAL PLANT BREEDING

GENE EDITING TO SUPPORT BREEDING IN ORNAMENTAL SPECIES

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Breeding of ornamental species is primarily focused on creating new varieties with unique ornamental value, but in recent years developing resistance to pests and diseases has become increasingly important. Traditional breeding approaches based on crossing and selection can be made more efficient e.g. by using marker-assisted selection. However, for many ornamental species molecular selection methods are not available and breeding remains challenging. The emergence of New Plant Breeding Techniques, such as gene editing using CRISPR-Cas, holds great promise for supporting the breeding of ornamental crops. Targeted mutagenesis via gene editing can modify specific ornamental features without altering other unique variety characteristics. Additionally, by targeting susceptibility genes, gene editing can provide a new source of disease resistance. To realize these promises, several conditions must be met. For instance, the delivery of gene editing tools into plants depends on the availability of efficient transformation and regeneration methods. Moreover, knowledge of gene function and genomic information in target crop species and genotypes is a prerequisite for the application of gene editing.

This lecture will explore the possibilities and challenges of applying gene editing in ornamental species. It will provide examples of gene editing applications in some ornamental crops and discuss the opportunities offered by this technique for breeding new ornamental varieties with improved resistance to pests and diseases.

KEY WORDS: Gene editing, CRISPR-Cas, Targeted mutagenesis, Ornamental species, New Plant reeding Technique.

EFFICIENT GENOME EDITING IN CARNATION (*DIANTHUS CARYOPHYLLUS*) USING CRISPR/CAS9 SYSTEM

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Carnation (*Dianthus caryophyllus* L.) is one of the most popular and widely used ornamental flowers worldwide. Although the genome sequencing, the mechanism of flower color formation and petal senescence have been reported, the efficient CRISPR-Cas genome editing toolkits in carnation has not been established.

In this study, we identified two DcU6 promoters (DcU6-1 and DcU6-2), and also established a fast and effective carnation protoplast screening system using NanoLUC as the reporter to screen the high-efficiency elements in CRISPR/Cas9 system. The results demonstrated that DcU6-2 promoter exhibited more effective role than other promoters (DcU6-1 and AtU6) in the editing efficiency in carnation protoplast.

Besides, we found that SlEF1α promoter from tomato could significantly increase the editing efficiency by driving the expression of AtCAS9. DcPDS was chosen as a target to further identify the editing efficiency using the CRISPR/Cas9 system containing DcU6-2 promoter, SlEF1α promoter and AtCAS9. Our study provides an optimization of the CRISPR/Cas9 system in carnation.

The application of gene editing technologies will be used for the improvement, especially for disease resistance, abiotic stress tolerance, herbicide tolerance, shelf life, and yield in flower plants.

KEY WORDS: *Dianthus*, carnation, genome editing.

GENOMIC TOOLS TO ENABLE DEVELOPMENT OF NEW SUSTAINABLE ORNAMENTAL PRODUCTS

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Ornamentals constitute a diversified portfolio of products where more and more differential advantages and wide sustainability goals are expected. While agricultural crops have received a large amount of attention and consortia/public funding to advance molecular breeding, efforts in ornamentals are often individual and more modest.

In this talk, we would like to present the latest state-of-the art technologies that enable molecular breeding in ornamentals. We will show use cases to clarify how such tools can contribute to a faster and more efficient selection. The challenges and opportunities of molecular tools for ornamental crops will be elucidated. Starting from simple molecular markers to select for resistant varieties to editing certain targets for complex traits. Traits such as disease resistance, transportability, vase life, new aesthetic shapes, and lower pesticide input are high on the breeders' agenda. Understanding the natural variation within the germplasm and exploiting that variation is crucial to reach such goals in a reasonable amount of time despite long generation times and complexity genetics and genomics.

KEY WORDS: genomics; natural variation; complex genetics; disease resistance.

SUSTAINABILITY IN ORNAMENTALS NEEDS EFFECTIVE PROTECTION OF INTELLECTUAL PROPERTY

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The UPOV Plant Breeders´ Rights system is the tailormade system for the protection of new plant varieties. However, the latest UPOV Act is from 1991, thus more than 30 years old. The advent of new breeding techniques, climate change, the ongoing globalization and the demand of the consumers for sustainable production bring challenges for both the breeders and for the UPOV system. Climate change urges breeders to develop new varieties which adapt to more extreme climatic conditions, such as increased heat or cold, drought, floodings or humidity. Consumers demand flowers and fruits with new colors, shapes and taste, from sustainable production, produced regionally with less use of pesticides and water. Both climate change and consumer demand require higher investments in breeding and research, which requires more financial means.

The ongoing globalization results in an increased production in territories where the protection of Intellectual Property for plants is not or not sufficiently effective. The flowers and fruits so produced are shipped to consuming regions. If breeders cannot monetarize their innovations/varieties in the producing countries or cannot act effectively against infringers, they rely on a solid and robust protection of their varieties in the consuming territories.

Finally, the new breeding technologies put at risk the conventional breeding, because they allow multiple changes to one or a series of varieties in a short period of time. The so developed new varieties compete on the market with their initial varieties. Conventional breeders must be in the position to negotiate with the biotech companies about the commercialization of the new varieties, which at the end carry almost the entire genome of the initial variety. Otherwise, the conventional breeders lose a significant part of their income, which is the basis for a sustainable continuation of their business. The presentation shows how UPOV and its members respond to these challenges

KEY WORDS: UPOV, EDV, Climate Change, Conventional Breeding, PBR, New Breeding Techniques.

HIGH -DENSITY GENETIC MAP CONSTRUCTION AND QTL ANALYSIS OF FLOWER COLOR TRAITS BASED ON A TETRAPLOID ROSE GENOME

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Modern roses, with its high economic values and irreplaceable cultural symbols, is one of the most important ornamental plants. Because of its complicated genetic background and tetraploid identity, the creation of high-density genetic maps has been a tricky problem, which slows the pace of molecular breeding progress for modern roses. Based on the existing diploid rose genomes, the construction of tetraploid genetic maps may lead to inaccurate marker information and genotyping results, thus a tetraploid rose genome is needed to meet the demand.

In this study, we presented the first tetraploid, highly heterozygous rose genome of *R.* 'Yunzheng Xiawei' based on Illumina, PacBio, and Hi-C scaffolding technologies. Combining this reference genome and tools for polyploid analyses, we performed the inheritance analysis of modern roses and constructed a near-saturated rose genetic map. We identified several key genes related to flavonoid and carotenoid biosynthesis based on the genome-wide QTL analysis. The study provides basis for the genetic analysis of highly heterozygous tetraploid roses and may facilitate the progress of marker assisted breeding in modern roses.

KEYWORDS: *Rosa*, tetraploid genome, genetic linkage map, QTL analysis, flower color.

BREEDING FOR DROUGHT TOLERANCE IN *CHRYSANTHEMUM* WITH *AGROBACTERIUM RHIZOGENES* AS A NATURAL GENE-DONOR

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Climate change and its consequences, we are all aware of it. The urgence to weapon ourselves in the battle against drought is high and plants, also ornamentals, must be bred to withstand longer periods of drought.

In this project we use chrysanthemum as a model to develop a new sustainable breeding technology that focuses on the roots, using *Agrobacterium rhizogenes* (Riker et al.) Conn (= *Rhizobium r.* Riker et al.). Natural strains of *A. rhizogenes* contain a unique Ri plasmid (which includes the *rol* genes) that allows them to transfer and incorporate the T-DNA genes lying on this plasmid into the plant genome. The result is extreme root formation (so called hairy roots). When these roots in turn regenerate into a plant, one obtains Ri plants. The presence of the Ri genes in these plants results in a typical phenotype with a more pronounced root system and more compact growth, as well as changes in flowering and leaf morphology.

We want to evaluate the effect of the altered rooting on the drought tolerance of the plants. We will present our results on the creation of the first Ri plants in chrysanthemum, which were fully characterized morphologically as well as genetically. Copy-numbers of the inserted genes as well as the position of these genes on the chromosomes were identified (using digital PCR and FISH, respectively). Since Ri-plants are to be seen as pre-breeding material due to unwanted negative phenotypical features, we will also demonstrate the successful outcrossing of the genes in an F1 and F2 population. Also, the outcome of the first experiments on root physiology and the relatedness to drought will be presented

KEY WORDS: dPCR, qPCR, breeding, FISH, root physiology.

This work was done as part of the project 'RootsPlus', which is carried out under the second call of the ERA-NET Cofund SusCrop, being part of the Joint Programming Initiative on Agriculture, Food Security and Climate Change (FACCE-JPI). SusCrop has received funding from the European Union's Horizon 2020 research and innovation programme under grant agreement No 771134. More information on the project partners can be found on www.rootsplus.eu.

THE FIRST REFERENCE GENOME OF *RANUNCULUS ASIATICUS* L. REVEALS A KEY REGION FOR ANTHOCYANIN PIGMENTATION

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Persian Buttercup (*Ranunculus asiaticus* L.; 2x=2n=16; estimated genome size: 7.6Gb) is an ornamental and perennial crop native of Asia Minor and Mediterranean basin, marketed both as cut flower or potted plant. In 2019, its production counted for the 0,4% of the total turnover of cut flowers and foliage, with the highest values in Italy (132 million of stems) and 300-350 ha of cultivated surface. The large size of the genome, its repetitively and the high level of heterozygosity has made challenging the assembly of a reference sequence, using a short-reads sequencing approach.

Thanks to the recent advances in long-reads sequencing (e.g.: Oxford Nanopore Technology, ONT), the first version of the *R. asiaticus* genome was here assembled and annotated. Furthermore, genotyping data on two F1 mapping populations, sharing a common progenitor, were used for the construction of the first genome-anchored linkage map in the species, which successfully identify eight linkage groups (correspondent to the haploid chromosome number of the species). The developed map was then used for quantitative trait loci (QTLs) analysis on vegetative and floral traits gathered for three growing seasons (2020-2022), identifying a major QTL for anthocyanin pigmentation. Finally, the sequence information was used for the development of a derived cleaved amplified polymorphic sequences (dCAPS) marker, the first reported molecular tool for varietal development in *R. asiaticus*.

KEY WORDS: *Ranunculus*; genome sequencing; QTLs; anthocyanins; linkage maps; ornamentals.

UNRAVELLING THE GENETIC BASIS OF ROSE TRANSFORMATION WITH *AGROBACTERIUM RHIZOGENES* USING A GENOME-WIDE ASSOCIATION STUDY

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The 'RootsPlus' project is a European collaboration between research institutions in Belgium, Germany, Poland and Romania and two companies. Natural transformation with *Agrobacterium rhizogenes* (Riker et al.) Conn (= *Rhizobium r.* Riker et al.) wildtype strains shall be developed as a new breeding technology for apple, chrysanthemum, sunflower and rose. *Agrobcterium r.* contains a unique Ri plasmid, that allows it to transfer and integrate its T-DNA into the plant genome. The integrated genes lead to the development of an enhanced root formation, so called "hairy roots". Subsequently, Ri plants can be regenerated from these roots in vitro. The presence of the Ri genes results in a typical phenotype with a more pronounced root system and more compact growth, as well as changes in flowering and leaf morphology. In many countries, for these natural transformants the regulations for GMOs (genetically modified organisms) do not apply.

The major objective of this study is to investigate the genetic basis of transformability in rose. Using a GWAS (genome-wide association study), genes and genomic regions will be identified that are important for transformability by *A. rhizogenes*. For a panel of 105 cut and garden roses the genotypic data is already present in form of the 68k Axiom WagRhSNP chip. Per genotype, thirty in vitro leaf explants were transformed in two repetitions with a bacterial strain carrying the reporter gene GFP (green fluorescent protein) and thus delivered the phenotypic data by counting of fluorescent hairy roots. Peaks were detected in genomic regions, that co-localize with genes, that are already known for their importance for adventitious root formation. In addition, peaks in other genomic regions were identified which do not seem to have a direct connection to root formation. These will be evaluated for SNPs with significant effects and the underlying genes, since they are expected to contain crucial factors for the transformation process itself.

KEY WORDS: *Agrobacterium rhizogenes*, transformation, GWAS, Ri plants, hairy roots.

NONCODING GENOME: CONTROLLING PLANT MORPHOLOGY AND FLOWERING

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Understanding the control of plant morphology and floral transition in plants is critical to ensure production under changing climate conditions. However, the molecular mechanisms of phase changes of crop plants are less well studied primarily due to the complexity of the genome as the crop of agricultural significance usually has a polyploid genome where genes are present in multiple copies. Recent advances in genomics have highlighted the vital role of the non-coding part of the genome. The protein-coding regions account for only a tiny portion of the eukaryotic genome, and most of the genomic regions transcribe copious amounts of non-coding RNAs. MicroRNAs (miRNAs) are small non-coding RNAs consisting of 20 to 24 nucleotides that play central regulatory roles in gene expression. miRNAs, such as miR156 and miR172, have been shown to play significant roles in various pathways of the model plant, *Arabidopsis*, that regulate the onset of flowering by acting in either negative or positive ways to initiate the reproductive phase. miR156, a highly conserved plant miRNA, plays master regulator roles in vegetative development and the extension of the juvenile stage of plant development. On the other hand, the expression of miR172 has been correlated with the reproductive stage.In this presentation, we will discuss our work on the functional analysis of soybean microRNA in ornamental tobacco.
Our results showed that ectopic expression of the microRNAs, *gma-miR156a* and *gma-miR172a*, in tobacco regulate multiple morpho-physiological traits that could be used to enhance crop yield under changing climate conditions.

KEY WORDS: Tobacco, Floral Transition, MicroRNA, Morpho-Physiological Traits.

TOWARDS GENETIC CHARACTERIZATION OF REMONTANCY IN *HYDRANGEA MACROPHYLLA*

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Hydrangea macrophylla (Thunb.) Ser. is a perennial shrub used as an ornamental crop plant for indoor and outdoor cultivation. Natural floral initiation occurs in autumn under cool temperatures and short-day conditions. Then, initiated flowers rest over winter in buds and bloom in the summer of the following year on two-year-old shoots. However, some cultivars are remontant and initiate flowers also on newly emerging, one-year-old shoots, which bloom within the same season. Because of its ornamental attractiveness, remontancy is a highly desired trait for breeding. However, the physiological and molecular mechanisms of remontancy are widely unknown in *H. macrophylla*.

Therefore, we performed floral induction experiments using the remontant cultivars 'Diva fiore' and 'Mak20' and as control the non-remontant cultivar 'Libelle'. In these experiments, we found that 15°C and short-day-conditions promote floral initiation in 'Libelle', whereas both of the remontant cultivars initiate flowers independently from temperature and photoperiod treatments. On the other hand, we detected two mechanisms of remontancy, which act most likely separately in *H. macrophylla*: i) floral initiation of apical and lateral meristems after floral induction, and ii) permanent "*de novo*" floral initiation of apical meristems of new growing shoots.

Besides these findings, we will present our current status of QTL analysis based on F1 populations derived from crosses between 'Diva fiore' x 'Mak20' and 'Libelle' x 'Diva fiore' in comparison to a backcross population derived from a cross between a F1 offspring plant of 'Libelle' x 'Diva fiore' and 'Diva fiore'. Furthermore, we present first data of a transcriptome analysis based on vegetative and generative shoot apices of 'Diva fiore', 'Libelle' and 'Mak20', which was performed to identify the molecular mechanism of "*de novo*" floral initiation in *H. macrophylla*.

KEY WORDS: floral initiation, flowering.

USING TRANSCRIPTOMICS TO IDENTIFY GENE MARKERS FOR FLOWER LONGEVITY IN DAHLIAS

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Floral longevity is a key feature of importance to cut flowers, however its regulation is not fully understood especially in composite flowers such as dahlias. Understanding how floral longevity is regulated could help to devise new strategies for reducing waste in the supply chain for cut flowers. Dahlia flowers are composed of whorls of florets within a capitulum or flower head, which develop sequentially such that inner florets are younger than the older florets at the margins. Hence, within a single flower head, a range of floret ages are represented. How the senescence of the different florets is coordinated is of fundamental interest, but is also important as the deterioration of the outermost florets determines vase life of the whole flower.

Thus, dahlia flower heads can be staged sequentially from the bud stage through opening of the majority of the florets to the sequential senescence of the floret whorls. In some flowers, ethylene is an important regulator of longevity but in others it is not. In dahlia the role of ethylene varies across different cultivars suggesting the involvement of other plant hormones in the control of longevity. Transcriptomics is a powerful technique for revealing important genes and pathways that may regulate developmental processes, especially in species for which there is no genome sequence and few genetic resources.

A transcriptomic analysis of *Dahlia pinnata* Cav. cv. Sylvia revealed that the expression of hundreds of genes differs both between outer and inner florets on the same flower head but also between inner florets on flower heads of different ages, suggesting a complex regulation of floret development. Abscisic acid (ABA) is known to accumulate during floral senescence in some species and in dahlia the expression of ABA related genes was assessed in the transcriptome analysis showing that genes encoding proteins involved both in biosynthesis and response to ABA altered in expression during floret senescence, suggesting a complex role of this plant hormone.

KEY WORDS: flower senescence, dahlias, transcriptome, abscisic acid.

SESSION IV: ORNAMENTALS FOR A SUITABLE WORLD

ORNAMENTAL PLANTS IN DIFFERENT CONTEXTS THAT FAVOR SUSTAINABILITY AND QUALITY OF LIFE

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Ornamental plants, include a group of flowering and not flowering plants that are grown purchased and used in gardens and landscapes projects, house plants for cut flower and specimen display mainly for their aesthetic proprieties primarily decorative and, as such, are appreciated according to their visual characteristics. But beyond this, we need to be concerned of other aspects while making these choices.

It's important to move the consideration and the focus while selecting the plants during the process of planting design and landscape horticulture from a largely cosmetic, decorative and functional role, to one that is also central and plays a major role within climate change, ecology and sustainability for the future, taking also in consideration how these have an impact to the quality of our life.

We will be exploring work and examples where the key element is an understanding of the 'horticultural ecology' of designed plantings, and working with 'plant communities' that are suited to site conditions, and which mimic the processes in 'natural' vegetation.

Nowadays, ornamental plants can represent important living components of urban areas, and if appropriate species are used, they can also provide important ecosystem components. It's important to remember that plants provide multiple benefits. Even those that do not excel in one particular 'service' may provide other 'services' well.

KEY WORDS: ornamental plants, sustainability, urban areas.

THE USE OF PSAMMOPHILOUS FLORA IN THE MEDITERRANEAN LOW-IMPACT GARDENS

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Climate change is one of the greatest challenges of our time on all fronts. Rising temperatures and extreme weather phenomena have driven a trend in garden design and consumer demand to increasingly require plants that are resistant to high temperatures and long periods of drought. In addition, finding solutions that save water, energy and nutrients are crucial for sustainability in agriculture. Traditionally, agriculture has responded to such needs with exotic species, but these have often proved to be invasive and thus an additional problem.

In this framework, within our flora, psammophilous plants are the perfect candidates due to their specific morpho-physiological adaptations to the harsh environmental conditions in which they grow: insolation, lack of water and nutrients, high salinity, mechanical action of the wind, inconsistent substrate.

During the PSR PSAMMbeach project, about twenty species of the indigenous psammophilous flora were selected for their aesthetic value. Ligurian (Italy) propagation material was collected and a zero-impact floricultural production was experimented. The promotion of these plants was made to land managers, parks and an educational campaign was carried out at some primary schools.

The resulting plants were used in the decoration of bathing establishments, city flowerbeds and public and private gardens with low management impact.

KEY WORDS: Sustainability, psammophilous flora, Liguria, low impact.

DRYING, A SUSTAINABLE WAY OF BRINGING DIVERSITY IN THE ORNAMENTAL SECTOR

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One of the breeding aims in the ornamental sector is to add diversity and novelty to the ornamental components. Bypassing the conventional modus-operandi of breeding or the tasks of breeders, the said breeding objectives could be achieved by the drying techniques. Dried flowers are one of the most sought-after interior trends around and this sector of the floriculture industry has gained popularity over the last ten years. Dried and preserved ornamental products offer a wide range of qualities like novelty, longevity, aesthetic properties, year around availability, eco-friendly and flexibility for flower arrangement products. Due to this increased interest, the flower drying process is moving from an empirical approach to a sophisticated interdisciplinary science where genetics, physiological, chemical-physical, biochemical and technologic knowledges are required to interact. Different strategies could be applied for the generation of dried attractive plant material including air drying, press drying, embedded drying, oven drying and freeze drying. In order to ensure best quality and color retention a fast drying and low light conditions during the process should be applied. The most challenging aspect is that it is difficult to avoid fading of colors and browning of foliage.

In our work, we would like to evaluate the best protocols to obtain dried flowers of Bunny rose and *Crossandra*, otherwise known as "firecracker flower", and dried foliage of *Polyalthia longifolia* (Sonn.) Benth. & Hook.f. ex Thwaites. *Crossandra* sp. and Bunny rose (*Rosa chinensis* Jacq. x *R. hybrida*) are popularly grown in mass to meet the regional demand as loose ornamental flowers in Southern India (especially in Tamil Nadu and Karnataka) while sometimes their over-production also causes the market glut situation and the subsequent wastage. Hence, here we had aimed to minimize the degree of loss and to give an added value as dried flowers also considering their high perishability as well as the virgin status in the dried ornamental sector. Embedded drying methods using silica gel, saw dust and white sand had been adopted in both cases. For bunny rose varieties i.e 'Rimosa-79' (red colored) and 'Gold bunny' (yellow colored), saw dust, as embedding material, under ambient condition for 12 and 12.6 days respectively had been proven its efficacy being qualifying all requisite qualitative attributes. In the case of *Crossandra* Salisb. var. 'Delhi Princess' (orange colored), silica gel as embedded material under microwave oven condition for 5 mins (temp. 118.760F) had yielded optimum quality score. The anthocyanin and carotene pigment content in the two dried bunny rose varieties were 0.078and 0.366 mg g-1 respectively while 5.99mg g-1 of carotene pigment had been retained in desiccated Crossandra

flowers. Sun drying method was found inappropriate due to causing the bleaching effectand the air drying did not attain the qualitative values. 'Leaf skeletonizing' is a process that occurs spontaneously in nature but a technique could be applied to make skeleton leaves which could be included in dry floral arrangements. Here, we had used the foliage of *P. longifolia* var. 'Pendula', commonly used in landscaping as wind break and tall to medium height ornamental hedge. The tissue digestion treatments using sodium hypochlorite (NaOCl), sodium carbonate (Na2CO3) and sodium hydroxide (NaOH), each at three graded concentrations of 5, 10 and 15%, for 60-120mins had been used here. The treatments of 10 and 15% of NaOH for 90mins had been found optimum to get qualitatively satisfactory skeletonized leaf. Boiling the leaf skeletons in a mixture of hydrogen peroxide (2%), sodium carbonate and lemon essential oil for two (2) hours allowed to yield fully bleached leaf skeletons. Conclusively, our work wanted to focus on the potential of the dry ornamental sector to enhance the multi-functional approach to ornamentals. Moreover, we would like to draw the attention of ornamental breeders concerning the potency of this sector to develop sustainable diversified ornamental products.

KEY WORDS: Dehydration, dried ornamental, sustainable, additional alternative, breeding, diversity.

WILDFLOWERS VALORISE SUSTAINABLE AGRICULTURE, LOCAL CULTURAL IDENTITY AND TERRITORIAL DEVELOPMENT

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Wildflowers are acquiring a growing attention of researchers and policy makers as a fundamental resource to be recognized as part of rural territories identity and biocultural heritage. They can be employed for the promotion of agricultural sustainability and territorial growth. Significantly, the (re)integration of wildflowers in cultivations and rural landscapes can favor the realization of bee-friendly low impact practices, promoting biodiversity preservation, and contribute to the development of integrated valorization approaches, involving cultural and touristic promotion.
Our study aims at designing testing and promoting a new model of agricultural development, based on local wildflower exploitation. Specifically, this paper presents the results of the ongoing participatory action-research (PAR) project Ecoflorland, funded on EU Rural Development funds, involving a collaboration of public and private key local stakeholders, both. The project focuses on the case of the San Gimignano territory, a rural area of Tuscany (Italy). The latter is well-known for the quality of local wines and the beauty of the landscape, characterised by small vineyards nestled on gentle slopes. Of relevance, cultivations in this area are characterized by a high sustainability, considering the presence of extensive agriculture - carried out with organic techniques, and avoiding phytochemicals - making the agro-environment rich in spontaneous flowering species, attracting pollinators and other components of the trophic web, and maintaining local biodiversity. The territory presents a large variety of traditions, typical products, and cultural heritage, attracting the interest tourists.
Project actions are intended to: i. the enhancement of San Gimignano territory agricultural sustainability and biocultural potential, by strengthening cultivations and rural landscapes through the introduction of flowering plants of beekeeping interest, respecting the care of the land and the environment; ii. the realization of an integrated territorial offer, combining both wildflowers-related traditional goods (e.g., seeds, flowers, honey, processed agri-food products) and services (e.g., touristic experiences); iii. its market qualification and territorial promotion by the development of a dedicated collective mark. As a result, a process of (economic, environmental, social, and cultural) development can be fostered, based on a synergic connection between rurality, typical products, and tourism development, and leading to an increase of incomes and diversification of agricultural businesses and other activities in the area.

KEY WORDS: wildflowers, agroecosystems, biodiversity, vineyards, San Gimignano, biocultural heritage, rural territorial development, rural landscape.

NEW INSIGHTS ON MANAGEMENT STRATEGIES FOR INVASIVE SPECIES IN URBAN ENVIRONMENT AND SELECTION OF NEW SUSTAINABLE ORNAMENTALS

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Invasive alien plants are a global issue that impacts many local ecosystems and economies. The establishment and spread of alien ornamental plants (e.g. *Buddleja davidii* Franch., *Spiraea japonica* L. and *Lonicera japonica* Thunb.) in the new areas where they have been introduced can be detrimental to biodiversity and ecosystem processes, and result in ecological and socio-economic problems. Although ornamental horticulture has been a source of invasive plants, it may also be able to provide some solutions, for example, through the development of non-invasive cultivars and the phasing out of the sale of invasive ornamentals. Particularly in urban context, a plant's invasive potential is determined by its capacity for rapid and efficient colonization, high levels of phenotypic plasticity, vegetative reproduction, high propagule pressure, fast growth rates, highly competitive ability, and avoidance of genetic bottlenecks. Some of these traits can be targeted using genetic tools and traditional plant breeding to reduce invasiveness. The processes are complex and employ forms of genetic mutation and traditional breeding to create sterile male and female plants as well as plants that produce only sterile seeds.
Novel and diverse technologies are needed to face invasive species, which requires bridging gaps between disciplines, pushing agronomists, botanists, ecologists and molecular biologists to share information and address potential alternative approaches. In the field of managing invasive species, there may be more than one formula for success. Here, we present the research framework and ideas included in the research program titled "Analysis of alien and invasive plant species in the urban environment" within the Project "National Biodiversity Future Center - NBFC" to identify and develop novel non-invasive ornamental plants for future sustainable urban ornamental horticulture.

KEY WORDS: biodiversity, genetic transformation, in vitro culture, introduced species, molecular biology, ornamental crops, invasive plants.

"FLORA & GREEN" ARE THE NEW MUST IN COMMUNICATING YOUR SUSTAINABLE REPUTATION

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"*I get so happy when you talk to me*", … WHAT if you received this message from a plant?!
An increased number of academic studies demonstrate specific reactions of plant species to certain *stimuli*, notably chemicals, sound-based or photoreceptive. Even if defining all this as a communication process does not find common consensus on a scientific level, it leads us to reflect on the importance of creating an adequate *communication ecosystem* to apply whenever we come into contact with *flora*. Not by chance, agricultural companies breeding ornamental and flowering plants are rediscovering ancient values to foster sustainability or introducing innovation respectful of the natural rhythms according to the specific needs of each plant species. *Peter Seabrook*, paramount British gardening writer and television broadcaster, has focused for more than forty years on a clear communication, understandable by the large audience, even if based on scientific sources, to create a storytelling from the point of view of plants. One of his central ideas was in fact to start from the 'heritage' of each plant species to create the best conditions of cultivation, both in plant nurseries and at home. His legacy has influenced the new generations of professional gardeners. Notably, emerging entrepreneurs in the horticultural industry following his teachings are selecting plant species that require less maintenance and are suitable for local climatic conditions of the geographical areas where they are distributed. Thus, it triggers the *Triple Bottom Line* (TBL) business strategy investing in environmental, social and economic sustainability, breaking away from the fast fashion model. *But how to transmit effectively the corporate sustainable values and concrete actions of a breeding and gardening company?* We cannot escape the pragmatic and constitutive nature of eco-communication, conveyed through the support of experts in the field of *green marketing*. Each message follows a phased path from the sender to the receiver in which several variables are involved. Managing an appropriate business communication plan is essential to get to the point with the least risk of 'noise', to avoid *green washing*.
A case study conducted by *Stanford University* in 2016 shows how you can easily be misled by examining a photo of a plant species anchored to a specific news event. Reputation, trust, empathy, clarity, responsiveness, storytelling based on reliable and traceable sources are key factors to make a difference on the market and foster eco-responsibility in the target audience. This was clear for *Rachel Louise Carson*, American biologist, who gave birth to modern ecology and *environmental communication*. Maurizio Abbati's speech will offer an overview of the evolution of eco-communication from its birth to the present, with a look to the future through a targeted case study.

KEY WORDS: #communication #storytelling #sustainability #corporate #reputation

POSTER

LINARIA MILL. - A GENUS WITH ORNAMENTAL VALUE AND BREEDING POTENTIAL

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The genus *Linaria* Mill., which consists of more than 150 species and belongs to the family Plantaginaceae, is related to many commercially important ornamental plants such as *Antirrhinum majus* L., *Angelonia* Humb. et Bonpl., *Digitalis* L., *Globularia* L., *Penstemon* Schmidel, *Veronica* L. etc.

Due to the genus' diversity and its ornamental potential, a breeding program has started six years ago at the Humboldt University Berlin. Primary selections from seed sources (*Linaria aeruginea* (Gouan) Cav., *Linaria genistifolia* ssp. *dalmatica* (L.) Mill., *Linaria maroccana* Hook., *Linaria purpurea* (L.) Mill.) and clonal accessions collected in urban areas and in wild habitats (*Linaria vulgaris* Mill.) resulted in the genotypes later used as breeding partners. Cross-breeding was performed in a partial diallel cross system. All 23 combinations produced seeds but only seeds from 14 cross-combinations germinated. Several interspecific combinations showed symptoms of bastard bleaching in the F1 which could be a consequence of incompatibility between parents' genomes. A selected F1 from *L. vulgaris* × *L. purpurea* with intermediate flower coloration and growth pattern was successfully back-crossed. F1 and F2 selections show potential for cut flower plants as well as garden and pot plants. The cross-breeding between *L. vulgaris* and *L. genistifolia* shows scenting big yellow flower with an orange mark on the lower lip. Due to heterosis effects, this combination seems to have the highest potential for ornamental purposes since flower size appears as an important breeding goal in *Linaria*. According to the presented results, the genus *Linaria* can be added into the group of "New Ornamentals" with high horticultural potential. In addition, further breeding approaches such as polyploidization, additional interspecific hybridization, and strong selection will result in first cultivars in the near future.

KEY WORDS: *Linaria*, interspecific hybridization, heterosis.

MORPHOLOGICAL CHARACTERIZATION OF AMARYLLIS (*HIPPEASTRUM* HERBERT) GENOTYPES

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Due to their visual appeal, the *Hippeastrum* flowers are economically important and have been the goal of many breeding programs, in which the objective is to modify the characteristics and quality of flowers.

To explore the variability of *Hippeastrum* accessions and obtain plant genetic resources to be used in future breeding programs, in this study, the phenotypic traits from 64 *Hippeastrum* genotypes were evaluated using 39 morphological descriptors. Five main components were identified from the principal component analysis (PCA) carried out by morphological descriptors, which explained 70 % of the total variability.

The genotypes were associated in three groups based on Gower distances ranking from 0.18 to 0.49 and in according to the Ward's clustering method; these three groups explained 72 % of the total variability, and outstanding 12 highly discriminant quantitative characteristics. The three groups identified are distinguished for the flower main colours, the patterns of distribution in the tepals and also statistically differences respect to the length, height and width of the flower.

This is the evidence of the genetic variability of the accessions and their potential to be used in futures breeding programs and also the usefulness of the floral characteristics: length, height and width for discriminating between accessions and select the material.

KEY WORDS: ornamental, flower, principal component analysis, variability.

GENETIC DIVERSITY OF WILD POPULATION AND IDENTIFICATION OF UNCERTAIN CULTIVARS OF *HYDRANGEA SERRATA* BY SSR ANALYSIS

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Hydrangea serrata (Thunb.) Ser. distributed throughout mountainous areas in Japan and southern Korea. The inflorescence of *H. serrata* consists of decorative sterile flowers and non-decorative fertile flowers. The color of decorative flowers varies from white to blue and pale to vivid. Such as flower color, *H. serrata* has great morphological diversity. Thus, accessions that have higher ornamental values are selected from various wild *H. serrata* populations, and they are given a name and sold as cultivars. Many of them are named after their collected place and horticultural characteristics. However, these cultivar names are freely given by the person who collected them, and most of them are not applied for variety registration. Therefore, it is uncertain that their names are surely related to their collected places. In this study, we investigated microsatellite (SSR) profile of wild populations collected from various parts of *H. serrata* distribution range and its cultivars. This study aims to evaluate genetic diversity of *H. serrata* wild populations and to reveal uncertain named cultivars corresponding with their collected places.

KEY WORDS: Japanese hydrangea, cultivar name, microsatellite, structure analysis.

CONSERVATION, CHARACTERIZATION AND ENHANCEMENT OF ACCESSIONS AND SPECIES OF THE *SALVIA* GENUS

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The recovery and conservation of biodiversity are priority activities worldwide for the protection of species of agricultural and ornamental interest from the risk of genetic erosion and will be strategic activities in a future conditioned by significant climate and environmental changes. The CREA of Sanremo has since years been engaged in the study of aromatic plants both in the Mediterranean basin and in other parts of the world (in particular, Africa and the Americas) and has set up collections of numerous species and accessions of sage and rosemary (*Salvia rosmarinus*), highlighting the multifunctional aspect: ornamental, food and industrial (extracts for cosmetic and/or pharmaceutical or antiparasitic use). In recent years, species, varieties, and accessions of plants present in the collections have been maintained and renewed and specific descriptors were used for the morphological characterization thanks to which descriptive cards were prepared for many of the accessions of *S. rosmarinus* and *S. officinalis*. As part of various research projects, the use of flowers for consumption as edible flowers of various species and varieties of the genus Salvia was also evaluated because of the interest to offer these products as a garnish for dishes or as ingredients in sweet and savoury preparations. For this purpose, the species have been carefully selected, the methods of organic cultivation have been applied for their production. The results obtained have shown that within the rosemary and sage collections there are species, accessions and varietal selections with multifunctional characteristics usable not only for ornamental and extraction purposes, but also as plants suitable for the production of edible flowers. Many of the accessions considered adapt well to sunny, arid climates with significant temperature ranges and, having low nutritional and water requirements, their cultivation can be considered with low environmental impact.

KEY WORDS: flower description; selection; sage; rosmary; edible flowers.

MOLECULAR CHARACTERIZATION OF AMARILLYS (*HIPPEASTRUM* HERBERT)

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Hippeastrum ornamental plants, valued for floral esthetics, are the target of breeding programs that seek to modify their appearance and quality. These programs are based on the knowledge and description of the genetic variability of accessions through a molecular and morphological approaches. ISSR molecular markers are reproducible, polymorphic, and neutral. With the objective of knowing the genetic variability of *Hippeastrum* in the central part of Mexico and obtaining resources that can be used in future plant breeding programs, in this research 64 accessions were characterized to explore their genetic variability using 23 ISSR primers. 167 bands were obtained, and 88.02 % of them were polymorphic with an average of 7 bands per primer. Primers ISSR1, ISSR3 and ISSR6 showed PIC values of 0.35, 0.28 and 0.27, respectively. Jaccard distances ranged from 0.30-0.81, and according to the Ward´s method three groups were identified, nevertheless it is recommended to complement these results with morphological characterizations for selection of accessions when planning and designing breeding programs.

KEY WORDS: ISSR, molecular markers, clustering analysis, genetic variability.

DETERMINATION OF A PROTOCOL FOR THE IN VITRO GERMINATION OF *TILLANDSIA USNEOIDES*

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The excessive collection of *Tillandsia usneoides* (heno, paxtle or Spanish moss) and the loss of natural habitat are putting wild populations at risk and loss of traditions by being used even to perform dances (in Mexico). Hence, it seeks to establish some techniques for the legal production or conservation of viable germplasm. Therefore, it was sought to observe their behavior during in vitro germination to obtain innocuous explants for other experiments.

In the first tests, contamination was observed in 100 % of the establishments, so two bioassays were designed. 1. Completely randomized design with five treatments in the growth medium: a) 0.5 and 4 ml.L^{-1} of Plant Preservative Mixture® (PPM), b) 250 mg.L^{-1} and 4 g.L^{-1} of chitosan and a witness, all of them with 15 g.L^{-1} of sugar, MS (Murashige and Skoog) at 25 %, coal 2 g.L^{-1} and agar 5.5 g.L^{-1}.

With the results, a second bioassay designed as a completely randomized factorial was established: factor one, 250 mg.L^{-1} chitosan and 2.5 ml.L^{-1} of PPM, factor two, four disinfection techniques. No contamination was observed in medium one when combined with wash one, nor in medium two with wash four, however, twice the germination was observed in the first. Being the best treatment the medium where they were added 250 mg.L^{-1} of chitosan and washing consisted of soaking with 70 % alcohol (3 min) and 5% commercial chlorine (40 min). With this technique, it can be expected to germinate the greatest amount of hay for subsequent experiments which are useful for the multiplication and conservation of the species.

KEY WORDS: Bromeliaceae, conservation, use traditional, medicinal.

GENETIC IMPROVEMENT PROGRAM OF SOME ORNAMENTAL MEXICAN SPECIES AT AUTONOMOUS UNIVERSITY CHAPINGO

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Mexico is one of the 10 most megadiverse countries in the world, due to the diversity of its plant, animal and microbial species. It has around 25,000 species with ornamental potential, of which 2,500 are cultivated and less than 10 species, such as poinsettia, dahlia, marigol, cosmos, tigridia and echeveria, have a consolidated genetic improvement programme in countries such as Japan, Europe and the United States of America. In our country, government investment in this area has been minimal and there is no vision that ornamental plants can bring foreign exchange to the country in the future, as in Holland.

For all these reasons, in 2010 a Field Germplasm Bank was created on the Campus of the Autonomous University Chapingo. Its aim is to conserve, characterise, improve and use Mexican phylogenetic resources for ornamental purposes. This bank has 65 accessions of poinsettia (*Euphorbia pulcherrima*), 17 species of dahlia (*Dahlia* spp.), 5 species of cosmos (*Cosmos* spp.) and 15 accessions of *Euphorbia strigosa*.

During these years, improvement techniques such as the use of irradiation and hybridization have been applied and currently 13 varieties of poinsettia, 14 varieties of dahlia, 2 varieties of cosmos and 1 variety of *E. strigosa* have been registered. It is concluded that, in order to have a greater impact, varieties should be registered with breeders' rights and deliver them to the producers for their evaluation and commercial reproduction.

KEY WORDS: Biodiversity, breeder's rights, improvement techniques.

A NEW MINIATURE DWARF EGGPLANT WITH INTEREST AS POTENTIAL ORNAMENTAL PLANT

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Eggplant (*Solanum melongena* L.) is an important vegetable crop widely cultivated around the world due to its culinary and nutritional value. Despite its primary agricultural use, eggplant is also an aesthetically pleasing ornamental plant with considerable decorative value for gardens and landscaping.

With the emergence of climate change, the application of wild relatives in breeding programs is playing an important role for the development of new eggplant genotypes with enhanced resistance to biotic and abiotic stresses.

As a result of introgression breeding with the wild species *S. anguivi* Lam., new phenotypes appeared which showed increased resilience and an interest for ornamental use. After a second backcrossing between these offsprings and the hydrid eggplant, a miniature dwarf white-fruited plant was observed. This individual plant was selfed and, for several generations, selection was performed for the dwarf phenotype followed by selfing.

A stabilized miniature eggplant line was then selected. The new accession exhibits unique morphological and agronomic features like short internodes, small leaves and multiple inflorescences with purple flowers with white stripes. It is highly prolific and produces miniature (2-3 cm long) white fruits. It has early flowering and adapts well to growth in small pots (0.5-1.5 L). It flowers well both under a wide range of conditions including full exposure to sun or in shadowy places. Its unique characteristics make it as a potential ornamental crop for cultivation in pots, hydroponic or in soil for home gardens and landscaping.

KEY WORDS: dwarf eggplant, breeding, potential ornamental.

SEED AND EMBRYO CRYOPRESERVATION AS STRATEGY TO ORNAMENTAL PLANTS AND GENETIC RESOURCES CONSERVATION

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Ornamental market is eager for new products and tropical species have the potential supply trends from cut flowers to landscape design. However, as varieties are developed and new species with ornamental use are discovered, plant germplasm conservation arises as an important tool for plant breeding, as well as safeguarding genetic resources. Long term conservation can be achieved through cryopreservation techniques, storing plant material in very low temperatures. For cryopreservation success, it is important to avoid ice crystals formation inside cells, in order to maintain viability. This could be achieved with different explants and/or use of cryoprotection techniques. Seeds and also embryos naturally hold lower water content, compared to plant cells, therefore facilitating cryopreservation. Such character enables direct immersion in liquid nitrogen recovery and normal germination in species like *Hibiscus acetosella* Welw. ex Hiern., *H. sabdariffa* L. and *Handroanthus serratifolius* (Vahl) S.O.Grose. Also, simple dehydration with silica or laminar air flow could reduce water percentage in different species, such as *Strelitzia reginae* Banks, *Zinnia elegans* Jacq., *Xyris cipoensis L.B.Sm. & Down* and *Handroanthus serratifolius*, which vary in botanical aspects and ornamental usage. Hence, cryopreservation is an appealing technique to support ornamental plants in use or in potential germplasm conservation. Different species can be preserved through seed storage with assured recovery for germination and plant formation.

KEY WORDS: Micropropagation, plant preservation, explant cryoprotection.

Support: Capes, FAPEMIG and CNPq, Brazil.

SCREENING FOR *PHYTOPHTHORA* RESISTANCE IN *LAVANDULA* SPP.

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Lavender (*Lavandula* spp.) is a shrub with origins in the Mediterranean. Its popularity as an ornamental garden plant is increasing in the face of climate change due to its good drought tolerance. *Phytophthora* spp. are important pathogens in lavender, causing lethal root and stem rot. Breeding for *Phytophthora* resistance is desirable but requires a representative and efficient bioassay. In this study, we aimed to 1) select appropriate isolates for such a bioassay based on the genetic and pathogenic diversity of the pathogen population; and 2) apply the bioassay to a collection of lavender species and cultivars to screen for *Phytophthora* resistance. *Phytophthora* isolates were obtained from symptomatic lavender plants after a survey in Flemish nurseries and private gardens. Taxonomic classification and diversity analysis of the isolates were done using barcode sequencing and genotyping-by-sequencing (GBS). A total of 16 *Phytophthora* isolates were identified, comprising eight species and one hybrid species. Variation in pathogenicity among the isolates was determined using an optimized bioassay. Disease symptoms were monitored and scored using visual observation and final aboveground biomass of the inoculated plants was measured. The most pathogenic isolate, *P. nicotianae*,, was used to inoculate 64 lavender species and cultivars to screen them for *Phytophthora* resistance. Identification of variation in *Phytophthora* resistance in *Lavandula* could enhance breeding programs in facilitating the creation of resistant plants.

KEY WORDS: bioassay, breeding, disease resistance, GBS, lavender.

ANALYSIS OF SECONDARY METABOLOME AND CHINESE MEDICINAL COMPONENTS IN ROSEHIPS BASED ON WIDELY TARGETED METABOLOME

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Rosehip is an underutilized nutrient source, rich in a variety of bioactive substances, and has a long history of utilization in the diet and medicine. China is rich in *Rosa* resources, and metabolomics profiles of different rosehips are needed to explore the specific uses of underutilized rosehips.

Secondary metabolites of 12 rosehips in China were identified by UPLC-ESI-MS/MS, and a total of 523 metabolites were detected. Through the search of TCMSP database, 151 metabolites were found to be chemical components of Chinese medicine, and 74 metabolites were identified as KAI in five *Rosa multiflora* Thunb. fruits. Different rosehips have unique uses. For example, *R. persica* Michaut ex Juss. contains more abundant phenolic acids, while *R. roxburghii* Tratt. contains more abundant terpenoids.

This study enriched the metabolic pool knowledge in rosehips and provided a reference for the comprehensive development and utilization of fruit food and medicine of *Rosa* species.

KEY WORDS: Rosa, Hip Metabolome.

COMPACT HARDY *GERANIUM* BY USE OF *AGROBACTERIUM RHIZOGENES*

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The growth habit of the plant is an important factor affecting the quality of the plant and market demand. Hardy geranium is a popular garden plant. Despite its wide variety, there is a need for compact varieties, which was the main motivation of this study. Leaf explants of five genotypes (*Geranium* 'Rozanne', *G. macrorrhizum* L. 'Czakor', *G.* x *cantabrigiense* P.F.Yeo 'Biokovo', *G. pratense* L. 'Galactic' and 'Blushing Turtle') were co-cultivated with wild type *Agrobacterium rhizogenes* (Riker et al.) Conn (ATCC15834) (= *Rhizobium r.* Riker et al.). All genotypes produced hairy roots, but transformed plants could only be regenerated from the hairy roots of 'Blushing Turtle'. qPCR confirmed that seven Ri lines contained Ri genes in different copy numbers. After acclimatization, petiole length, number of leaves and branching (number of axillary shoots) were measured each month. For all traits, clear differences were seen between the Ri lines and control plants.

KEY WORDS: Hardy geranium, Compactness, *Agrobacterium rhizogenes*, *Geranium.*

IDENTIFICATION OF *BOTRYTIS CINEREA* TOLERANT PLANTS OF *HYDRANGEA MACROPHYLLA*

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Botrytis blight caused by the necrotrophic fungus *Botrytis cinerea* Pers. is one of the most destructive diseases in *Hydrangea macrophylla* (Thunb.) Ser. Symptoms appear on the plant's leaves, stems, flowers and buds. Fungicide applications are increasingly impractical due to the development of fungal resistances. Therefore, usage of disease-resistant cultivars is considered as the most sustainable approach to manage Botrytis blight. For this aim, we established a resistance test based on 11 cultivars of *H. macrophylla*. This resistance test was conducted as a detached leaf assay based on mycelial plug inoculation using the highlyaggressive *B. cinerea* isolate BcHyd21. The lesion areas were measured at 3- and 5-days post inoculation (dpi) to determine the speed of infection and fungal development. With this test, we identified reliably the most tolerant and susceptible cultivars. An independent *in vivo* infection test confirmed these results, which indicates the robustness and transferability of our test system. Next, we screened 75 *H. macrophylla* cultivars for *Botrytis* tolerance. The size of lesion areas differed clearly between cultivars and allowed the identification of highly tolerant and susceptible cultivars to select crossing partners. To produce segregating F_1 populations for QTL analysis, different crosses were made between cultivars showing different levels of tolerance/susceptibility. Independent F_1 populations derived from crosses between medium susceptible cultivars and between highly and medium susceptible cultivars were phenotyped at 3 different time points. Each of these populations segregated for tolerance against *B. cinerea*. Preliminary data indicated that F_1 individuals derived from a cross between medium susceptible parents show a lower variation, because these individuals were more tolerant than individuals derived from highly susceptible parents. These first results suggest that tolerance against *B. cinerea* is inherited in *H. macrophylla* and breeding of resistant plants is possible.

KEY WORDS: *Hydrangea macrophylla*, *Botrytis cinerea*, tolerant.

EVALUATION OF PH TOLERANCE IN *RHODODENDRON* GENOTYPES

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Rhododendron is a popular garden plant, but its use is limited because it prefers acidic soils. Breeding and selection for *Rhododendron* cultivars able to grow on neutral to alkaline soils will broaden the use of this species.

We evaluated *Rhododendron* genotypes for pH tolerance in a greenhouse experiment with potted plants. For this study, we used different genotypes: the established cultivars 'Gomer Waterer' and 'Cunningham's White', seedlings from *R. fortunei* Lindl., and hybrids of a cross between 'Pink Purple Dream' and 'Belami'. Rooted cuttings of each genotype were planted either in a substrate with low pH (pH 4.5) or elevated pH (pH 6.3). To evaluate growth and pH stress different parameters were measured: chlorophyll fluorescence by Fv/Fm, fresh weight, dry weight, a rooting score after 70 and 140 days of growth, plant height, number of shoots, number of growth flushes, length of the new flush and leaf color analysis by CIELab.

Except for the number of shoots, all other parameters were able to distinguish stressed and non-stressed plants. Fresh weight and root scores had a very strong correlation ($r = 0.96$) showing that plants with stress were affected in both their above and belowground size. A good correlation was found between Fv/Fm and the CIELab measurements with a negative correlation between Fv/Fm and L* ($r = -0.75$) and b* ($r = -0.76$), and a positive correlation between Fv/Fm and a* ($r = 0.71$), as plants grown on higher pH had a paler leaf color. The analysis showed variation in pH stress tolerance between the genotypes tested. Especially one of the 'Pink Purple Dream' x 'Belami' genotypes showed a remarkable tolerance to the high pH in the substrate, which indicates that breeding for pH-tolerant genotypes is possible.

KEY WORDS: abiotic stress, chlorophyll fluorescence, CIELab, pH resistance, stress tolerance.

WOODY ORNAMENTAL BREEDING AND SELECTION ASSISTED BY UAV-IMAGERY

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With regard to woody ornamental breeding interspecific hybridization, polyploidisation and mutagenesis are very important tools to increase genetic variation and are at the basis of many modern assortments.
At ILVO, Belgium, these tools are exploited in the breeding program of different genera: *Sarcococca, Lavandula, Photinia*, and others. Selection within offspring populations require evaluation of a wide variety of traits, including aesthetic beauty and tolerance to (a)biotic stresses. This is a time consuming and laborious process. Methodologies based on data derived from visual imagery captured with a drone equipped with an RGB (red, green, blue) camera might facilitate selection and ranking of genotypes.
Using our *Sarcococca* selection program as an example, we will demonstrate how drones (unmanned aerial vehicles, UAV) and high throughput field phenotyping (HTFP) can be applied. Data was gathered in 2019 in two well-established breeding trials and plant architecture traits, such as plant height and shape, were determined to evaluate individual plants. Also, the variability among open-pollinated half-sib populations of Sarcococca was analysed per mother plant. This approach allows for the use of quantitative evaluation standards that exclude breeder bias and speed up data collection. Correlations were calculated between on-ground measurements and UAV-derived methods and were significant. The advantages and disadvantages of the methodology and the approach used, will be presented.

KEY WORDS: High throughput field phenotyping, drone.

EVALUATION OF THE EFFECT OF NITROUS OXIDE ON THE PLOIDY LEVEL OF LISIANTHUS (*EUSTOMA RUSSELLIANUM*)

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Lisianthus [*Eustoma russellianum* (Hook.) G.Don = *Eustoma grandiflorum* (Raf.) Shinners] is an ornamental species that has gained relevance in the last decade. Originally from northern Mexico and southern United States, it is a new floral crop that has become one of the main cut flowers, where larger flowers are coveted. Polyploidization consists of doubling the chromosome number of a species, obtaining larger plants, it exists naturally and can be induced by chemical agents, colchicine being the most widely used, however, the success rate is low. Nitrous oxide, a gas, acts in a similar way to colchicine and has been used for induction of 2n gametes, but not in somatic cells.

The objective of this work is to know the effect of nitrous oxide under different times and pressures on the ploidy level of *E. russellianum*. Seeds of *E. russellianum* were disinfected and germinated in ½MS medium. Once the cotyledons emerged, they were treated at 5 and 6 atm in a pressure chamber for 24, 48, and 72 hours, with 50 seedlings per treatment. Also, 3 colchicine treatments were carried out, subjecting 50 seeds per treatment to 0.1% and 0.2% during 2, 4 and 6 hours. The treated seedlings were transferred ½MS medium added with Benzyladenine (BA) and gibberellic acid (GA3) for their growth, the survival rate and the size of the plants were recorded. Subsequently, they were transferred to ½MS medium with casein hydrolyzate and indolebutyric acid (IBA) to induce roots.

The highest survival and mortality in the N2O treatments were 6 atm at 72 hours and 5 atm at 72 hours respectively, while in the colchicine treatments they were 0.1% for 2 hours and 0.2% for 6 hours. The size of the plants obtained were registered, among which there are approximately 190 with sizes that double or triple the size of the control plants. Currently there are approximately 3000 multiplied plants and both, chromosome counts and DNA content are under development.

KEY WORDS: Lisianthus, polyploid, nitrous oxide, colchicine, chromosomes.

GENETICALLY MODIFIED ORNAMENTALS (ARDISIA, PETUNIA, AND ROSE) DEVELOPED IN NATIONAL INSTITUTE OF HORTICULTURAL AND HERBAL SCIENCE OF KOREA

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The National Institute of Horticultural & Herbal Science (NIHHS) started studies to develop new rose, petunia, and ardisia in 2003, 2004, and 2006, respectively, using genetic transformation technique. We have reported the research results.

In rose, plant regeneration technique through somatic embryogenesis, which is a prerequisite to develop this genetic transformation technique, was successfully done in 2007 (Lee et al. 2008. J. Plant Biotech. 35:223-228; Lee et al. 2010. Acta Hort. 870:219-215). And then, in 2010, genetic transformation technique, which is not easy to develop in rose, was developed (Lee et al. 2010. J. Plant Biotech. 37:511-516; Lee et al. 2013. Hort. Environ. Biotechnol. 54:172-176). In 2015, we obtained 7 *SOD2*-transgenic rose plants enhanced in resistance to drought Lee et al. 2020 Hort. Environ. Biotechnol. 61:569-576). In 2017, we obtained 8 *COPB2*- transgenic rose plants preventing scattering of *Tetranychus urticae* C.L. Koch, 1836 (Lee et al. 2022. Scientia Horticulturae 301: 111113). In 2020, we obtained color-modified two *VtF3'5'H/RhNHX*-transgenic rose lines (changed white into pink-purple).

Moreover, we developed *SOD2*- or *NDPK2*-transgenic petunia lines resistant to abiotic stresses (Lee et al. 2009, J. Plant Biotech. 36:144-148, 289-293; Lee et al. 2010, J. Plant Biotech. 37:562-566; Lee, et al. 2010, J. Plant Biotech. 38: 215-220), *NDPK2/SOD2*-transgenic petunia lines resistant to sulphur dioxide gas (Lee et al. 2016. Korean J. Hortic. Sci. Technol. 34:154-162), and *AtFALDH*-transgenic petunia line enhanced in ability to detoxify formaldehyde gas (Lee et al. 2015 Hort. Environ. Biotechnol. 56:247-254).

Furthermore, we succeeded in setting up the optimal condition for shoot regeneration from internodes of ardisia (*Ardisia pusilla* A. DC.) in 2006 (Lee et al. 2008, J. Plant Biotech. 35:209-213). Recently, NIHHS developed *AtNDPK2*- or *CYP2E1*-transgenic ardisia plants enhanced in removing exogenous toluene gas (Ahn et al. 2020. Hort. Environ. Biotechnol. 61:949-957; Kim et al. 2021. Hort. Environ. Biotechnol. 62:619-627).

KEY WORDS: Genetically modified ornamentals.

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EVALUATION OF THE EFFECT OF NITROUS OXIDE (N2O) UNDER PRESSURE ON THE PLOIDY OF *TAGETES ERECTA*

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Polyploidization is a relevant strategy in plant breeding with benefits as the increase in plant size, leaves or fruits. To date, colchicine remains the most widely used antimitotic agent, but it has drawbacks that make it ineffective; It can induce aneuploidy or chromosomal rearrangements, it has a high toxicity for animal and plant cells. As a relevant alternative, we have used nitrous oxide, since it has been shown to work as an inhibitor of the mitotic spindle, without the toxicity of colchicine.

This work aims to know the effect of nitrous oxide in the induction of *Tagetes erecta* L. polyploidy and demonstrate its efficacy against colchicine. Seeds germinated in vitro in MS medium were used. At the beginning of germination different treatments were applied; the seeds were subjected to two concentrations of colchicine (0.1%, 0.2%) for different times (2, 4 and 6 hours) with 13 seeds per treatment. Another treatment consisted of exposing the seeds in the same germination stage to nitrous oxide at 6 atm pressure for 24, 48 and 72 hours with 30 seeds per treatment. After the treatments, the seedlings were transferred to petri dishes for an observation period where the mortality rate of the plant material after contact with the polyploidizing agent was measured. The live plants were taken to a stage of growth, propagation, acclimatization and sowing in a greenhouse to obtain roots that served for the chromosome count analyses.

Nitrous oxide had no mortality in any of its treatments, while colchicine had a mortality of 68.5% for the treatment with the least toxic effect. Polyploid cells have been observed in nitrous oxide treatments; some of the plants in the greenhouse, resulting from the treatments with the polyploidizing agents, present typical morphology of polyploid plants, such as larger flowers, leaves, and stems.

KEY WORDS: Colchicine, Ornamental, Polyploidization.

RATING *EUCALYPTUS* SPECIES FOR THEIR ADAPTABILITY AS CUT FOLIAGE PRODUCTION IN LIGURIA

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The Ligurian floriculture industry has a 150-year tradition and nowadays shows a saleable production estimated to be 400 million of euros, being the export nearly 75%. Cut foliage has always been an economic contributor to this floriculture industry since cut foliage species are grown as perennial crops in open field and with low costs of production. Foliage plants are relatively easy-to-grow with a less extensive pest and fertigation management programs compared to the most important flower species. Far from being a simple green background for flowers, cut foliages are increasing their importance as component of flower arrangements. In addition, cut foliages are important in our Region since thanks to these crops it is possible to guarantee an all-year-round production and hence the presence of our growers in the international markets. Therefore, cut foliage crops fulfil the criteria of sustainable cultivation both from an economic and an environmental point of view; the cut foliage crop management can be carried out in accordance with the European and National Strategic Programmes, imposing severe restrictions for the use of synthetic chemical substances and pesticides harmful to the environment and to the human health. The genus *Eucalyptus* (family: Myrtaceae) includes numerous species, mainly used in the wood and cellulose industry, for the production of essential oils but also for ornamental purposes. Several species of *Eucalyptus* have shown a considerable potential for providing a good complement in flower arragements, particularly when the cut foliage at a juvenile stage of development is used.

At Regional Institute for Floriculture, Sanremo, a study was undertaken to select several genotypes suitable for the Mediterranean and Western Liguria conditions. In cooperation with growers and exporters/retailers, we rated several *Eucalyptus* species for their ornamental value and the adaptability to the cut foliage production. Due to the high variability scored among seedlings, the vegetative propagation by cuttings was undertaken in order to get a homogeneous production of the superior genotypes. In details, specific propagation protocols were pointed out for the selected genotypes of the following species: *E.gunnii* Hook f., *E. cinerea* F.Muell. ex Benth., *E. parvula* L.A.S.Johnson & K.D.Hill, *E. populnea* F. Muell., *E. albida* Maiden & Blakely, *E. kruseana* F.Muell., *E. crucis* Maiden. Agronomic trials are underway on the application of biostimulants during both the rooting process and the cultivation of young plantlets. The work will present a qualitative evaluation of the selected species with regard to: ornamental characteristics, adaptability to Mediterranean conditions, preliminary indications on the aptitude for vegetative propagation by cuttings and results on the use of biostimulants.

KEYWORDS: *Eucalyptus*, sustainable cultivation, vegetative propagation, cuttings, biostimulants.

CYTOGENETIC COMPARISON OF *HIBISCUS SYRIACUS* WITH *HIBISCUS MOSCHEUTOS* BY FISH AND FLOWCYTOMETRY

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Determination of chromosome number, nuclear DNA content, genome size, and ploidy level and information on cytogenetic characteristics are all prerequisite of modern plant breeding. The high number, small size, and similar morphology of mitotic chromosomes in *Hibiscus* species make it challenging to identify individual chromosomes. The goal of the study was to ascertain the chromosome number, number of 5S and 18S rDNA signals, as well as genome sizes, 2C-DNA content of *Hibiscus syriacus* L. and *Hibiscus moscheutos* L. Two cultivars from each species 'Shintaeyang' and 'Blue Bird' for H. syriacus and 'Luna Red' and 'Luna Pink Swirl' for *H. moscheutos* were selected for analyses. The root tip squash method was used to check the chromosomal numbers. The number of chromosomes were 84 for *H. syriacus* and 38 for *H. moscheutos*. 5S rDNA and 18S rDNA signals were detected by fluorescence in situ hybridization (FISH). FISH results, *H. syriacus* possesses two signals for 5S rDNA and four signals for 18S 18S rDNA, while *H. moscheutos* showed two loci for 5S rDNA and six loci for 18S rDNA. The lengths of mitotic metaphase chromosomes ranged from 2.0 to 7.4 µm and 2.4 to 7.1 µm for *H. syriacus* 'Shintaeyang' and 'Blue Bird' respectively, and 2.9 to 5.2 µm and 2.2 to 5.5 µm for *H. moscheutos* 'Luna White' and 'Luna Pink Swirl', accordingly. The 2C-DNA content of *H. syriacus* was 4.0 and 4.2 pg, for 'Blue Bird' and 'Shintaeyang' respectively whereas, *H. moscheutos* had nearly half and that amount was 2.1 and 2.0 pg for 'Luna Red' and 'Luna Pink Swirl' respectively.

KEY WORDS: *Hibiscus*, Cytogenetics, Fluorescence in situ hybridization, 5S rDNA, 18S rDNA, Nuclear DNA content.

This work was supported by Korea Institute of Planning and Evaluation for Technology in Food, Agriculture and Forestry (iPET) through Technology Commercialization Support Program, funded by Ministry of Agriculture, Food and Rural Affairs (MAFRA) (821005-3).

APPLICATION OF K-SEQ GENOTYPING PROTOCOL IN *RANUNCULUS ASIATICUS* L. AND *ANEMONE CORONARIA* L.

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Persian buttercup (*Ranunculus asiaticus* L.) and poppy anemone (*Anemone coronaria* L.) are ornamental, outcrossing, perennial species, belonging to the Ranunculaceae family, characterized by large and highly repetitive genomes. Reduced-representation sequencing (RRS) techniques are an advantageous approach to generate genome-wide high-throughput sequencing data obtaining a large number of genetic polymorphisms.

Here we applied K-seq protocol in both species to generate high-throughput sequencing data and produce a large number of genetic polymorphisms. The technique entails the application of Klenow polymerase-based PCR using short primers designed by analysing k-mer sets in the genome sequence. Since to date the genome sequence of both species has not yet been released, we designed primer sets using the genome sequence of the close-related species *Aquilegia oxysepala* var. *kansuensis* (Brühl) as reference. A set of 11,542 SNPs were applied for assessing genetic diversity of eighteen commercial varieties of *R. asiaticus*, while 1,752 SNPs were used for assessing genetic diversity in six cultivars of *A. coronaria*. No previous fingerprinting information were available for Persian buttercup for comparison purposes, while the results obtained in poppy anemone were compared with a previously published SSR-based fingerprinting, proving K-seq to be an efficient protocol for the genotyping of complex genetic backgrounds. Our findings make it possible to optimize genotyping protocols in such a complex species, potentially lowering wet-lab and sequencing costs for wide genotyping projects.

KEY WORDS: K-seq; genotyping; ornamentals; fingerprinting; NGS.

MICROSATELLITE-BASED IDENTIFICATION OF DI-HAPLOID PLANTS BY ANDROGENESIS IN *ANEMONE CORONARIA* L.

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Anemone coronaria L. is a perennially, allogamous, and highly heterozygous plant belonging to the family of Ranunculaceae, marketed both as cut flower and as garden plant. Its marked inbreeding depression following selfing hampers the development of highly homozygous inbred lines crossable for the production of F1 hybrids. Commercial cultivars are thus developed by selecting traits of interest in segregating genotypes obtained by crossing heterozygous genotypes, which are then propagated and commercialized through rhizomes. In the species, successful protocols for the obtainment of androgenetic haploid plants have been developed, but both flow-cytometric analyses and chromosome counts in meristematic cells of root apexes highlighted the high frequency obtainment of diploid plants. Recently, microsatellites (SSR) markers have been developed from the draft genome of the cultivar 'Magenta Mistral' and two of the, originating heterozygous alleles in the plant from which the anthers were taken, were selected. Their application in 'in vitro' seedlings made it possible the quick and easy discrimination between diploid plants resulting from spontaneous diploidization of an haploid embryo (one allele) in respect to the ones originated from somatic embryogenesis of anther wall cells (two alleles).

KEYWORDS: poppy anemone; androgenetic haploid plants; SSRs; genome sequencing.

APPROACHES TO UNDERSTANDING BUD OPENING IN LILIES

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Bud opening is an important feature for purchasers of cut flowers and a failure of buds to open leads to waste, reducing sustainability of the ornamental flower industry. Lilies are widely sold as a cut flower, grown both in Europe but increasingly, also, transported long distances. To delay flower development and opening during storage and transport, the flower stems are subjected to low temperature. To encourage bud opening the vase water is supplemented with sugars. However, in Oriental lilies that produce several buds per stem the youngest buds may fail to open when subjected to commercial treatment. Understanding the mechanisms that prevent bud opening in these youngest buds would help identify breeding targets that may be useful for producing more consistent bud opening for consumers. Transcriptomics combined with physiological experiments offers a powerful tool analysing processes occurring in buds at risk of bud opening failure. Here we assessed whether bud opening is directly affected by position on the stem, and the effects of commercial treatment, finding that indeed both affect bud opening. Metabolomic and transcriptomic approaches were also used to investigate flower opening indicating that important changes in both metabolites and gene expression are activated during the opening phase. This opens the way to future studies to identify specific gene targets.

KEY WORDS: flower development, Lilium, transcriptome, metabolites.

MICROSCOPIC ANALYSIS OF THE STEM SURFACE AND COMPARISON OF EXPRESSION OF MYB FAMILY TRANSCRIPTION FACTORS ACCORDING TO PRICKLE FORMATION STAGES IN ROSE 'PINK BEAUTY'

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Rose (*Rosa hybrida*) is one of the major cut flowers in Korea and is a crop with high economic value in the flower industry. The prickles of rose are very important to defend itself against herbivores or pests, but large and numerous prickles injure workers during cultivation or damage flowers during transportation, resulting in reduced marketability. In addition, wounds on stems caused by removing prickles cause a decrease in the longevity of cut rose flowers. So, rose varieties with few or no prickles are highly preferred, and breeders are trying to breed roses without prickles. However, some morphological mutants show regrowth of prickles after exposure to environmental stressors. Therefore, it is necessary to study the mechanism of prickles formation in order to develop non-prickly rose varieties and stably maintain their characteristics. In this study, the stem surface of the rose cultivar 'Pink Beauty' was confirmed using a scanning electron microscope and a stereo microscope. In addition, RT-PCR was used to confirm the difference in the expression of genes involved in formation according to the prickle morphology. For candidate gene selection, stems with/without spines were selected, and after RNA sequencing, the difference in expression was classified as up or down. And, among the groups with up-expression in the stem with prickles, the MBW complex, which has been reported to be involved in formation and development of trichome in Arabidopsis thaliana, was searched. RNA was extracted from stems with/without prickles, and RT-PCR was performed with primers targeting 10 candidate genes. As a result, it was confirmed that the expression level of 10 candidate genes was higher in the prickly stem than in the non-prickle stem. And, as a result of qRT-PCR by dividing the development of prickles into 4 stages, it was confirmed that the expression of three transcription factors, including MYB5, increased as the prickle developed. These results can be used as basic data for transformation experiments for the development of prickless roses. In addition, further research is needed to clarify the functions of transcription factors involved in prickle formation.

KEY WORDS: Microscope analysis, MYB family transcription factors, prickle formation, rose.

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HOW MUCH IS ENOUGH? CONSIDERATIONS ON THE USE OF MICROSATELLITE FOR GENOTYPING ORNAMENTAL PLANTS

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Genotyping through molecular markers can significantly speed up breeding programs, not only for Plant Variety Protection, but also for crosses and selection management. The progressive collapse of sequencing costs made HTS (High-Throughput Sequencing) platforms accessible also for marker assisted breeding programs. However, these costs continue to be a major obstacle for minor species, including ornamental ones. For this reason, the use of more cost-effective markers, such as Simple Sequence Repeats (SSRs), can still represent the most convenient choice. The discriminating potential of a single SSR is generally much higher than that of a Single Nucleotide Polymorphism (SNP), but the debate related to the minimum number of markers to be used for correctly discriminating breeding lines, is still ongoing. Here, we present two comparable case studies dealing with the use of SSR to genotype ornamental breeding lines, belonging to *Petunia hybrida* (2n=2x=14, 2C DNA content: 3,24 pg) and *Lantana camar*a (2n=2x=22, 2C DNA content: 3,0 pg). Preliminarily, a flow cytometry analysis has been carried out in both species, since the level of ploidy may strongly influence the success of a genotyping analysis. The *Petunia* genotypes did not show any difference in terms of ploidy, whereas in *Lantana* two different levels of ploidy were observed, allowing to discriminate diploid accessions from tetraploid one. Furthermore, the genome size estimates of both species were found to be consistent with the values available from the scientific literature.
For the subsequent SSR analyses, only diploid genotypes from both species were considered.

KEY WORDS: *Petunia hybrida*, *Lantana camara*, SSR markers, flow cytometry analysis, genotyping.

USING SELECTED HABITAT EUROPEAN DIRECTIVE SPECIES AS GARDEN PLANTS: CHALLENGES AND OPPORTUNITIES

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The Life+ SEEDFORCE project aims at improving the conservation status of 29 Annex II Habitat European Directive plant species, reported in bad conditions according to the 2013-2018 report on the trends of habitats and species according to art. 17 of the Directive. The project will identify and remove threats to the survival of these species in their native range, will assess their the genetic makeup, model their ecological niche and massively propagate them to bridge the isolation and re-establish gene flow between isolated population. To raise awareness of these species and to engage the public in their active conservation a selection of this species with ornamental and/or garden value will be tested for performance in suitable garden setting, including dry Mediterranean gardens [*Cytisus aeolicus* Guss., *Primula palinuri* Petagna, *Valeriana amazonum* (Fridl. & A.Raynal) Christenh. & Byng, *Dracocephalum austriacum* L., *Eryngium alpinum* L.] and wet boggy gardens [*Woodwardia radicans* (L.) Sm., *Adenophora liliifolia* (L.) A.DC., *Kosteletzkya pentacarpos* (L.) Ledeb.]. The project can therefore represent an example of a bridge linking the conservation of natural biodiversity and the production sector of ornamental plants.

KEY WORDS: Habitat European Directive, threatened plant species, ornamentals.

This work was carried out with the support of the Life+ SEEDFORCE.

ROLE OF BOTANIC GARDENS FOR ORNAMENTAL PLANTS CONSERVATION THROUGH SUSTAINABLE MANAGEMENT: CASE STUDIES AT HANBURY BOTANIC GARDENS

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Botanic Gardens have been collecting ornamental plants from all over the world for years; the collections represent scientific, historical, biological and genetic diversity assets. Recent years have seen an increase in parasitic infestations on many plants collections, as a result of both global change and greater international trade, which aid to the spread of several alien species, most of which are pests of ornamental plants. In order to preserve plants acclimatized in these places over time and to safeguard environment and health of gardeners and visitors from the widespread use of synthetic substances, the management of these parasites using sustainable methods becomes a priority.

The Hanbury Botanic Gardens are home to important ornamental plants collections; among the most significant, distinguished by high diversity, are those of the genus *Agave*, *Aloe*, and *Citrus*, while of historical-cultural value, the palms. However, several pests currently threaten their conservation. *Scyphophorus acupunctatus* (Gyllenhal, 1838), the agave black weevil, causing rot and death of the host plants (Agavoideae), is monitored with the use of pheromone traps and to limit its diffusion the eradication and rehabilitation of infested plants and low impact treatments are carried out. *Aceria aloinis* (Keifer) eriophyid mite causes physiological and morphological alterations of the aloe species, is controlled with the release of other predatory mites such as *Neoseiulus californicus* (McGregor, 1954) (= *Amblyseius c.)* and *Amblyseius swirskii* Athias-Henriot, 1962, as well as different citrus scales, which involve a subtraction of sap, are controlled releasing predators and parasitoids. The two palm parasites, the red weevil *Rhynchophorus ferrugineus* (Olivier, 1790) and the moth *Paysandisia archon* (Burmeister, 1880), are controlled using entomopathogenic products based on *Beauveria bassiana* (Bals.-Criv.) Vuill., 1912 and nematodes *Steinernema carpocapsae* (Weiser, 1955). As a result, the fight against these enemies is conducted through monitoring, agronomic techniques, and the employment of antagonistic organisms. These management strategies are effective sustainable control mechanism against natural parasites, for the conservation and defence of ornamental plants. These tools avoid use of dangerous chemicals products and are potentially applicable both in urban greenery and in private gardens.

KEY WORDS: sustainable management, pests, conservation, ornamental plants.

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SUCCULENT PLANT DIVERSITY AS A BOOST FOR A SUSTAINABLE LIGURIAN FLORICULTURE

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Succulents are drought-tolerant plants increasingly recognized as a resource to mitigate the consequences of climate change. Succulent plants offer many options for growers to provide low-maintenance plants that work well for green infrastructure, green roofs, and other eco-friendly projects thanks to their durability and flexibility. This added value could offer new possibilities for growers to increase plant sales. Liguria Region has a strong tradition of growing cacti and succulents. Compared to other production realities in the world, our growers are used to having a wide collection of cacti and succulents which is sometimes reaching more than 100 species for each farm. The production is addressed to several market channels and international trade is important. For this reason, it becomes mandatory to have innovative products to launch into the market in order to satisfy the consumers' needs and to support our niche market. The vegetative propagation of succulent plants is carried out by cutting or by plant division. In vitro propagation is a valuable mean thanks to the fast introduction of micro-plantlets into the market, the obtention of healthy plants to be used in the in vivo stock mother plants, and to the possibility to perform an in vitro conservation of valuable genotypes. In this framework, we would like to highlight the work which has been carried out at the Regional Institute for Floriculture (IRF), Sanremo, with the aim to give support to the local growers and to provide them with micropropagated plant material of several species of succulents and cacti. In particular, we worked on the following families and genders: Asphodelaceae (gender: *Aloe, Gasteria, Haworthia*), Asparagaceae (gender: *Agave*), Asteraceae (gender: *Senecio*), Cactaceae (gender: *Mammillaria, Maihueniopsis, Opuntia, Tephrocactus*) and Crassulaceae (gender: *Crassula, Echeveria*). In this work, the best-performing micropropagation protocols for each gender will be provided beside the average multiplication rate. Generally speaking, our approach was characterized by using Murashige and Skoog-based media with a low concentration of growth regulators (kinetin-Kin or 6-Belzylaminopurine- BAP at 0-2 mg L-1). The rooting phase was carried out under in vivo conditions.

KEY WORDS: Succulent plant, cactus, micropropagation, drought-tolerant, Liguria floriculture, plant diversity.

METABOLITE PRODUCTION FROM HAIRY ROOT BIOMASS IN *SALVIA* SPP.

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Salvia is the largest genus in the Lamiaceae family, with around 1000 species of shrubs, perennials, and annuals. The current research sought to cultivate the hairy roots of various *Salvia* species to enhance the production of secondary metabolites with pharmacological activity. Because of their rapid growth and genetic and metabolic stability, transformed roots are prospective biotechnological systems that produce significant amounts of secondary metabolites. Hairy root lines of *Salvia karwinskii* Benth. and *Salvia oxyphora* Briq. were established by co-cultivating the explants with a suspension of two strains of *Agrobacterium rhizogenes* (Riker et al.) Conn (ATCC and LB9402). The formation of hairy roots was observed 3-4 weeks following inoculation in both nodal and leaf explants under dark conditions. When compared to the ATCC strain, the LB9402 strain grown on Yeast/Mannitol/Broth (YMB) medium supplemented with acetosyringone developed noticeably more roots on each explant. On Murashige and Skoog media devoid of phytohormones, these roots demonstrated robust development and an abundance of lateral branching. In order to completely eradicate the bacterium, cefotaxime was added in decreasing amounts from 300 µg/l to 50 µg/l. We continued the research using various clones to establish biomass for the examination of secondary metabolites (diterpenoids, triterpenoids and phenolic acids) from the developed roots. The polymerase chain reaction was used to validate the transformation through examination of the *virC1* and *rolC* genes.

KEY WORDS: *Salvia*, Hairy roots, *Agrobacterium rhizogenes*, Lamiaceae.

MORPHOLOGICAL CHARACTERISTICS OF INFLORESCENCE AND TRUNK IN GOLDEN-RAIN TREES (*KOELREUTERIA PANICULATA* LAXM.) SELECTED FOR THE DEVELOPMENT OF NOVEL STREET TREES FROM SIX REGIONS OF KOREA

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Golden-rain tree (*Koelreuteria paniculata* Laxm.) indigenous in Korea, Japan and China has a high ornamental value owing to its yellow flowers that bloom in summer season. Moreover, its tolerance to air pollution, salt, and dry soil make it have a lot of potential not only as a landscape tree but also as a potential street tree in urban environment.

This study was conducted to collect basic data on tree form and infloresence characteristics of the golden-rain trees growing in the six regions including Taean, Yeongkwang, Wando, Pohang, Andong and Jecheon from late June to mid July 2022. We investigated the length and width of inflorescence (LI and WI, respectively), the number of flowers per inflorescence (NFI), and the presence of bract and red color on petal bases. Additionally, tree from, clear-bole length, shape and straightness grade of trunk were also examined. A total of 45 individuals were investigated, averaging 7.5±1.8 trees per region. Superior individuals were selected by the number of inflorescences, however, there was no significant difference in the size of inflorescences.

NFI was higher in the coastal regions, for example, YeongKwang (1223.2±734.5), Wando (958.8±450.6), Taean (985.3±530.5) and Pohang(714.8±219.4) than it in other regions. The individuals selected in Andong had the smallest LI (20.0±10.9cm), WI (17.8±10.4cm) and NFI (555.93±87.7). Most individuals of all regions have bracts (97.1%) and red color in petal base (86.5%). Tree form accounted for 77.8% of irregular form and 22.2% of round form. In most areas except Taean, main stems of the trees were straight with a gentle curve, however, the individuals from Taean had seriously wavy stem and short clear-bole length.

KEY WORDS: golden-rain tree, inflorescence, tree form, street trees, selection.

DAHLIA PARVIBRACTEATA FOR AGRICULTURE AND FLORICULTURE IN MEXICO

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Dahlia is the national flower of Mexico whose beauty is recognized worldwide. *Dahlia parvibracteata* Saar & P.D.Sørensen was discovered in 1995 north of Taxco in the state of Guerrero, Mexico. Under cultivation this species has a life cycle of 150 days from sowing to blooming, has tuberous roots and develops up to 4 basal long stems up to 180 cm, which are hollow, reddish brown with abundant flowers, ideal for cut flower. Plants produce up to 14 tuberous roots, with an average weight of 170 g per plant, resulting in a yield of more than 3.5 tons/ha of tuberous roots in a density of 20,750 plants. Tubers has an inulin concentration of 24.25% (dw) an important source for food, medicine and industry applications.

D. parvibracteata species developed a high number of tuberous roots and a higher concentration of inulin content compared to other species of the genus such as *Dahlia brevis* P.D.Sørensen (21.7%) but lower than *Dahlia coccinea* Cav. (70%) this shows the potential that the species has to produce tuberous and inulin roots and to be used in future studies or plant breeding programs for its exploitation as a cut flower, food and source of inulin.

KEY WORDS: inulin, tuberous roots, food, cut flower.

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