



# 32<sup>nd</sup> ANNUAL MEETING

SEPTEMBER 5-8, 2021

**BOLOGNA - ITALY**

Plesso di Agraria - University of Bologna



*Under the auspices of*



**CHAIR OF THE MEETING**

**Federica Zanetti** - University of Bologna - Italy

## PROGRAM



## Hybrid Conference Bologna, 5-8 September 2021

Venue: Plesso Agraria - University of Bologna  
Viale G. Fanin 44  
Bologna

### Program

#### DAY 1 (SUNDAY - 05/09/2021)

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- 16.00 Picking up at Savoia Regency Hotel  
*Via S. Donato, 159 - Bologna*
- 16.20 Picking up at DISTAL  
*Viale Fanin, 44 - Bologna*
- 16.30 Arrival at Cadriano experimental station
- 16.30 - 18.00 **PRESENTATION OF THE EXPERIMENTAL STATION ACTIVITIES  
AND SHORT FIELD TOUR**
- 18.00 - 20.00 Welcome reception at Cadriano experimental station
- 20.15 Return to hotels - end of day 1

Room: AULA MAGNA

- 9.00 - 12.00 **MAGIC project event**
- 9.00 The MAGIC project  
**Efthymia Alexopoulou, CRES, MAGIC project coordinator**
- 9.15 Development of genetic tools for rapid improvement of orphan biomass crops for marginal lands  
**Francesco Pancaldi, UW**
- 9.40 An overview of industrial crops on European marginal lands  
**Danilo Scordia, UNICT**
- 10.05 *Coffee break*
- 10.30 Utilization of industrial crops for the phytomanagement and remediation of heavy metal contaminated soils  
**Eleni Papazoglou, AUA**
- 10.55 Integrated sustainability assessment of selected products from marginal land: from industrial crops to value chains and the assessment of impact  
**Nils Rettenmaier, IFEU**
- 11.20 Biomass Production on Marginal Land: Mapping the Economic Feasibility Prospects for Multiple Value Chain  
**Lazaros Karaoglou, AUA**
- 11.45 Wrap up session
- 12.00 - 13.00 *Lunch*

## DAY 2 (MONDAY - 06/09/2021)

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- 13.15 Starting time of  
**32<sup>th</sup> AAIC ANNUAL MEETING program**
- 13.20 **Dr. Federica Zanetti** - AAIC President  
*Greetings and conference opening*
- 13.25 **Prof. Giovanni Molari** - Director of DISTAL  
*Greetings from the DISTAL*
- 13.30 **Prof. Luca Fontanesi** - Research delegate at DISTAL  
*Research activities at the Department of Agricultural and Food Sciences*
- 13.45 **Prof. Fabio Fava** - FF University of Bologna & IT Representative in the BBI JU SRG and EU Bioeconomy Policy Forum  
*The Biobased industry in Europe and Italy: state of the art and perspectives*
- 14.00 **Prof. Marisol T. Berti** - Editor in Chief of Industrial Crops and Products Journal  
*Presentation of the Congress Special Issue on ICP*
- 14.10 - 14.30 *Short Break*
- 14.30 - 16.55 **Plenary Speakers**
- 14.30 **Nicola Di Virgilio** - EC DG AGRI (Belgium)  
*EU main policies for renewables*
- 14.55 **Jack Grushcow** - CEO SmartEarth Camelina (Canada)  
*Moving Camelina from Novelty to Mainstream - Our 15 year Journey*
- 15.20 **Giacomo Fanin** - Business Development Manager - Cerealdocks (Italy)  
*General perspective and potential opportunities of non-food crops in Italy from one national Agribusiness leader, Cereal Docks case*
- 15.45 - 16.05 *Coffee Break*
- 16.05 **Alan Garosi** - Head of marketing at Fulgar (Italy)  
*Castor oil as an ingredient for the biobased textile: EVO by Fulgar*
- 16.30 **Jean Luc Dubois** - R&D responsible at Arkema (France)  
*Risk analysis of vegetable oils conversion to monomers.  
Main lessons learned*
- 16.55 **Round table discussion**
- 17.05 - 17.20 *Short Break*

17.20 - 19.00 **POSTER PRESENTATION - SESSION 1**  
(GENERAL CROPS AND PRODUCTS - MEDICINAL AND NUTRACEUTICAL PLANTS)  
Chairs: **Prof. Ana Luisa Fernando / Prof. Diana Jasso de Rodríguez**

## GENERAL CROPS AND PRODUCTS

- 17.20 - 17.23 P.1  
**Salvador Carlos-Hernández**  
LCA BASED STRATEGY FOR TELEMETRIC MONITORING OF AN  
AQUAPONICS SYSTEM
- 17.23 - 17.26 P.2  
**Salvador Carlos-Hernández**  
PROSPECTIVE LIFE CYCLE ASSESSMENT OF A BASED ORANGE WAX  
FUNGICIDE
- 17.26 - 17.29 P.3  
**María L. Flores-López**  
EDIBLE COATING BASED ON BLACK CHIA (SALVIA HISPANICA) SEED  
MUCILAGE CONTAINING MYRTILLOCACTUS GEOMETRIZANS FRUIT  
PHENOLIC EXTRACTS
- 17.29 - 17.32 P.4  
**María L. Flores-López**  
RHUS MICROPHYLLA LEAF EXTRACTION OBTAINED BY OHMIC  
HEATING AND THEIR PHJSICOCHEMICAL CHARATERIZATION
- 17.32 - 17.35 P.5  
**María L. Flores-López**  
CHIA (SALVIA HISPANICA L.) SEED MUCILAGE-  
CHITOOOLIGOSACCHARIDES BASED SYSTEM FOR ENCAPSULATION  
OF  $\beta$ -GALACTOSIDASE
- 17.35 - 17.38 P.6  
**Jaqueline de Mattia**  
INDUSTRIAL PROCESSING OF SUGARCANE JUICE EXTRACTED  
FROM DIFFERENT VARIETIES AIMING THE PRODUCTION OF ENERGY  
DRINK
- 17.38 - 17.41 P.8  
**Anna Karova**  
PEST CONTROL APPROACHES IN ORGANIC CULTIVATION  
OF OIL-BEARING ROSE (ROSA DAMASCENA MILL.)



- 17.41 - 17.44 P.12  
**Emilia Mihaylova**  
AERIAL MULTISPECTRAL IMAGING TO DISCRIMINATE BETWEEN DIFFERENT GENOTYPES OF COMMON WINTER WHEAT
- 17.44 - 17.47 P.13  
**Roberta Paris**  
BY-PRODUCTS FROM INDUSTRIAL HEMP INFLORESCENCES
- 17.47 - 17.50 P.14  
**Mariana Petkova**  
SOLANACEAE PLANTS GROWTH-PROMOTING AND ANTIFUNGAL ACTIVITIES OF TWO ENDOPHYTIC YEAST STRAIS
- 17.50 - 17.53 P.15  
**Slaveya Petrova**  
GENOTYPE REACTION OF SORGHUM SPECIES TOWARDS ALLELOPATHIC INFLUENCE OF SOME TYPICAL WEEDS
- 17.53 - 17.56 P.16  
**Nikolina Shopova**  
EFFECT OF MICROORGANISMS ON THE GROWTH OF TOMATO SEEDLINGS
- 17.56 - 17.59 P.17  
**Nikolina Shopova**  
HERBICIDE CONTROL OF THE WEEDS IN TOMATO (SOLANUM LYCOPERSICUM L.)
- 17.59 - 18.02 P.18  
**Krasimira Uzunova**  
USE OF PCA (ANALYSIS OF THE MAIN COMPONENTS) IN WHEAT CULTIVATION UNDER UNCONVENTIONAL CONDITIONS
- 18.02 - 18.05 P.19  
**Ciro Vasmara**  
THERMO-KOH PRE-TREATMENT AND CO-DIGESTION WITH PIG SLURRY IMPROVE METHANE YIELD AND DIGESTATE QUALITY FROM GIANT REED (Arundo Donax L.)
- 18.05 - 18.08 P.21  
**Flavia Fulvio**  
CHARACTERIZATION AND COMPARISON OF ESSENTIAL OIL COMPOSITION FROM 11 CANNABIS SATIVA GENOTYPES FROM TWO CULTIVATION SEASONS

## MEDICINAL AND NUTRACEUTICAL PLANTS

- 18.08 - 18.11 P.22  
**Mattia Alpi**  
SAFFRON AQUAPONICS CULTIVATION TECHNIQUES:  
PRODUCTIVITY EVALUATION
- 18.11 - 18.14 P.23  
**Luciana Gabriella Angelini**  
COMPOSITION AND ANTIFUNGAL ACTIVITY OF THE ESSENTIAL OILS  
HYDRODISTILLED FROM THREE ACCESSIONS OF PASTINOCELLO  
CARROT
- 18.14 - 18.17 P.24  
**Violina**  
PHYTOREMEDIATION POTENTIAL OF VETIVER GRASS  
(CHRYSOPOGON ZIZANIOIDES L.)
- 18.17 - 18.20 P.25  
**María L. Flores-López**  
COMPOSITION AND BIOLOGICAL PROPERTIES OF RHUS  
MICROPHYLLA AND MYRTILLOCACTUS GEOMETRIZANS FRUIT  
EXTRACTS
- 18.20 - 18.23 P.26  
**Maria Lourdes Diaz Jimenez**  
STABILITY ENHANCEMENT OF GARLIC-ALLICIN BY ENCAPSULATION  
IN ORGANIC AND INORGANIC MATRICES
- 18.23 - 18.26 P.27  
**Félix Martín**  
SETTING UP PRELIMINARY TESTS TO PROVE THE EFFECT OF  
COCONUT FATTY ACID AS AN APHID REPELLENT IN PEPPER
- 18.26 - 18.29 P.28  
**Elettra Frassinetti**  
NEW PERSPECTIVE FOR THE GREEN ROOF SECTOR: SEDUM SPP.  
COVERING ACCESSIONS WITH BIOMEDICAL APPLICATIONS
- 18.29 - 18.32 P.29  
**Adelina Harizanova**  
THE EFFECT OF THE PREDECESSOR AND THE NITROGEN RATE  
ON THE PRODUCTIVITY AND THE ESSENTIAL OIL CONTENT OF  
CORIANDER (*CORIANDRUM SATIVUM L.*) IN SOUTH-EAST BULGARIA

- 18.32 - 18.35 P.30  
**Dennise Anahí Carrillo-Lomelí**  
FLOURENSIA MICROPHYLLA: EFFECTS OF ULTRASOUND ASSISTED EXTRACTION ON PHENOLIC COMPOUNDS, ANTIOXIDANT AND ANTIFUNGAL ACTIVITY OF PHENOLIC EXTRACT
- 18.35 - 18.38 P.31  
**Juliana Navarro Rocha**  
CULTIVATION OF AROMATIC-MEDICINAL PLANTS, A MEETING POINT BETWEEN RESEARCH AND SOCIAL INCLUSION
- 18.38 - 18.41 P.32  
**Juliana Navarro Rocha**  
WORMWOOD CANDIAL VARIETY ESSENTIAL OIL YIELD UNDER CULTIVATION IN ARAGÓN, SPAIN
- 18.41 - 18.44 P.33  
**Valtcho D. Zheljazkov**  
ALLELOPATHIC EFFECTS OF JUNIPER ESSENTIAL OILS ON SEED GERMINATION AND SEEDLING GROWTH OF SOME WEED SEEDS
- 18.44 - 18.47 P.34  
**Mariyana Shishkova**  
OPPORTUNITIES FOR SUSTAINABLE PRODUCTION OF ROSA DAMASCENA THROUGH APPLICATION OF THE PRINCIPLES OF BIOECONOMY - A CASE STUDY FROM PLOVDIV REGION
- 18.47 - 18.50 P.35  
**Valtcho D. Zheljazkov**  
ANTIMICROBIAL ACTIVITY AND ALLELOPATHIC EFFECTS OF ESSENTIAL OILS ON SEED GERMINATION OF BARLEY
- 18.50 - 18.53 P.36  
**Ali Baghdadi**  
IMPACT OF HARVESTING TIME ON PHYTOCHEMICAL CONSTITUENT AND ANTIOXIDANT PROPERTIES OF SWEET BASIL VARIETIES
- 18.53 - 18.56 P.37  
**Diana Jasso de Rodríguez**  
FLOURENSIA RETINOPHYLLA: AN OUTSTANDING PLANT, FROM NORTHERN MÉXICO WITH ANTIBACTERIAL ACTIVITY
- 18.56 - 18.59 P.38  
**Roberta Paris**  
CANNABIS MEDICA NAZIONALE - CAMED: INNOVATION AND ENHANCEMENT OF THE PRODUCTION OF MEDICAL CANNABIS PLANT MATERIAL FOR NATIONAL DEMANDS AND NEW VARIETAL CONSTITUTION FOR PHARMACEUTICAL USE



8.00 - 12.20 **STUDY TOUR**

- 8.00 Picking up at Savoia Regency Hotel  
*Via S. Donato, 159 - Bologna*
- 8.10 Picking up at DISTAL  
*Viale Fanin, 44 - Bologna*
- 8.15 Departure to **Ferrari Museum** (Maranello)
- 9.15 Arrival to Maranello & Visit of Ferrari Museum
- 11.00 Departure from Maranello
- 12.20 - 13.00 Arrival in Conference Venue and *quick lunch*

**Room: AULA MAGNA**

13.00 - 14.40 **Concurrent scientific sessions**

**GENERAL CROPS AND PRODUCTS**

Chair: Prof. **Ana Luisa Fernando**

13.00 - 13.20 **Luigi Pari (Keynote Speaker)**

A PROTOTYPE TO CREATE SUBSURFACE WATER RETENTION SYSTEM (SWRS) TO FACE CLIMATE CHANGE: FIRST ASSESSMENT OF WORK PERFORMANCE

13.20 - 13.32 **Marisol T Berti**

CAN FORAGE SORGHUM TYPES BE GROWN AS FEEDSTOCK FOR BIOENERGY IN NORTHERN LATITUDES?

13.32 - 13.44 **Maha Elbana**

PRODUCTION OF CACTUS PEAR CROP UNDER WATER AVAILABILITY/DROUGHT CONDITIONS AND ITS IMPACT ON FRUIT PHYTOCHEMICAL CHARACTERISTICS

13.44 - 13.56 **Leandro Gomes**

PROSPECTS OF CULTIVATING GIANT REED (*Arundo donax* L.) AND SWITCHGRASS (*Panicum virgatum* L.) IN SOILS CONTAMINATED WITH HEAVY METALS - BRIDGING BIOENERGY AND BIOMATERIALS PRODUCTION WITH ECOLOGICAL REMEDIATION

- 13.56 - 14.08 **Mariusz Jerzy Stolarski**  
HOW DOES THE DOUBLE HARVEST OF PERENNIAL HERBACEOUS CROPS IN ONE YEAR AFFECTS THE BIOMASS YIELD AND ITS QUALITY?
- 14.08 - 14.40 Questions & Answers

**Room: AULA 3**

- 13.00 - 14.40 **Concurrent scientific sessions**  
**MEDICINAL AND NUTRACEUTICAL PLANTS**  
Chair: Prof. **Diana Jasso de Rodríguez**
- 13.00 - 13.20 **Dimitrios Argyropoulos (Keynote)**  
UNLOCKING THE POTENTIAL OF DATA-DRIVEN RESEARCH AND INNOVATION IN MEDICINAL PLANT VALUE CHAIN
- 13.20 - 13.32 **Dimitrios Argyropoulos**  
CURRENT RESEARCH ON DRYING OF MEDICINAL AND AROMATIC PLANTS (MAPs) BELONGING TO LAMIACEAE FAMILY
- 13.32 - 13.44 **María J. Pascual-Villalobos**  
CHARACTERIZATION OF BIOACTIVE VOLATILE BASED PRODUCTS AND ITS EFFICIENCY IN APHID POPULATION CONTROL ON A PEPPER CROP
- 13.44 - 13.56 **Hristo Djugalov**  
CONTENTS OF CERTAIN MACRO, MICRO ELEMENTS AND BIOLOGICALLY ACTIVE SUBSTANCES IN THE FRUIT OF THE GOJI BERRY VARIETIES (LYCIUM BARBARUM L.)
- 13.56 - 14.08 **Rumyana Georgieva**  
USE OF SOME PRODUCTS FOR FOLIAR APPLICATION FOR IMPROVING THE PRODUCTIVITY, QUALITY AND ESSENTIAL OIL CONTENT OF CORIANDER SEEDS (CORIANDRUM SATIVUM L.) UNDER SOUTH-EASTERN BULGARIA CONDITIONS

## DAY 3 (TUESDAY - 07/09/2021)

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14.08 - 14.20 **Diana Jasso de Rodríguez**  
ANTIOXIDANT AND ANTIPROLIFERATIVE ACTIVITIES OF  
FLOURENSIA SPP

14.20 - 14.40 Questions & Answers

14.40-15.00 *Coffee Break*

### Room: AULA MAGNA

15.00-16.30 **Concurrent scientific sessions**  
**GENERAL CROPS AND PRODUCTS**  
Chair: Prof. **Ana Luisa Fernando**

15.00 - 15.12 **Mariusz Jerzy Stolarski**  
SHORT ROTATION WOODY CROPS AS A SOURCE OF BIOACTIVE  
COMPOUNDS

15.12 - 15.24 **Manuel Cantó-Tejero**  
ANISEED ESSENTIAL OIL BOTANICAL INSECTICIDES FOR THE  
MANAGEMENT OF THE LETTUCE APHID

15.24 - 15.36 **Carolina Rodrigues**  
PECTIN EXTRACTION FROM OPUNTIA SPP. CLADODES: PROCESS  
OPTIMIZATION AND CHARACTERIZATION

15.36 - 15.48 **S. Joseph Asadauskas**  
BIO-DERIVED FEEDSTOCKS FOR NATURAL DEEP EUTECTIC  
SOLVENTS

15.48 - 16.00 **Ewelina Olba-Zięty**  
ECONOMIC ANALYSIS OF THE PRODUCTION OF SUPERCRITICAL  
EXTRACT CONTAINING BIOACTIVE SUBSTANCES FROM POPLAR

16.00 - 16.30 Questions & Answers

**Room: AULA 3**

- 15.00 -16.30 **Concurrent scientific sessions**  
**MEDICINAL AND NUTRACEUTICAL PLANTS**  
Chair: Prof. **Diana Jasso de Rodríguez**
- 15.00 - 15.12 **Heriberto Torres Moreno**  
SEASONAL EFFECT ON THE ANTIPROLIFERATIVE AND  
ANTIINFLAMMATORY ACTIVITIES OF BURSERA MICROPHYLLA
- 15.12 - 15.24 **Susana Fisher**  
BIOACTIVE COMPOUNDS IN FRUITS OF WILD MAQUI IN DIFFERENT  
RIPENING STAGES AND ENVIRONMENTS
- 15.24 - 15.36 **Ilaria Marotti**  
AGRONOMIC AND NUTRACEUTICAL CHARACTERISTICS OF  
STINGING NETTLE GROWN UNDER ORGANIC FARMING IN ITALIAN  
ENVIRONMENTS
- 15.36 - 15.48 **Eugenia Mazzara**  
MICROWAVE-ASSISTED EXTRACTION OF HEMP (CANNABIS SATIVA  
L.) TO RECOVER THREE VALUABLE FRACTIONS (ESSENTIAL OIL,  
PHENOLIC COMPOUNDS AND CANNABINOIDS): A CENTRAL  
COMPOSITE DESIGN OPTIMIZATION STUDY
- 15.48 - 16.00 **Alan Taylor**  
HEMP FUNGICIDE SEED TREATMENTS TO CONTROL DAMPING-OFF
- 16.00 - 16.30 Questions & Answers
- 16.30-16.45 *Short Break*

Room: AULA MAGNA

16.45 - 18.03 **POSTER PRESENTATION - SESSION 2**  
(OILSEEDS - FIBERS AND CELLULOSIC CROPS - NATURAL RUBBER AND RESIN)  
Chair: **Dr. Hussein Abdel-Haleem / Dr. Efthymia Alexopoulou**  
**Dr. Sam Wang**

**OILSEEDS**

- 16.45 - 16.48 P.39  
**Efthymia Alexopoulou**  
CRAMBE - WHICH VARIETY FITS BEST IN GRECE?
- 16.48 - 16.51 P.40  
**Giulio Balestriero**  
COMPOSITION IN FATTY ACIDS AND TOTAL POLYPHENOLIS IN  
DIFFERENT GENOTYPES OF CANNABIS SATIVA L.
- 16.51 - 16.54 P.41  
**Sara Berzuini**  
CAMELINA A CASH COVER CROP FOR THE MEDITERRANEAN  
REGION: PRELIMINARY RESULTS FROM THE 4CE-MED PROJECT
- 16.54 - 16.57 P.42  
**Petar Čanak**  
CAMELINA GERMINATION UNDER OSMOTIC STRESS - TREND LINES,  
TIME-COURSES AND CRITICAL POINTS
- 16.57 - 17.00 P.43  
**Sarah Chen**  
TOWARDS AUTOMATING EARLY VIGOR RATINGS FOR BRASSICA  
PLANTS
- 17.00 - 17.03 P.45  
**Flavia Fulvio**  
IDENTIFICATION OF A FAD2 POINT MUTATION PUTATIVELY  
RESPONSIBLE FOR HIGH OLEIC SEED OIL PHENOTYPE IN AN EMS-  
MUTAGENIZED MILK THISTLE POPULATION
- 17.03 - 17.06 P.46  
**Adelina Garapova**  
FATTY ACID COMPOSITION OF THE OIL FROM EXPRESS-SUN®  
SUNFLOWER HYBRIDS, DEPENDING ON SOIL FERTILITY
- 17.06 - 17.09 P.47  
**Hristofor Kirchev**  
OIL CONTENT AND YIELD OF TRIBENURON-METHYL RESISTANT  
SUNFLOWER HYBRIDS IN CONDITIONS OF DIFFERENT SOIL  
NUTRITION



- 17.09 - 17.12 P.48  
**Marina Marcheva**  
MUTAGENESIS AS TOOL FOR ENHANCEMENT OF FATTY ACID COMPOSITION OF RAPESEED (BRASSICA NAPUS L.)
- 17.12 - 17.15 P.49  
**Marina Marcheva**  
DIVERSITY OF POTENTIAL AND QUALITY OF CAMELINA (CAMELINA SATIVA) FOR HEALTHY FOODS AND BIO-ECONOMY
- 17.15 - 17.18 P.52  
**Luigi Pari**  
CAMELINA ON MARGINAL LAND: A CULTIVATION TRIAL ON STEEP SLOPE IN ITALY
- 17.18 - 17.21 P.53  
**Noemi Codina**  
WHO'S BEST? A FIELD TRIAL WITH 10 CAMELINA VARIETIES TO STUDY THEIR ADAPTATION TO SEMIARID MEDITERRANEAN CONDITIONS
- 17.21 - 17.24 P.54  
**Federica Zanetti**  
CAMELINA (CAMELINA SATIVA L. CRANTZ) A NEW OILSEED CROP FOR MEDITERRANEAN AND BALKAN EUROPEAN CLIMATES
- 17.24 - 17.27 P.55  
**Ana Luisa Fernando**  
PHYTOREMEDIATION POTENTIAL OF DIFFERENT OIL CROPS IN HEAVY METALS CONTAMINATED SOILS
- 17.27 - 17.30 P.56  
**Roque Evangelista**  
COMPARATIVE PROCESSING OF WILD-TYPE PENNYCRESS AND LIGHT-COLORED COVERCRESS TM SEEDS
- 17.30 - 17.33 P.57  
**Aritz Royo-Esnal**  
DOES SOWING PATTERN AFFECT THE COMPETITIVE ABILITY OF CAMELINA (CAMELINA SATIVA (L.) CRANTZ) AGAINST WEEDS?
- 17.33 - 17.36 P.58  
**Federica Zanetti**  
UNCOVER MORPHO-PHYSIOLOGICAL DIVERSITY IN CAMELINA (CAMELINA SATIVA L. CRANTZ) UNDER DIFFERENT ENVIRONMENTAL CONDITIONS IN EUROPE
- 17.36 - 17.39 P.59  
**Emilia Mihaylova**  
PHENOLOGY OF BRASSICA NAPUS L. FROM REMOTE SENSING DATA

## FIBERS AND CELLULOSIC CROPS

- 17.39 - 17.42 P.61  
**Ana Luisa Fernando**  
UNDERSTANDING THE POTENTIAL OF KENAF (HIBISCUS CANNABINUS L.) IN SOILS CONTAMINATED WITH HEAVY METALS IN MOZAMBIQUE
- 17.42 - 17.45 P.62  
**Michał Krzyżaniak**  
LIFE CYCLE ASSESSMENT OF SUPERCRITICAL EXTRACT OBTAINED FROM POPLAR BIOMASS
- 17.45 - 17.48 P.63  
**Emilia Mihaylova**  
LASER RADIATION TO STIMULATE TOBACCO GROWTH
- 17.48 - 17.51 P.64  
**Walter Zegada-Lizarazu**  
POTENTIAL SOC ACCUMULATION OF TWO FIBER CROPS
- 17.51 - 17.54 P.65  
**Eleni G. Papazoglou**  
COULD RAMIE BE AN ALTERNATIVE CROP FOR REMEDIATION OF CONTAMINATED SOILS IN SOUTH EUROPE?

## NATURAL RUBBER AND RESIN

- 17.54 - 17.57 P.66  
**M. Engracia Carrión**  
IDENTIFICATION OF NEW POLYPHENOLS IN LEAF OF GUAYULE AND ITS HYBRIDS
- 17.57 - 18.00 P.67  
**M. Mercedes García-Martínez**  
GUAYULE RESIN AND PORMENORIZED GUAYULIN CONTENT BY NEAR-INFRARED SPECTROSCOPY (NIR)
- 18.00 - 18.03 P.68  
**Juana Rozalén**  
COMPARISON OF ACCELERATED SOLVENT EXTRACTION EQUIPMENT'S FOR RESIN AND RUBBER DETERMINATION IN GUAYULE STEM

Room: AULA MAGNA

- 9.00-12.00 **Workshop**  
**"National and EU perspectives on non-food crops, circular and biobased economy"**  
(Moderator: **Dr. Federica Zanetti**, UNIBO)
- 9.00 **Giorgio Matteucci** (Director of CNR Bioeconomy Unit, Italy)  
*Potential and possible limitations of non-food crops for a circular bioeconomy in the EU*
- 9.20 **Giorgio Testa** (SIA executive committee member)  
*The relevance of non-food crops in Italy: the perspective of the Italian Society for Agronomy (SIA)*
- 9.30 **Ákos Kristóf** (Head of Unit - Hungarian Ministry of Agriculture)  
*The BIOEast Initiative: challenges and opportunities for non-food crops*
- 9.50 **Marina Montedoro** (Regional director of Coldiretti)  
*Actual and future regional policies for the development of the biobased economy and non-food crops*
- 10.10 - 10.30 *Coffee Break*
- 10.30 **Andrea Monti** (UNIBO, Italy, BECOOL project coordinator)  
*Advanced results from the BECOOL (Brazil-EU Cooperation for Development of Advanced Lignocellulosic Biofuel) project*
- 10.50 **Efthymia Alexopoulou** (CRES, Greece, GOLD project coordinator)  
*The GOLD project: Bridging the gap between phytoremediation solutions on growing energy crops on contaminated Lands and clean biofuel production*
- 11.10 **Ana Luisa Fernando** (FCT-UNL, Portugal, MEDIOPUNTIA project coordinator)  
*The MediOpuntia project: Promoting cactus plantation on large scale in marginal lands of Mediterranean countries*
- 11.30 **Andreas Kiessel** (University of Hohenheim, Germany, GRACE project coordinator)  
*Progress and interim results of the BBI demo project GRACE*
- 11.50 Final roundtable discussion
- 12.00 - 13.00 *Lunch*

Room: AULA MAGNA

- 13.00-14.36 **Concurrent scientific sessions**  
**OILSEEDS**  
Chair: Dr. **Hussein Abdel-Haleem**
- 13.00- 13.12 **Barbara Alberghini**  
SCREENING OF SPECIALIZED METABOLITES IN SIX CAMELINA VARIETIES
- 13.12- 13.24 **Efthymia Alexopoulou**  
LONG-TERM FIELD SCREENING TRIALS FOR CAMELINA IN GREECE
- 13.24- 13.36 **Luigi Pari**  
SWATHING AS A SUITABLE ALTERNATIVE FOR HARVESTING CAMELINA
- 13.36- 13.48 **Christina Eynck**  
A LONG TIME COMING: DEVELOPMENT OF THE HERBICIDE RESISTANT CAMELINA CULTIVAR SES1154HR
- 13.48- 14.00 **Aritz Royo-Esnal**  
WINTER WEED SUPPRESSION CAPACITY OF CAMELINA (CAMELINA SATIVA (L.) CRANTZ)
- 14.00- 14.12 **James V Anderson**  
ANALYSIS OF MOLECULAR MECHANISMS ASSOCIATED WITH LOW TEMPERATURE INDUCED FREEZING TOLERANCE AND FLORAL COMPETENCE IN CAMELINA SATIVA
- 14.12- 14.24 **Hussein Abdel-Haleem**  
GENETIC DIVERSITY AND POPULATION STRUCTURE OF USDA COLLECTION BRASSICA JUNCEA POPULATION
- 14.24- 14.36 Questions & Answers

Room: AULA 3

- 13.00-14.32 **Concurrent scientific sessions**  
**FIBERS AND CELLULOSIC CROPS**  
Chair: Dr. **Efthymia Alexopoulou**
- 13.00- 13.20 **Francesco Mirizzi (Keynote)**  
OVERVIEW AND PERSPECTIVES ON THE HEMP FIBRE VALUE CHAIN  
IN EUROPE
- 13.20- 13.32 **Dilpreet Bajwa**  
BIOBASED PLASTICIZER AND CELLULOSE NANOCRYSTALS IMPROVE  
MECHANICAL PROPERTIES OF POLYLACTIC ACID COMPOSITES
- 13.32- 13.44 **João Pires**  
CHARACTERIZATION OF CHITOSAN BIOFILMS REINFORCED  
WITH NANOCELLULOSE EXTRACTED FROM DIFFERENT  
LIGNOCELLULOSIC BIOMASSES
- 13.44- 13.56 **Ciro Vismara**  
GIANT REED HYDROLYSATE FOR SINGLE CELL OIL PRODUCTION  
BY OLEAGINOUS YEASTS LIPOMYCES STARKEYI AND  
RHODOSPORIDILOBOLUS AZORICUS
- 13.56- 14.08 **Dilpreet Bajwa**  
ENHANCING UV-SHIELDING AND MECHANICAL PROPERTIES OF  
POLYLACTIC ACID NANOCOMPOSITES BY ADDING LIGNIN COATED  
CELLULOSE NANOCRYSTALS
- 14.08- 14.20 **Francesco Pancaldi**  
GROWING NOVEL PERENNIAL BIOMASS CROPS ON MARGINAL  
LANDS: CHALLENGES FOR PLANT BREEDING
- 14.20- 14.32 Questions & Answers
- 14.36-14.50 *Short Break*



Room: AULA MAGNA

- 14.50-16.15 **Concurrent scientific sessions**  
**OILSEEDS**  
Chair: Dr. **Hussein Abdel Haleem**
- 14.50 - 15.02 **Mukhlesur Rahman**  
OILSEED BREEDING PROGRAM AT NORTH DAKOTA STATE UNIVERSITY
- 15.02 - 15.14 **Russ W. Gesch**  
IMPROVING PENNYCRESS ESTABLISHMENT THROUGH EARLIER CORN HARVEST
- 15.14 - 15.26 **Liv S. Severino**  
CASTOR MEAL FOR ANIMAL FEEDING AND CONTROLLING NEMATODES STUDIES IN PROGRESS
- 15.26 - 15.38 **Federica Zanetti**  
SAFFLOWER (CARTHAMUS TINCTORIUS L.) A MULTIPURPOSE OILSEED CROP FOR THE MEDITERRANEAN REGION
- 15.38 - 15.50 **Nesho Neshev**  
AMELIORATIVE BIOSTIMULANT APPLICATION AT SUNFLOWER HYBRIDS TREATED WITH INAPPROPRIATE HERBICIDES
- 15.50 - 16.15 Questions & Answers

Room: AULA 3

- 14.50-16.20 **Concurrent scientific sessions**  
**NATURAL RUBBER AND RESIN**  
Chair: Dr. **Sam Wang**
- 14.50 - 15.10 **Evan Sproul (Keynote)**  
SUSTAINABILITY ASSESSMENT OF PRODUCING GUAYULE RUBBER WITH COPRODUCTS
- 15.10 - 15.22 **Olivier Taugourdeau**  
ADAPTING INDUSTRIAL CROPS TO URBAN BROWNFIELDS: THE FRENCH GUAYULE CASE
- 15.22 - 15.34 **Jose Antonio Reche-Vilches**  
COLD RESISTANCE OF GUAYULE CULTIVATED IN CASTILLA-LA MANCHA, SPAIN

## DAY 4 (WEDNESDAY - 08/09/2021)

---

- 15.34 - 15.46 **Amandine Rousset**  
COMPOSITION OF GUAYULE RESIN
- 15.46 - 15.58 **Hussein Abdel-Haleem**  
GUAYULE, A PROMISING BIOFUEL AND BIOPRODUCTS CROP
- 15.58 - 16.20 Questions & Answers
- 16.20 - 16.40 *Coffee Break*

### Room: AULA MAGNA

- 16.40 - 17.30 **Concurrent scientific sessions**  
**FIBERS AND CELLULOSIC CROPS**  
Chair: Dr. **Efthymia Alexopoulou**
- 16.40 - 16.52 **Efthymia Alexopoulou**  
HOW THE IRRIGATION AFFECTS THE MISCANTHUS YIELDS IN THE DRY MEDITERRANEAN REGION
- 16.52 - 17.04 **Danilo Scordia**  
SOIL WATER AVAILABILITY ON BIOMASS YIELD AND WUE OF PERENNIAL GRASSES IN A SEMIARID AREA
- 17.04 - 17.16 **Eleni G Papazoglou**  
TOLERANCE TO AND ACCUMULATION OF CADMIUM IN THREE BAST FIBER CROPS
- 17.16 - 17.30 Questions & Answers

### Room: AULA 3

- 16.40 - 17.42 **Concurrent scientific sessions**  
**NATURAL RUBBER AND RESIN**  
Chair: Dr. **Sam Wang**
- 16.40 - 16.52 **Francisco Miguel Jara**  
GUAYULE: ALTERNATIVE CROP FOR SEMI-ARID REGIONS IN SPAIN
- 16.52 - 17.04 **Sophia Alami Tazi**  
IMPLEMENTATION GAPS FOR THE BUILDING OF A SUSTAINABLE BIOECONOMY VALUE CHAIN. LESSONS FROM GUAYULE CASE IN OCCITANIA

## DAY 4 (WEDNESDAY - 08/09/2021)

---

- 17.04 - 17.16 **Guayente Latorre**  
IMPROVING GUAYULINS SELECTIVE EXTRACTION
- 17.16 - 17.28 **Daniel Alberto Zuniga Vazquez**  
OPTIMAL DESIGN OF GUAYULE AND GUAR SUPPLY CHAINS FOR THE AMERICAN SOUTHWEST
- 17.28 - 17.40 **Sam Wang**  
IRRIGATION EFFECTS ON SEASONAL GROWTH AND RUBBER PRODUCTION OF DIRECT-SEEDED GUAYULE
- 17.40 - 17.50 Questions & Answers

### Room: AULA MAGNA

17.50-19.00 *Closing and Award Ceremony*

20.00-23.00 *Gala Dinner*  
*at Savoia Regency Hotel*



# TIMETABLE

<p style="text-align: center;"><b>DAY 1</b> <b>SUNDAY 05/09/2021</b></p>	<p style="text-align: center;"><b>DAY 2</b> <b>MONDAY 06/09/2021</b></p>
<p>16.00</p> <p><b>WELCOME RECEPTION &amp; PRESENTATION OF THE EXPERIMENTALE STATION ACTIVITIES AND SHORT FIELD TOUR</b></p> <p>20.00</p>	<p style="background-color: #008000; color: white; text-align: center;">AULA MAGNA</p> <p style="background-color: #d3d3d3; text-align: center;">MAGIC EVENT</p> <p>09.00</p> <p style="background-color: #ffff00; text-align: center;">Lunch</p> <p>12.00 13.00</p> <p style="background-color: #008000; color: white; text-align: center;">AULA MAGNA</p> <p style="background-color: #add8e6; text-align: center;">OPENING SESSION</p> <p>14.10 14.30</p> <p style="background-color: #ffff00; text-align: center;">Short break</p> <p style="background-color: #add8e6; text-align: center;">PLENARY SESSION</p> <p>15.45 16.05</p> <p style="background-color: #ffff00; text-align: center;">Coffee break</p> <p style="background-color: #add8e6; text-align: center;">PLENARY SESSION</p> <p>17.05 17.20</p> <p style="background-color: #ffff00; text-align: center;">Short break</p> <p style="background-color: #add8e6; text-align: center;">Poster presentation Session 1</p> <p>19.00</p>

# TIMETABLE

## DAY 3 TUESDAY 07/09/2021

08.00	<b>STUDY TOUR</b> <b>Motorvalley : Ferrari - Museum</b>	
12.00	Lunch	
13.00	<b>AULA MAGNA</b>	<b>AULA 3</b>
	<b>GENERAL CROPS AND PRODUCTS</b> (oral presentations)	<b>MEDICINAL AND NUTRACEUTICAL</b> (oral presentations)
14.40	Coffee break	
15.00	<b>GENERAL CROPS AND PRODUCTS</b> (oral presentations)	<b>MEDICINAL AND NUTRACEUTICAL</b> (oral presentations)
16.30	Short break	
16.45	<b>Poster presentation</b> <b>Session 2</b>	
18.03		

## DAY 4 WEDNESDAY 08/09/2021

	<b>AULA MAGNA</b>	<b>AULA 3</b>
09.00	<i>Workshop</i> <b>National and EU perspectives on non-food crops, circular and biobased economy</b>	
12.00	Lunch	
13.00	<b>AULA MAGNA</b>	<b>AULA 3</b>
	<b>OIL SEED</b> (oral presentations)	<b>FIBER AND CELLULOSIC CROPS</b>
14.36	Short break	
14.50	<b>OIL SEED</b> (oral presentations)	<b>NATURAL RUBBER AND RESIN</b> (oral presentations)
16.20	Coffee break	
16.40	<b>FIBER AND CELLULOSIC CROPS</b>	<b>NATURAL RUBBER AND RESIN</b> (oral presentations)
17.50		
19.00	<b>CLOSING &amp; AWARD CEREMONY</b>	



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# 32<sup>nd</sup> ANNUAL MEETING

SEPTEMBER 5-8, 2021

**BOLOGNA - ITALY**  
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# **ABSTRACTS GENERAL CROPS DIVISION**

**ORAL + KEYNOTE PRESENTATIONS**

**CHAIR**

**ANA LUISA FERNANDO, NOVA UNIVERSITY OF LISBON, LISBON, PORTUGAL**

## A PROTOTYPE TO CREATE SUBSURFACE WATER RETENTION SYSTEM (SWRS) TO FACE CLIMATE CHANGE: FIRST ASSESSMENT OF WORK PERFORMANCE

Luigi Pari, Walter Stefanoni, Francesco Latterini, Nadia Palmieri and Sandu Lazar

Consiglio per la Ricerca in Agricoltura e l'analisi dell'Economia Agraria (CREA)—Centro di Ricerca Ingegneria e Trasformazioni Agroalimentari, Monterotondo, Rome, Italy

Global warming and climate change are threatening the effectiveness of agriculture in Mediterranean area, which is worldwide considered as a hotspot of climate change. In particular, increasing aridity and occurrence of extreme rainfall events can have detrimental effects on agriculture, therefore there is the strong need of increasing its resilience. One of the possible solutions to tackle this issue consists of applying rain water harvesting technology (RWH), and in particular Subsurface Water Retention System (SWRS). This last consists of applying an impermeable membrane in the root zone, preventing water loss via deep percolation in sandy soils. SWRS has been studied in the recent years, highlighting its effectiveness in increasing several crops yield in arid and semi-arid climate. However, its application is limited only to laboratorial scale, due to the lack of an efficient machinery to set up the SWRS extensively. One of the aims of MediOpuntia project was properly the development of a SWRS prototype. This work represents a preliminary study, in which the evaluation of work productivity and costs of SWRS installation via this prototype was performed. The obtained results are encouraging, indeed the prototype reached target depth of 1 m for the installation of the film. Effective Field Capacity (EFC) of 0.19 ha h<sup>-1</sup> was reached, but it can be further improved working in conditions of lower presence of crop residues, which in the studied case caused clogging of the machine being in a very high amount. SWRS installation costs resulted in more than 4800.00 € ha<sup>-1</sup>. It is however worth to highlight that SWRS is permanent installation and therefore the costs would incur only once. Further studies are needed to evaluate the effectiveness of SWRS to increase crop yield in extensive field trials.

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## CAN FORAGE SORGHUM TYPES BE GROWN AS FEEDSTOCK FOR BIOENERGY IN NORTHERN LATITUDES?

Marisol T. Berti<sup>1</sup>, James V. Anderson<sup>2</sup>, Dulan Samarappuli<sup>1</sup>, and Andrea Cecchin<sup>1</sup>

<sup>1</sup>Department of Plant Sciences, North Dakota State University, Fargo, ND 58108, USA

<sup>2</sup>USDA-ARS Edward T. Schafer Agricultural Research Center, Fargo, ND, USA

Forage sorghum [*Sorghum bicolor* (L.) Moench.] cultivars and hybrids have been selected for forage yield and nutritive value, for many years. Forage sorghum is used primarily for silage and grazing. Generally, farmers use a single-cut strategy that results in the highest biomass yield at the lowest cost. In addition, currently private industry and institutes are breeding forage sorghum and sorghum-sudangrass hybrids for northern climates. Some of these new cultivars are photoperiod sensitive, which can increase biomass productivity due to the longer vegetative stage. The objective of this study was to determine the biomass yield potential of forage sorghum cultivars and hybrids grown in northern latitudes of North America and its correlation with accumulated growing degree-days (GDDs). For this study, forage sorghum yield trial data from several US states and Canadian Provinces (above the 40-degree latitude) was compiled and correlated with accumulated (GDDs). In addition, forage sorghum yield data from European countries in latitudes above 40-degrees was obtained from available literature. The data was used to conduct a meta-analysis for determining the locations where high yielding forage sorghum cultivars and hybrids have potential to be grown as feedstocks for biofuels. In the northern Great Plains, dry matter yield of biomass sorghum in trials (2009 to 2019) ranged from 8 to 30 Mg ha<sup>-1</sup> year<sup>-1</sup>, depending on cultivar and GDDs accumulated during each growing season. In the northern Great Plains, the mean dry matter yield averaged across all locations and cultivars tested was 13 Mg ha<sup>-1</sup>. However, with many cultivars yielding above the mean in most years, there is great potential for improving dry matter yield. In Europe, Germany, and many eastern European countries sorghum is grown as a feedstock for biogas production to replace the use of maize (*Zea mays* L.) silage. Raw sorghum biomass has the advantage of lower lignin and ash content compared with maize and other perennial grasses used for biofuels. Environmental impact analysis of cropping systems have further demonstrated that integrating biomass sorghum has the added benefit of reducing the carbon footprint and eutrophication compared with maize in monoculture. In conclusion, high yielding forage sorghum cultivars can be used for production of bioenergy feedstocks in northern climates. More research focused on integrating biomass sorghum into current cropping systems is needed to produce food, feed, and energy, and to attain long-term sustainability and resilience of agricultural production in northern latitudes.

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# PRODUCTION OF CACTUS PEAR CROP UNDER WATER AVAILABILITY/DROUGHT CONDITIONS AND ITS IMPACT ON FRUIT PHYTOCHEMICAL CHARACTERISTICS

Maha Elbana<sup>1</sup>, Mahmoud Elwakeel<sup>2</sup>, and Mohamed Rashad<sup>3</sup>

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<sup>3</sup>Land and Water Technologies Department, Arid Lands Cultivation Research Institute, Alexandria, Egypt

Water availability is a limiting production factor in the agricultural sector especially, in arid and semi-arid regions such as the south Mediterranean region. On the other hands, field and industrial crops play a valuable role in food security and hunger defeating. Encouraging the plantation of these crops and/or finding out the potential of other crops to be planted under drought and heat conditions characterizing the Mediterranean region for food production is one of the ways toward more sustainable food security. Cactus pear (*Opuntia ficus-indica*) is drought -heat tolerant crop that is well adapted to marginal lands in arid regions such as Egypt. The presented study investigates the effect of water availability and scarcity on fruit production and quality of cactus pear. The plant was cultivated in the experimental field of City of Scientific Research and Technological Applications in Alexandria, Egypt for two successive seasons from March 2019 to September 2020. Three irrigation regimes were applied to the crop. The first (T1) implicated no water stress on the plant applying irrigation with 12 l plant<sup>-1</sup> week<sup>-1</sup> as recommended by Ministry of Agriculture and Land Reclamation in Egypt. The second and the third irrigation regimes (T2 and T3) implicated severe drought conditions to the crop. Irrigation was applied only when water content in soil profile dropped below 35% and 30% of field capacity for T2 and T3, successively. The effect of applied irrigation regimes on the vegetative growth, water use efficiency, and fruit phytochemical properties represented in antioxidants and vitamin C contents in the fruit were studied. Results revealed no significant difference in the plant canopy cover development among the three applied irrigation regimes. It ranged from 13.63±1.28 % in T1 to 10.22±1.13 % in T3. Higher fruit yield was obtained under T1 irrigation regimes (342 kg ha<sup>-1</sup>) than T2 (226 kg ha<sup>-1</sup>) and T3 (227 kg ha<sup>-1</sup>). Yet, lower water use efficiency was observed under T1 (0.06 and 0.31 kg ha<sup>-1</sup> mm<sup>-1</sup> for 1<sup>st</sup> and 2<sup>nd</sup> seasons, respectively) followed by T2 (0.52 and 0.60 kg ha<sup>-1</sup> mm<sup>-1</sup>) and was the highest under T3 (0.59 and 0.61 kg ha<sup>-1</sup> mm<sup>-1</sup>). On the other hands, the produced cactus pear fruits under T1 had the lower nutritional value compared to the produced fruits under T2 and T3 irrigation regimes. Total phenolics (TPC) and flavonoid (TFC) content were significantly the lowest (P≤0.05) under T1 and the highest under T3. TPC content significantly varied (P≤0.05) from 82.54±1.54 GAE mg 100 ml<sup>-1</sup> in T1 to 89.51±1.08 GAE mg 100 ml<sup>-1</sup> in T2 and 92.34±0.75 GAE mg 100 ml<sup>-1</sup> in T3. TFC as well was significantly (P≤0.05) the highest under T3 (1.38±0.04 GAE mg 100 ml<sup>-1</sup>) and the lowest in T1 (0.96±0.02 GAE mg 100 ml<sup>-1</sup>). No significant difference was found among the three applied irrigation regimes in vitamin C content. Yet, it was slightly lower in T1 (4.32±0.06 g AA kg<sup>-1</sup>) than in T2 (4.43±0.02 g AA kg<sup>-1</sup>) than in T3 (4.51±0.30 g AA kg<sup>-1</sup>). In conclusion, cactus pear crop (*Opuntia ficus-indica*) is capable of producing high nutritional value fruits accompanied with high water use efficiency under severe drought conditions where soil water content could drop below 30% of field capacity. It is recommended to encourage the cultivation of this crop in marginal lands and degraded areas around cities for more sustainable food security, job opportunities and new markets openings.

Contact: Maha Elbana, Soil Dept., Faculty of Agriculture, Beni-Suef University. 320-acre East Nile Campus, East the Nile, Beni-Suef, Egypt.

PROSPECTS OF CULTIVATING GIANT REED (*ARUNDO DONAX* L.) AND SWITCHGRASS (*PANICUM VIRGATUM* L.) IN SOILS CONTAMINATED WITH HEAVY METALS – BRIDGING BIOENERGY AND BIOMATERIALS PRODUCTION WITH ECOLOGICAL REMEDIATION

Leandro Gomes<sup>1</sup>, Jorge Costa<sup>1,2</sup>, Fernando Santos<sup>3</sup>, Federica Zanetti<sup>4</sup>, Andrea Monti<sup>4</sup>, and Ana Luisa Fernando<sup>1</sup>

<sup>1</sup>MEtRICs, Departamento de Ciências e Tecnologia da Biomassa, Universidade NOVA de Lisboa, Caparica, Portugal

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Soil contamination caused by anthropogenic activity gives rise to a number of environmental problems such as desertification, contamination of water resources and/or contamination of food crops, which can lead to serious health problems for humans directly or indirectly and to disturbance of the ecosystems. To avoid these problems, soil decontamination becomes essential to guarantee land availability in the future. Phytoremediation, a bioremediation process that uses various types of plants to remove, transfer, stabilize, and/or destroy contaminants in the soil and groundwater, is an interesting option. The cultivation of energy crops, such as the perennial grasses giant reed (*Arundo donax* L.) and switchgrass (*Panicum virgatum* L.), in heavy metals contaminated soils, combines the potential soil decontamination with the production of a sustainable and renewable feedstock for bioenergy and biomaterials, providing an additional income to the farmers. Besides contributing to the bioeconomy, the process also contributes to reduce the dependence on fossil and non-renewable feedstocks, and to alleviate the soil contamination. Therefore, this work aimed to study the effects of different heavy metals (Zn, Cr, Pb, Cd, Ni, Cu) on the productivity and biomass quality of giant reed and switchgrass along three growing seasons. The soils were artificially contaminated, and the established concentrations were the double of the limits established by the Decree-Law 276 of 2009 (Portuguese regulation that establishes the regime for the use of sewage sludge in agricultural soils) - Zn: 900 mg/kg; Cr: 600 mg/kg; Pb: 900 mg/kg; Cd: 8 mg/kg; Ni: 220 mg/kg and Cu: 400 mg/kg. Overall, after three growing seasons, switchgrass showed higher yields (almost three times more) than giant reed, on the control soils and on the contaminated soils. Giant reed showed high tolerance to Zn, and switchgrass to Pb, once in these soils, the yield reduction was lower than 25%. Both crops showed medium tolerance to Cd, Ni and Cu, because the yield reduction was in the range 25-50%. Giant reed also showed medium tolerance to Cr and switchgrass to Zn. Giant reed showed low tolerance to Pb, because the yield reduction was higher than 50% and switchgrass showed no tolerance to Cr, because the seeds did not germinate. Biomass obtained in the contaminated soils presented higher ash content and lower heating value, which may limit the technological exploitation of the biomass for bioenergy, but the total fiber content was not affected by the contamination, which is important considering its use for biomaterials.

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## HOW DOES THE DOUBLE HARVEST OF PERENNIAL HERBACEOUS CROPS IN ONE YEAR AFFECTS THE BIOMASS YIELD AND ITS QUALITY?

Mariusz Jerzy Stolarski, Michał Krzyżaniak, and Ewelna Olba-Zięty

Dept. of Genetics, Plant Breeding and Bioresource Engineering, University of Warmia and Mazury, Olsztyn, Poland

The biobased economy consists of all options to generate bioproducts and energy services from different sources of biomass. Therefore sustainable supply of biomass for bioproduct or energy generation and multiple uses is needed. Solid biomass is mainly obtained from forests, wood processing, roadside, urban green care and agriculture residues. Perennial herbaceous crops (PHC) can also play an important role as a source of lignocellulosic biomass in the form of semi-woody biomass and straw. The diversity of PHC requires proper selection of genotypes with high biomass production. Moreover the crops can be harvest at different stage of vegetation period depending of their final utilization. Therefore, the objective of the study was to determine the yield and biomass quality of seven genotypes of PHC, including three dicotyledon plants: *Helianthus tuberosus* L., *Helianthus salicifolius* A. Dietr., *Silphium perfoliatum* L. and four monocots plants – grasses: *Miscanthus × giganteus* J.M. Greif & M. Deuter, *Miscanthus sinensis* (Thunb.) Andersson, *Miscanthus sacchariflorus* (Maxim.) Hack and *Spartina pectinata* Bosc ex Link during double harvest (June and October) in one vegetation period. The study was based on a three-year field experiment (2018–2020) with PHC, conducted at the Didactic and Research Station in Bałdy, University of Warmia and Mazury in Olsztyn, Poland. The highest yield of dry biomass in the first harvest in June was achieved by the *S. perfoliatum* (over 7 Mg ha<sup>-1</sup> y<sup>-1</sup> d.m. on average). However, in the second harvest (October), the yield of this species was not so high and in the total of two harvests (June + October) it amounted to over 11 Mg ha<sup>-1</sup> y<sup>-1</sup> d.m. On the other hand, among all studied genotypes, the highest total dry matter yield from two harvests was obtained by *S. pectinata* (over 13 Mg ha<sup>-1</sup> y<sup>-1</sup> d.m.). *M. sacchariflorus* and *M. sinensis* also yielded high. In turn, the yields of *H. tuberosus*, *M. × giganteus* and *H. salicifolius* were much lower. It should also be added that the yields of dry biomass of PHC harvested twice in one vegetation period, obtained in the third year of the study, were lower compared to the yields obtained in the first and the second years. It was shown that the moisture content of PHC biomass obtained in the first harvest and its nitrogen content was higher in all species compared to the second harvest. It was also found that dicotyledon plants contained more ash and lignin than grasses. On the other hand, the grasses had a higher content of cellulose and hemicelluloses than dicotyledon plants.

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GENOTYPING AND IDENTIFICATION OF DISEASE RESISTANT BARLEY LINES AND VARIETIES (*HORDEUM VULGARE L.*) TO NET BLOTCH (*PYRENOPHORA TERES DRECHSLER*)

Silviya Vasileva<sup>1</sup>, Milena Kostova<sup>2</sup> and Marina Marcheva<sup>1</sup>

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The barley crop can be affected by large number diseases such as Fusarium, leaf stripe, smuts, snow rot etc., but mostly foliar diseases can be prevalent in both winter and spring barley (Rhynchosporium, mildew, brown rust, Ramularia and Net blotch). The mosaic viruses (barley yellow mosaic virus (BaYMV) and barley mild mosaic virus (BaMMV)) have become a problem where winter barley has been grown too frequently. Sustainable control of all diseases is by rotation and mostly by the use of resistant varieties. *Pyrenophora teres* is a plant pathogen that causes Net blotch on barley (*Hordeum vulgare L.*). Nowadays Net blotch is a very important disease. It can cause typical yield losses of 10–40% where it is not well controlled. It also reduces the quality of grain, increasing screenings and the risk of crop rejection for malting. The disease can be particularly damaging when symptoms continue to develop through the winter and into the early spring, producing an early epidemic as the crop develops. It is a disease that is distributed worldwide and can be found in all regions where barley is grown. The aim of current research is an identification and genotyping reaction of barley lines and varieties to fungus disease net blotch (*Pyrenophora teres Drechsler*) in Bulgaria. The subject of the study is national and foreign elite barley genotypes, included in the core collection at the Agricultural University 's breeding garden. The perspective lines and varieties, constituting the varietal structure of Bulgaria are valued as medium-sensitive (MS 4, 5) to very sensitive (VS 8, 9) to the observed pathogen according to the Scale of Tekauz. Genotypes used as resistance identifiers by lot of authors whole over the world and global gene banks, including National Small Grain Collection, USA and NordGen, Norway, also showed sensitivity to local isolates. On the territory of Bulgaria, only very few varieties such as Forrajeira, Nomini, Igri and Jet showed symptoms of the pathogen in the later stages of their development. Symptoms of net form (*Pyrenophora teres f. teres*) and spot form (*Pyrenophora teres f. maculata*) of net blotch are observed during the vegetation stage from tilling until full maturity. All barley forms with different origin and pedigree forming the studied trait collection are subjected to selection and genetic analysis. All the genotypes were phenotyped according to their reaction to the pathogen. An ISSR marker system was used to genotype certain barley forms with different phenotypic reaction in order to their resistance to *Pyrenophora teres*. On this basis the genetic map and genetic distance between genotypes was observed. The established polymorphisms of ISSR primer application revealed genetic distance between genotypes visualized on the dendrogram. Obtained genetic map gives an idea of the identified polymorphisms relation, which may further serve to establish their additional links with the behavior of the varieties with the examined plant pathogen. Moreover, the created map can also be used to identify associations of these markers with resistance to other pests' levels, thereby allowing the identification of genomic regions contributing to horizontal (multiple) resistance, such as those of particular interest.

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## SHORT ROTATION WOODY CROPS AS A SOURCE OF BIOACTIVE COMPOUNDS

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Short rotation woody crops are easily available sources of renewable raw materials, which can be used in various branches of the medical, chemical and power industry. However, this biomass can be used primarily for the production of high added-value substances. The main objective of the study, was to evaluate the usability of the bark from different woody genotypes, grown on agricultural land in short rotations, for use as a source of bioactive compounds. The specific objectives were to determine the proportion of bark in fresh and dry dendromass, seven bioactive substance content in bark, fresh and dry bark yield, potential bioactive substance yield. This research is based on the results of a field and laboratory experiment on dendromass obtained from a plantation of short rotation woody crops, conducted in north-eastern Poland. A two-factorial field experiment was carried and it comprised fourteen genotypes, including one *Robina* genotype, five *Populus* genotypes, seven clones and one cultivar of genus *Salix* and two rotations of biomass harvest: annual and quadrennial. Biodiversity, harvest rotation and the shoot diameter had a significant impact on the bark content in fresh and dry biomass of the studied genotypes. The mean bark content in dry biomass ranged from 15% in four-year-old shoots of *S. triandra* to 48% in shoots of *P. nigra* × *P. maximowiczii* obtained in an annual harvest rotation. The genetic variability had a significant impact on the qualitative and quantitative composition of the bioactive substances in the bark of the studied plants, whereas the biomass harvest cycle determined the quantitative content of individual bioactive substances. Extending the biomass harvest rotation decreased the mean content of the substance under analysis by ca. 20%. The highest total content of bioactive substances was found in poplar bark obtained from *P. balsamifera* shoots, whereas the lowest amounts were determined in the bark of *S. dasyclados*. Among the seven bioactive substances analysed in the experiment, the highest mean content was determined for salicin, followed by salicortin. A significantly lower content was determined for salicylic acid and salireposide, whereas kaempferol and quercetin were determined in trace amounts. The content of populin in dry bark was below the level of detection of the analytical procedure applied. The total potential yield of the analysed bioactive compounds was significantly varied, both with respect to genetic variability and depending on the biomass harvest rotation.

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## ANISEED ESSENTIAL OIL BOTANICAL INSECTICIDES FOR THE MANAGEMENT OF THE LETTUCE APHID

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Anise (*Pimpinella anisum* L.) is one of the most important crops in the production of essential oils. It has traditionally been used in medicine and currently has a wide range of applications in the food industry. It is composed mainly of phenylpropanoid (*E*)-anethole. *Nasonovia ribisnigri* Mosley (Hemiptera: Aphididae) is the most damaging aphid species of lettuce grown in the open field. The control of this species is based mainly on contact insecticides sometimes overused and very toxic to natural enemies and pollinators. Lettuce is grown in large areas of monoculture in the Murcia Region in Southeast Spain; the populations of *N. ribisnigri* are developing resistance to the less and less authorized insecticides. Resistant lettuce cultivars (Nr) to *N. ribisnigri* are commonly planted. However, in the last decade, some populations of *N. ribisnigri* throughout Europe have overcome this resistance. Essential oils can be used as an alternative for the aphid's control with synthetic pesticides. We assessed the effect of aqueous nano-formulations of aniseed essential oil and its main compound (*E*)-anethole against *Nasonovia ribisnigri* in laboratory tests, semi-field in spring during 2019 and 2020 and in the open field in a plot in Southeast of Spain (Torrepacheco, Murcia) in May 2019. Randomized complete block designs were used in semi-field and field trials. The essential oils were prepared with a laboratory dispersing machine (rotor IKA Labor Pilot) at a high-speed regime (10 min, 7940 revs/min, 15°C) using Tween80 as a surfactant at ratio 1:2. Foliar applications of aniseed essential oil at 0.2 and 0.4% in lettuce plants infested with homogeneous populations of *N. ribisnigri* reduced populations and their population growth ratio (ri) compared to the control in laboratory tests (ri = -0.49 at 24 hours after treatment) and semi-field (ri ≈ -0.20 at 24 hours after treatment). In the trial field, we also observed a reduction in the aphid populations (over 50% of efficacy respect the control) without causing damage to the crop. In the same way, (*E*)-anethole gave similar results in the laboratory, semi-field and field trials than aniseed essential oil (with efficiencies of up to 47% to the control) without damages to the plant. The effect of aniseed essential oil was lower than the reference insecticide based on pyrethrins but showed a significant reduction in the number of aphids and their growth. For this reason, we are pursuing more work on the formulation of this botanical to enhance its pest control properties.

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## PECTIN EXTRACTION FROM *OPUNTIA* SPP. CLADODES: PROCESS OPTIMIZATION AND CHARACTERIZATION

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Cactus (*Opuntia* spp.) is a xerophytic plant which is particularly adapted to arid and semiarid lands (e.g. Mediterranean area and North Africa) and severely degraded soils that are unsuitable for traditional crops, thus being considered a promising crop to overcome the climate change effects in agriculture. It is a spiny succulent plant of the *Cactaceae* family and its fruits, the cactus pear, with an edible portion of 54%, is a food with nutraceutical and functional importance. The fruits are widely used for human consumption and its vegetative part, the cladodes are essentially used as feed. Cladodes contain in their composition a complex of high molecular weight named mucilage. This mucilage contains pectin, a polysaccharide, capable to form viscous solutions with promising application in different areas, e.g. the food industry and as edible films and coatings. Application of the extracted pectin to substitute fossil-based products, e.g. plastics in the food packaging, will contribute to accomplish the targets of the European Green Deal in terms of reducing fossil-based packaging waste and reducing dependence on non-renewable, unsustainable resources, helping to decarbonize the economy. Thus, the aim of this work was to optimize a method to extract pectin from cladodes, in an efficient and sustainable manner. Pectin extraction procedure includes: (1) washing/cutting, (2) mixing with solvents, (3) centrifugation; (4) precipitation with ethanol; and (5) drying. To optimize the extraction process, different parameters were tested: pH, different solvents (water and aqueous solutions of citric and acetic acids), extraction temperature and time and also the liquid/solid ratio. The characterization of the initial biomass and the resulting materials was accessed in terms of moisture, ash, total fiber content, protein content, total sugars and pectin content through the evaluation of the galacturonic acid. The initial biomass characterization revealed that cladodes have a high moisture content (90-95 % w/w, wet basis), an ash content of about 16.5-20 % w/w (dry weight basis). The Neutral detergent fiber (NDF) content was in the range 26-29 % dry weight basis and the crude protein was on average 7-8 % in dry weight basis. Preliminary results indicate that an average yield (in dry basis) of 7.5 - 15 %, of pectin can be obtained. Higher extraction yields can be obtained through the use of low pH (2-3), and high extraction temperatures (70-80 °C). Also, the use of citric acid and acetic acid improves the extraction yields once they avoid pectin depolymerization. Increasing the extraction time to 50-60 minutes also ensures a higher extraction yield. The resulting extracted materials and commercial pectin were analyzed through ATR-FTIR and revealed similarities on the characteristic chemical groups. Work is in progress to identify which extraction procedure simultaneously presents good yields and pectin with optimal characteristics for the production of biopolymers, namely film forming ability. Also, application of pre-treatments (pulsed electric fields, ultrasound, microwave) are being tested to identify options for yields improvement.

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## BIO-DERIVED FEEDSTOCKS FOR NATURAL DEEP EUTECTIC SOLVENTS

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Deep eutectic solvents represent mixtures, where two or more compounds interact through hydrogen bonding (i.e. H-bonds) and dramatically reduce their melting point. As a result, even some solid substances can become liquid just by being mixed together without any chemical reaction, only by acting as H-bond donors or acceptors. Many metal salts or hydroxides can engage into such eutectic phenomena, driven not only by H-bond interactions, but also due to the processes between Lewis or Bronsted acids and bases. Although heating usually greatly accelerates the dissolution of solids and formation of deep eutectic solvents, thermodynamically ambient conditions might as well be sufficient for the liquids to form. Recently, researchers have shifted their focus to Natural Deep Eutectic Solvents (NADES), which are composed of organic compounds from bio-derived resources. Low melting points and unusual solvency of NADES is attracting a lot of interest from various industries, including food, cosmetics and plastics. Amino acids and sugars are among the most widely employed components, acting as H-bond acceptors and H-bond donors respectively and as a result these NADES are gradually becoming a significant player in bio-economy. Betaine and its salts have been studied with particular attention as H-bond acceptors by a number of teams. Nevertheless, fluidity and stability of betaine formulations in NADES remains quite uncertain. With a purpose to establish the relationships between fluidity or stability characteristics and the compositions of betaine-based NADES, a series of H-bond donors from bio-derived feedstocks were screened experimentally. The principal results demonstrate that the dissolution of solids strongly depends on the ratios between NADES components. In some formulations, the concentration interval is quite narrow for the blend to become liquid and retain fluidity. For example, NADES formulations of betaine and xylitol combine into sufficiently stable liquids only when betaine concentration is within 33% to 40% wt. Although clear uniform liquids might form after initial heating and retain their fluidity for many hours at room temperature, prolonged storage of some NADES often results in phase separation and formation of solids. These processes might be accelerated or inhibited, if some water is present in the formulations. Nevertheless, the tests show that betaine-based NADES can be produced as clear stable liquids with excellent low temperature stability by using a broad selection of various short chain sugars, polyhydric alcohols, dicarboxylic and functionalized acids or similar compounds as H-bond donors. These components can be obtained from diverse sources and not necessarily in a pure dehydrated form. It should be noted that purity is important for NADES fluidity and stability, so candidate formulations must be carefully screened for their suitability. In conclusion, despite remaining formulatory and technical challenges, employment of sugars, polyhydric alcohols and similar H-bond donors for betaine-based and other types of NADES promises high degree of innovation and a broad spectrum of possibilities for bio-derived material utilization.

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## ECONOMIC ANALYSIS OF THE PRODUCTION OF SUPERCRITICAL EXTRACT CONTAINING BIOACTIVE SUBSTANCES FROM POPLAR

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The concept of a biorefinery assumes the multi-product use of biomass for the production of high-value products first, and then the energetic use of post-production residues. In addition to environmental reasons for the validity of such concepts - the profitability of a project is also important. The main objective of the research was to assess the economic profitability of production, including the analysis of life cycle cost of the selected bioproduct value chain in terms of biorefining. The biomass raw material selected for the research was poplar (*Populus tremula* L.), harvested in a short rotation. The main bioproduct was dry extract from poplar lignocellulosic biomass, while post-extraction biomass was used to produce an energy co-product - pellets. The main bioproduct - supercritical dry extract (SFE) was obtained by supercritical extraction and is an innovative ingredient in high-value bioproducts, e.g., veterinary ones with anti-inflammatory properties. As part of the research, the costs of production in the SFE value chain from poplar and pellets were determined. The analysis included the determination of investment costs (CAPEX) and operating costs (OPEX) and a life cycle costing (LCC). Moreover, the production profitability analysis was carried out, the break-even point for the production of poplar chips, SFE and pellets was determined. For poplar chips and pellets, market selling prices were adopted and, on this basis, investment profitability ratios were determined, ie NPV, IRR, PI, DPBP, while for SFE, investment profitability ratios were determined assuming a margin of 10% from the break-even point. The scope of the research covered the following system boundaries: subsystem I, i.e., costs and revenues related to the production of poplar biomass in an annual cycle, including the establishment of a plantation, fertilization, harvesting and preparation of wood chips, plantation lifetime 20 years and liquidation of plantations. The next subsystem (II) covered the costs of transport to the pre-treatment plant and the costs of drying and grinding processes as well as the costs of transport to subsystem III. Subsystem III called biorefinery included costs and revenues related to the production of SFE as well as costs and revenues related to the production of pellets. The largest investment costs (CAPEX) were related to the purchase (73%) and installation (22%) of the supercritical extraction line, while the largest operating costs (OPEX) were personnel costs (59%) and costs of materials and utilities related to the supercritical extraction process (37%).

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# **ABSTRACTS GENERAL CROPS DIVISION**

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## LCA BASED STRATEGY FOR TELEMETRIC MONITORING OF AN AQUAPONICS SYSTEM

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Aquaponics is being studied as an alternative for intensive production systems. It consists in the combined production of fish (aquaculture) and vegetables (hydroponics). The nutrients produced in aquaculture are sent to the hydroponics crop to be assimilated by the vegetables; as a part of its growth process, vegetables will generate nutrients which are useful for fish. The studied system in this work is part of a structure intended to the integral transformation of waste fats in Tijuana, north-west of Mexico. In the metropolitan area, waste fats produced by restaurants and fat traps are revalorized by a local enterprise through several processes: mechanical separation, fractioned distillation, transesterification and composting. Those processes generate wastewater which is treated by electroflocculation. An aquaponics system is being studied in order to take advantage of the treated water. Lettuces and tilapias are considered as raising cultures by hydroponics and aquaculture processes, respectively. A telemetric monitoring system has been implemented to measure crop parameters (status of vegetable and fish, temperature and status of water, among others); this system is composed by a drone and informatics equipment for data processing. In order to perform an adequate operation of the aquaponics system, it was identified the necessity of a strategy to manage the different elements involved in the system. In view to design such strategy considering environmental indicators, a life cycle assessment (LCA) has been proposed. The system was divided in: aquaponics (tilapias and lettuces crops), monitoring system (drone) and telemetrics (data processing system). The functional unit was: 1 day of monitoring. The inventory was conformed from information provided by the enterprise owner of the aquaponics system and from official data related to electricity and water. The method to evaluate the environmental impacts was the one designed by the Institute of Environmental Sciences (CML from the Dutch) CML baseline v.4.4.4 by using the software openLCA v.1.10.3. Based on the results of the LCA, a monitoring protocol is proposed to reduce the environmental load, which is mainly due to nutrient recirculation, water pumping and number of drone flights. This includes an optimized flight plan, the generation of reference data from own measures, the continuous calculation of the status of the crop and its comparison with the ideal state. From the sensitivity analysis it is concluded that improving the three factor a reduction of 10%, 10% and 60% on average, respectively, can be achieved.

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## PROSPECTIVE LIFE CYCLE ASSESSMENT OF A BASED ORANGE WAX FUNGICIDE

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The use of pesticides is a recurrent strategy to reduce the negative effects of phytopathogens over crops. In recent years, fungicides based on natural products are being developed in order to reduce the negative characteristics of synthetic ones. A particular strategy is to obtain the active components from the byproducts of the food industry. In this context, Frutech International Corporation, an enterprise located at Monterrey, Mexico, produces orange (*Citrus × sinensis*) essential oils; in this process a solid wax waste containing waxy compounds (hydrocarbons, esters, alcohols) and phytochemicals (resveratrol and limonene, coumarins, furanocoumarins) is generated. The fungicide production from orange wax waste (OWW) has been identified as an alternative to revalorize this byproduct; the control of *Fusarium oxysporum* and *Colletotrichum gloeosporioides* has been proven in a previous work at lab scale. Then, the idea of the present study is to identify opportunity areas in the scaling up of the fungicide production process through a prospective life cycle assessment (LCA). This assessment was performed as described in next lines. Data from lab experiments were scaled up to a pilot plant considering as functional unit “the production of 1 m<sup>3</sup> of OWW fungicide”. The OWW dilution and the fungicide formulation were considered as the main stages of the studied process; two management scenarios for insoluble wax were considered: the traditional disposal and the transesterification for biodiesel production. The calculation of the environmental impacts was performed using the software OpenLCA v.1.10.3 considering the database ELCD v. 4.2. and the method CML (baseline); Climate change, Global warming, Human toxicity, Terrestrial ecotoxicity are among the impact categories included for the assessment. The obtained results show that the transport of supplies and the use of energy contribute the most to the environmental impacts of the fungicide production process; the main affected categories were climate change and global warming. By replacing the extraction solvent, the total environmental impacts are reduced around 30%; also by modifying the profile of energy consumption, the environmental impacts can be reduced near to 20%. Moreover, the transesterification of insoluble wax induces few benefits to the global process since the current biodiesel yield is low. It is concluded that from an environmental viewpoint, the fungicide production is a feasible alternative for the OWW revalorization.

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## EDIBLE COATING BASED ON BLACK CHIA (*SALVIA HISPANICA* L) SEED MUCILAGE CONTAINING *MYRTILLOCACTUS GEOMETRIZANS* FRUIT PHENOLIC EXTRACTS

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*Salvia hispanica* L is an annual herbaceous plant that belong to the *Lamiaceae* family, its popular name is chia, and it is native from sothern Mexico and northern Guatemala. The chia mucilage is considered a reliable source of soluble fiber, minerals, and polysaccharides. Edible film packaging has proven effective as bioactive compounds carriers (e.g., antioxidants, antimicrobials, nutraceuticals, and nutrients), acting as an active agent protector and prolonging its action. In this context, the application of edible coatings (EC) based on different natural matrices (e.g. polysaccharides, proteins, and lipids), has shown favorable effects to extend the shelf life of fruits and vegetables, in addition to being an appropriate vehicle for the incorporation of bioactive compounds. Extracts obtained from plants of arid zones of Mexico has different properties as antioxidants, antimicrobials, etc., which in synergy, potentiate their protective action. The objective of the present study was to evaluate the effect of a EC based on chia (*Salvia hispanica* L.) functionalized with extracts of *Myrtillocactus geometrizans* on the postharvest quality of blackberry. *M. geometrizans* fruit was characterized by its high content of carbohydrates (67.4 g 100 g<sup>-1</sup>), fiber (6.7 g 100 g<sup>-1</sup>), ashes (5.2 g 100 g<sup>-1</sup>), and protein (5.1 g 100 g<sup>-1</sup>); besides, it presented an interesting content of reducing sugars (854.9 mg AT g<sup>-1</sup>). These results confirmed the potential of *M. geometrizans* fruit as an alternative and innovative source for obtaining natural bioactive compounds. In the production of *M. geometrizans* extracts, a higher yield was obtained in the hydroalcoholic extract (EHA, 52.2%) compared to the aqueous extract (EA, 42.7%). Furthermore, it presented a higher total phenolic content (TPC) with values of 10.1 g GA kg<sup>-1</sup> extract, better antioxidant potential (CI<sub>50</sub> = 6.0 mg mL<sup>-1</sup>) and an interesting phenolic profile, highlighting the content of rosmarinic acid (12.4 mg L<sup>-1</sup>), ellagic acid (5.1 mg L<sup>-1</sup>), and rutin (2.8 mg L<sup>-1</sup>). EHA also demonstrated a greater inhibitory effect than EA on *Fusarium oxysporum*, a fungus of commercial importance in blackberry, with minimum inhibitory concentration (MIC) values of 1915 and 4881 mg L<sup>-1</sup> to inhibit 50 and 90% of fungal growth, respectively. Considering that EHA demonstrated better properties in all the parameters studied (phenolic profile, antioxidant, and antifungal potential), it was selected for its incorporation in a chia-based EC (EC + G) at 1915 mg L<sup>-1</sup> (MIC<sub>50</sub>), to evaluate its effect on the blackberry shelf life at 5 °C and 85% relative humidity (RH) for 15 d. In general, the application of EC + G was a protective barrier in blackberry, as the coated fruit did not show weight loss until day 6 of the evaluation; while, at the end of the test, it allowed to maintain the weight loss of the fruit below 3.2% compared to the uncoated fruit (19.6%). On the other hand, the color of the treated fruit was retained during storage, presenting greater luminosity (L\*) than the control treatment. However, the action of the EC + G was not prolonged during the test in other parameters (e.g. firmness and microbiological control), being associated with the biodegradable nature of the coating. This research provides a vision of the scope and importance of the development chia edible coatings of new alternatives using extracts of *M. geometrizans* to improve the shelf life of fruit of commercial importance in Mexico.

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## RHUS MICROPHYLLA LEAF EXTRACTS OBTAINED BY OHMIC HEATING AND THEIR PHYSICOCHEMICAL CHARACTERIZATION

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Plant species in arid zones of Mexico are considered of phytochemical importance, being commonly used in traditional medicine as a natural remedy for general ailments. Recently, they have been received a considerable attention for their great antioxidant and antifungal potential, which derives from their interesting presence of bioactive compounds. In this context, agrito (*R. microphylla*) shrub is distributed in Mexico and the southeastern United States and has been traditionally used by the Kickappos indigenous people in folk medicine; however, studies of its composition and bioactivity are limited. Therefore, the objectives of this work were to: (1) characterize the leaves of *R. microphylla*, (2) obtain hydroalcohol (HAE) and aqueous (AE) extracts by ohmic heating using two extraction times (5 and 10 min); and (3) evaluate their bioactivity in terms of antioxidant activity by the DPPH (2,2-diphenyl-1-picrylhydrazyl) assay and antifungal activity *in vitro* against *Fusarium oxysporum* and *Corynespora cassiicola*. The results showed a high content of carbohydrates and ashes in *R. microphylla* leaves ( $78.1 \pm 1.3$  and  $10.5 \pm 0.5\%$ , respectively); while the protein ( $0.8 \pm 0.1\%$ ) and lipid ( $8.9 \pm 0.4\%$ ) values were lower than those reported for other species of *Rhus*. The ANOVA indicated that there are no significant differences in the yields or antioxidant activity of the extracts, as there was no ( $p > 0.05$ ) effect of the solvent and extraction time in this parameter. However, these conditions influenced the recovery of sugars and total phenolic content. Total and reducing sugars were significantly higher in both aqueous extracts of *R. microphylla* leaves, being the highest content ( $p < 0.05$ ) for the AE obtained with 5 min of extraction ( $308.1 \pm 4.7$  and  $107.3 \pm 1.3$  g/L, respectively), followed by AE obtained with 10 min of extraction ( $252.1 \pm 1.3$  and  $82.6 \pm 0.8$  g/L, respectively). Regarding HAE, total sugars value was affected by the extraction time, with higher values for HAE obtained with extraction time of 10 min ( $235.2 \pm 4.0$  g/L); but there were no significant differences in the content of reducing sugars of HAE extracts ( $70.1$  to  $70.3$  g/L). All the extracts exhibited interesting antioxidant activity, which was expressed as  $IC_{50}$  value (i.e., concentration required to obtain 50% of inhibition of radical scavenging activity), being in the range of 0.33 - 0.36 mg/L. However, the extracts under study showed low antifungal activity on *F. oxysporum* and *C. cassiicola* (<30%). These results reveal for the first time the potential of the *R. microphylla* leaves extracts, although further studies are needed to establish optimal recovery conditions and elucidate their bioactivities.

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## CHIA (*SALVIA HISPANICA* L.) SEED MUCILAGE-CHITOLIGOSACCHARIDES BASED SYSTEM FOR ENCAPSULATION OF $\beta$ -GALACTOSIDASE

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Chia (*Salvia hispanica* L.) is an endemic seed of Mexico, which is traditionally consumed for its health benefits. It produces an interesting mucilaginous polysaccharide able of interacting with other biopolymers forming coating materials. However, its exploitation and appreciation in the Mexican market is still limited. On the other hand, the deficient digestion of lactose caused by the low activity of the  $\beta$ -galactosidase (lactase) affects 65% of the world population, for which various studies focus on the development of lactose-free products that maintain their properties. The objectives of this work were to design an encapsulation system based on chia mucilage and chitooligosaccharides and evaluate its ability to encapsulate  $\beta$ -galactosidase for its application in the goat milk delactosing process as model food. The capsule-forming solutions were based on a 2-factor Taguchi experimental design at two levels (high and low) of chia (40 and 60%, w/v) and chitooligosaccharides with a molecular weight of 1000 and 3000 Da (0.4 and 0.6%, w/v), thus generating eight combinations in total. The results showed an average capsule size of 2.5 - 3.2 mm, while capsule hardness was in the range of 6.8 - 8.2 N. The kinetic parameters (maximum velocity- $V_M$  and the Michaelis constant- $K_M$ ) of encapsulated  $\beta$ -galactoside were determined. The effectiveness of the encapsulation system to protect and release of  $\beta$ -galactoside activity was confirmed; however, additional studies are required to characterize the system and determine optimal conditions for its incorporation in the goat milk delactosing process. This study exhibits a novel use of chia as a vehicle of biomolecules, representing a positive impact on the sustainable exploitation of chia for its application in the agri-food industry.

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## INDUSTRIAL PROCESSING OF SUGARCANE JUICE EXTRACTED FROM DIFFERENT GENOTYPES, INSIGHTS TO INCREASE ITS ADDED VALUE

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Sugarcane (*Saccharum* spp) is one of the main crops in the world, cultivated in more than 120 countries, with Brazil, India and China being the main world producers. Sugarcane belongs to the Poaceae family and it is an economically important plant due to its fibrous stalk rich in sucrose. In Brazil, it is a crop of major economic and industrial significance, used for the manufacturing of several products among which white sugar and biofuel production. Sugarcane juice is the liquid extract obtained from sugarcane culms using mechanical compression. It is very rich in different compounds, such as phenolic compounds (anthocyanins, flavonoids, among others), which present bioactivity, melanoidins, melanins, and carbohydrates. In this sense, the objective of this work was to characterize and evaluate the chemical and biochemical composition of sugarcane juice from different genotypes in order to provide insights to increase its added value. A total of 8 fresh sugarcane juice samples of 4 genotypes were used, 4 samples extracted from sugarcane with husk and 4 samples from peeled sugarcane. The sugarcane (*Saccharum officinarum* L.) genotypes tested were RB016916, RB966928, RB925345 e RB935744. These genotypes were chosen because they are widely cultivated under different edaphoclimatic conditions in Brazil, especially in a milder climate, such as the one in Rio Grande do Sul, South Brazil. They present physiological maturity cycles from early to medium and productivity above the Brazilian average, 100 t/ha, with 35% broth production. In all samples, enzymatic activity, minerals, ICUMSA (International Commission for Uniform Methods of Sugar Analysis) color from broth and total characterization and quantification of the phenolic profile were analyzed. The average pH of the samples was 5.26 and the °Brix content was 21.31. The ICUMSA color presented values ranging between 1856 (RB016916) and 3927 (RB935744). The average content of ascorbic acid approached 1 mg per 100 mL of broth. The total acidity was below 0.1 g (citric acid)/100 mL for all samples. The average activity of polyphenol oxidase was 25 EU (enzymatic unit), with samples with husk showing higher value. The peroxidase activity was higher, presenting 64 EU in peeled samples, and samples with husk showed more than three times this result (194 EU). This indicates a greater presence of this enzyme in the sugarcane husk. The mineral results indicate the predominance of K, remaining between 438 and 906 mg/L of sugarcane juice. It was observed that the total phenolic content was higher for the samples of sugarcane juice with husk. The prevalent phenolic compounds identified in the sugarcane genotypes were catechin, luteolin, tricine, chlorogenic acid, coumaric acid and ferrulic acid. Catechin being more prevalent in samples with husk (206 mg/L) and ferrulic acid in samples without husk (85 mg/L). Given the preliminary results obtained, it can be concluded that samples of sugarcane with husk, genotypes RB016916 and RB966928, showed to have superior content of bioactive compounds and minerals. Results obtained in this study will allow to explore new potential uses of sugarcane juice in different sectors.

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PEST CONTROL APPROACHES IN ORGANIC CULTIVATION OF OIL-BEARING ROSE  
(*ROSA DAMASCENA* MILL.)

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Bulgaria is among the most important producers of rose oil in the world. The oil-bearing rose is grown mainly in the region of the Rose Valley, where the Karlovo and Kazanlak fields are situated, as well as in the districts of Stara Zagora, Plovdiv and Pazardzhik. The main part of the rose flower is processed into emblematic Bulgarian rose oil, and the rest into rose concrete and rose water. In recent years, there has been an increased interest in organic rose cultivation and production of organic rose oil, related to both greater marketing opportunities and higher financial support for organic production under the Rural Development Program. In order to receive optimal yields and pesticide free high quality products plant health protection strategy and successful pest management are needed. Plant protection measures in organic agriculture are totally different from those in conventional production and are not simple replacement of the synthetic pesticides with biological ones. This paper reviews main pests associated with oil-bearing rose in Bulgaria and focuses on the ecologically sound approaches based on production standards and rules. It also identifies the possible methods and preventive measures to be implemented on farm level and gives some recommendations for national policy concerning organic oil-bearing rose and rose oil production.

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## AERIAL MULTISPECTRAL IMAGING TO DISCRIMINATE BETWEEN DIFFERENT GENOTYPES OF COMMON WINTER WHEAT

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Multispectral reflectance phenotyping and genomic selection are two emerging technologies that have the potential to increase plant breeding efficiency by improving prediction accuracy for grain yield. Accurate retrieval of crop phenology information is a precondition for evaluating crop adaptation to climate change, modeling agricultural ecosystem carbon exchange, and predicting future agricultural production. Multispectral remote sensing is one of the advanced and effective techniques to discriminate crops phenology. The miniaturisation of multispectral sensors for use on drones have provided an opportunity to obtain multispectral data that may be used to discriminate between different genotypes of crops. Differentiating the characteristics between different wheat (*Triticum aestivum* L.) genotypes in the initial steps of their field testing is of fundamental importance. The objective of this study is to validate the effectiveness of multispectral data to characterize the differences generated from wheat genotypes. Various reactions to different stress factors of 12 registered varieties and 22 breeding lines of common winter wheat have been monitored during the vegetation period of 2020 – 2021. The late frost in April damaged in different degree all analyzed accession. The subsequent drought also hampered the wheat vegetation. The conditions of a humid and cool May and June enhanced the development of fungal diseases and insect attack. The application of fungicide (azoxystrobin) and insecticide (lambda-cyhalothrin) in BBCH 75 caused visually clear chlorosis to plants. The normalized difference vegetation index (NDVI) and Enhanced Vegetation Index (EVI) have been widely used for remote measurement of vegetation for many years. Higher NDVI values reflect greater vigor and photosynthetic capacity of vegetation leaves, whereas lower NDVI values for the same time period are reflective of vegetative stress. This study employed the NDVI and EVI to discriminate between different genotype modifications of *Triticum aestivum*. The results show a proven difference in the NDVI and EVI between different genotypes of common winter wheat one day after treatment with plant protection products. Thus, the aerial multispectral imaging proves to be a sensitive and useful tool for discriminating of wheat genotype modifications. It can also be employed to evaluate the effect of different herbicides on the crop's health. Early diagnosis of the plant response after mistreatment could help to take urgent measures to restore optimal crop vegetation.

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## BY-PRODUCTS FROM INDUSTRIAL HEMP INFLORESCENCES

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The cultivation of hemp (*Cannabis sativa* L.) is experiencing a rebirth that involves numerous industrial sectors thanks to the multifunctionality of the species. Once used mainly as a source of fiber and cellulose to obtain textiles and paper, today it finds application in the green building sector, in food production, in cosmetics, and in the field of ornamental plants and horticulture, as well as in the pharmaceutical industry.

*C. sativa* is typically characterized by phytocannabinoids, a class of terpenophenolic compounds accumulating in female inflorescences. The so called ‘Big four’ are the TetraHydroCannabinolic Acid (THCA), the CannaBiDiolic Acid (CBDA), the CannaBiChromenic acid (CBCA) and the CannaBiGerolic acid (CBGA), the last one precursor of the others through the action of specific enzymes. Cannabinoids are synthesized in plants in the acidic forms which are converted in non-enzymatic way to their de-carboxylated molecules, known for pharmacological properties. Only THCA is the precursor of the psycho-active molecule known as THC. Agro-industrial hemp differentiates from *marjuana* for the content in THCA; the legal limit for cultivation in Italy is 0.2% weight/weight, and with up to 0.6% THC in the crops no penal responsibility is attributed to the farmer. According to law 242/2016 hemp cultivation was promoted as a way to preserve biodiversity and reduce the environmental impact of agriculture. The law allows growers to plant only hemp varieties registered in the EU’s ‘Common Catalogue of Varieties of Agricultural Plant Species’.

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## SOLANACEAE PLANTS GROWTH-PROMOTING AND ANTIFUNGAL ACTIVITIES OF TWO ENDOPHYTIC YEAST STRAINS

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In this study, two yeast strains were enriched and isolated from different plant tissues. YP6 was isolated from *Triticum aestivum L.* seeds. YBS14 was isolated from the root of *Helichrysum italicum L.*, which is mainly used for its limited habitat and importance as a medicinal plant. Partial sequence analysis of ITS5-5.8-ITS4 region of the nuclear ribosomal DNA with universal primers identified YP6 as *Pichia fermentas* and YBS14 as *Saccharomyces cerevisiae*. Both isolated yeast strains produced indole-3-acetic acid (IAA) when cultivated in YPD medium supplemented with 0.2% L-tryptophan. The antimicrobial activities of yeast strains against several *Solanaceae* plant pathogenic fungi (*Alternaria solani*, *Rhizoctonia solani*, and *Fusarium solani*) were determined. YBS14 inhibited the growth of all tested pathogens, while YP6 inhibited all fungi except *Alternaria solani*. Both YP6 and YBS14 were tested for endophytic colonization of tomatoes, peppers, and eggplants by two different inoculation methods - soil drench and leaf spraying. To establish colonization in the various tissues of tested plants, samples were taken on 7th, 14th, and 21st-day inoculation and explants were inoculated on YPD agar. Both techniques of inoculation showed high frequency of colonization from 83.33% to 100%. The other method is directly observation of endophytic yeast inside plant tissues using a microscopic technique. YBS14 cells are located in the intercellular space of cells at the root of plants. Although, YP6 yeast cells were found in the cytoplasm of plant cells. In order to determine the effectiveness of the microbial endophytes, their effect on some physiological processes in the plant is analyzed, such as photosynthesis, stomatal conductivity, and transpiration intensity. Plants have the ability for structural and functional adaptation to stress effects of different nature. All treated plants have a higher content of photosynthetic pigments compared to the control, photosynthesis is probably more intense and growth stimulation was observed. The chlorophyll a / b ratio remained similar, and the total chlorophyll / carotenoid ratio slightly increased as a result of elevated chlorophyll levels. The most significant stimulating effect was recorded in plants treated by foliar spraying with YP6. The final aim of the present study is to evaluate endophytic yeast, physiological - biochemical, genetic and cultural characteristics in order to demonstrate PGP-activity and their possible application in organic farming.

## GENOTYPE REACTION OF SORGHUM SPECIES TOWARDS ALLELOPATHIC INFLUENCE OF SOME TYPICAL WEEDS

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Sorghum, which mainland is East Africa, is one of the first crops, which was taken into cultivation. At present, *Sorghum sudanense* hybrids are preferred as a fodder due to higher yields and sibling ratios and also their thin stems and higher leaf ratios. *Sorghum vulgare* var. *technicum* is mainly utilized for producing brooms, washing brushes, knittings, paper, wallboard, fences, biodegradable materials for packaging due to their peculiar resistance. Sorghum cultivar plants are more tolerance to drought and hot temperatures, diseases, pests, various soil types and have higher water use efficiency, higher production capacity for unit area. Their main disadvantage is the high sensibility against weed infestation in the first 30-40 days after sawing. In 2017-2018, some ex-situ experiments were carried out at the Agricultural university – Plovdiv aiming to assess and compare the allelopathic effect of cold aqueous extracts from some typical weeds (*Sorghum halepense* (L.) Pers., *Aristolochia clematites* L., *Cirsium arvense* Scop., *Sonchus arvensis* L. and *Xanthium strumarium* L.) in concentrations of 0.1%, 0.2%, 0.4%, 0.8% 1.6% and 3.2% w/v on the seed germination of *Sorghum vulgare* var. *technicum* Körn (local population) and *Sorghum sudanense* (Piper.) Stapf (300/43 mutant form). Tendency to reduce shoot and root length with an increment of the concentration was found in all experimental variants ( $p < 0.05$ ). Data revealed that the highest allelopathic effect on seed germination and initial development of both studied *Sorghum* crops at laboratory conditions was found for the extract from *Sonchus arvensis* L. which OAP (overall allelopathic potential) had maximum values of 9.56 (*S. sudanense*) and 13.84 (*S. vulgare* var. *technicum*). According to the OAP values from the experiment with *S. sudanense*, five studied invasive weed species could be arranged as follows: *S. arvensis* > *X. strumarium* > *A. clematites* > *C. arvense* > *S. halepense*, while in the experiment with *S. vulgare* var. *technicum* the descending order was: *S. arvensis* > *C. arvense* > *X. strumarium* > *A. clematites* > *S. halepense*. Two tested *Sorghum* sp. showed different sensibility to the allelopathic effect of the cold aqueous extracts from dry above-ground biomass of the typical weeds. *Sorghum sudanense* (Piper.) Stapf. had a relatively lower tolerance to the allelopathic action of *Cirsium arvense* Scop. and *Aristolochia clematites* L., while *Sorghum vulgare* var. *technicum* Körn - to the extracts from *Sorghum halepense* (L.) Pers. and *Xanthium strumarium* L. ( $p < 0.05$ ). Data from biometric measurements on seedling length make it possible to objectively evaluate differences in the initial stages of plant development depending on the type and concentration of the cold water extract used. Our results revealed that from two studied genotypes *S. sudanense* (300/43 mutant form) had higher tolerance to the allelopathic effect of studied weed species at laboratory conditions. This fact demonstrated that it is suitable for incorporation into various breeding programs as allelopathic tolerance donor. Reading of the genotype reaction to the seed germination of *Sorghum sudanense* and *Sorghum vulgare* var. *technicum* under the allelopathic influence of invasive weed species is proved as a convenient and fast method for determining the allelopathic tolerance of various species of genus *Sorghum* at different stages of the selection process.

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## EFFECT OF MICROORGANISMS ON THE GROWTH OF TOMATO SEEDLINGS

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The aim of this study was to determine the influence of some microorganisms on the development of tomato (*Solanum lycopersicum* L.) seedlings. The seeds were treated with some Plant growth-promoting (PGPR) bacterial and fungal (yeast) isolates. For the control was a used untreated seed. The seedling was grown in a peat-pearlite mixture. Some morphological and physiological indicators were followed – leaf area, shoot/root dry weight and dry matter content, chlorophyll content. It was found that the tested microorganisms stimulate plant growth in different directions. Some of the rhizobacteria enhance the growth of the aboveground part, others of the root system, and others have a complex action. The stimulation effect on the plants is enhanced by rhizobacteria Pnt 5. The stem height, leaf area, shoots /root dry weight and dry matter content increases. The total chlorophyll content increase compared to the control.

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## HERBICIDE CONTROL OF THE WEEDS IN TOMATO (*SOLANUM LYCOPERSICUM* L.)

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Effective weed control has a key role for the growth, development and productivity of tomato plants. The use of herbicides is one of the main methods of weed control. The aim of this study is to investigate a weed control system for tomatoes through the use of herbicides. In 2019-2020, on the experimental field of Agricultural University – Plovdiv, Bulgaria, a field experiment with tomato variety Opal F<sub>1</sub>, was performed. The experiment was performed by randomized block design in 4 replicates. The following herbicide treatments were studied: Dual Gold 960 EC (1.20 l ha<sup>-1</sup>), Stomp New 330 EC (4.00 l ha<sup>-1</sup>), Dual Gold 960 EC (1.20 l ha<sup>-1</sup>) + Targa Super 5 EC (1.75 l ha<sup>-1</sup>), Dual Gold 960 EC (1.20 l ha<sup>-1</sup>) + Sencor 70 WG (0.60 g ha<sup>-1</sup>), Stomp New 330 EC (4.00 l ha<sup>-1</sup>) + Targa Super 5 EC (1.75 l ha<sup>-1</sup>), Stomp New 330 EC (4.00 l ha<sup>-1</sup>) + Sencor 70 WG (0.60 g ha<sup>-1</sup>). The application with Dual Gold 960 EC and Stomp New 330 EC was done one day before planting. On the 30th day after planting was done application with Targa Super 5 EC and Sencor 70 WG. The dominant weeds on the field were *Portulaca oleracea* L., *Amaranthus retroflexus* L., *Convolvulus arvensis* L., *Sorghum halepense* (L.) Pers. developed from rhizomes, *Solanum nigrum* L. The system Stomp New 330 EC (4.00 l ha<sup>-1</sup>) + Sencor 70 WG (0.60 g ha<sup>-1</sup>) has the highest efficiency against total weeding - 93.1% efficiency against available weeds. Visual observations of phytotoxicity show that the studied herbicides have excellent selectivity for tomato Opal F<sub>1</sub>. Tested herbicide systems have some affect on the vegetative development of plants. Plant growth retardation was found in systems involving Sencor 70 WG, but the yield was not compromised. The highest yield was reported after application of Stomp New 330 EC (4.00 l ha<sup>-1</sup>) + Sencor 70 WG (0.60 g ha<sup>-1</sup>).

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## USE OF PCA (ANALYSIS OF THE MAIN COMPONENTS) IN WHEAT CULTIVATION UNDER UNCONVENTIONAL CONDITIONS

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Elements of productivity of Bulgarian wheat (*Triticum aestivum* L.) variety Venka 1, grown in 2019 and 2020 through four different regime technologies: biodynamic, biological, combination of the two technologies and control (untreated variant) were studied. It was found that there is a significant influence of factors of cultivation and year on two of the productive traits – number of spikelets and grain weight per spike, using analysis of variance. The aim of the study was to deepen the biometric analysis of the obtained results and to apply the analysis of the main components – PCA. With its help we found that a large enough part of the total variation – 99,7 % is determined by the total sum of components PC1 and PC2. Component 1 includes three of the studied traits – length of the class, number and weight of the grains, and component 2 – the height of the stem and the number of spikelets. According to the cultivation technologies, only the biological way is in the first place in the ranking of Factor 1, while the other three ways – biodynamic; a combination of both (biodynamic and biological); and the control, are combined in the second Factor. These analyzes allow to a more complete determination of which methods or group of growing methods significantly contributes to the better productive expression of a given genotype.

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THERMO-KOH PRE-TREATMENT AND CO-DIGESTION WITH PIG SLURRY IMPROVE METHANE YIELD AND DIGESTATE QUALITY FROM GIANT REED (*ARUNDO DONAX* L.)

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Biogas production from food/feed crops is no longer supported in EU. The new challenge for the CH<sub>4</sub> production in anaerobic digestion (AD) is to improve productivity using relatively quality-poor biomasses such as waste biomasses, by-products, or lignocellulosic biomass even though they require pre-treatments to enhance AD performance. To increase the total CH<sub>4</sub> production the co-digestion of substrates is mandatory. A pre-treatment is necessary when lignocellulosic biomasses are used. Removal of the lignin is necessary to increase the CH<sub>4</sub> yield from these biomasses since the lignin is recalcitrant to AD. The aim of this study was to evaluate the effects of a thermo-KOH pre-treatment of giant reed biomass, eventually followed by co-digestion with pig slurry, on CH<sub>4</sub> yield. The impact of possible inhibitors released by the pre-treatment was evaluated comparing the CH<sub>4</sub> production from unwashed and washed materials. Pre-treated biomass showed always higher CH<sub>4</sub> production than untreated; the co-digestion showed the highest CH<sub>4</sub> production levels and permitted to improve biogas production from giant reed by 2.5 folds without any change of reactor volume. As far as the specific CH<sub>4</sub> yield is concerned, the pre-treatment allowed to enhance the CH<sub>4</sub> yield by 24%, on average, in comparison with untreated biomass in mono-digestion and by 13% in co-digestion. Combined co-digestion and pre-treatment allowed to enhance specific CH<sub>4</sub> yield by 53% in comparison with untreated mono-digested biomass. No inhibitory effect of the liquid phase was detected; no washing step is necessary. Combined co-digestion and KOH-based pre-treatment improved digestate K content by 65%. In the digestates from co-digestion, a general increase in the concentration of total N and P of 1.13 and 2 times compared to the initial value, was observed. Of particular interest is the increase in lignin concentration (3 times compared to the initial concentration) since a digestate rich in lignin can be involved in the mechanism of C sequestration. Thermo-alkaline pre-treatment and co-digestion combination could effectively improve the AD process in agreement with the possibility to increase the CH<sub>4</sub> production from alternative biomasses using the size of the current reactors.

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## CHARACTERIZATION AND COMPARISON OF ESSENTIAL OIL COMPOSITION FROM 11 *CANNABIS SATIVA* GENOTYPES FROM TWO CULTIVATION SEASONS

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*Cannabis* is an annual plant genus, and together with *Humulus lupulus* L. (Hop), it belongs to the family of *Cannabaceae*. The species is known for its high content of beneficial ingredients, such as biologically active metabolites, for its biomass constituents, like natural textile fibres, and for the seeds, used as a source of nutrition, which have been harvested by humans for thousands of years. Hemp usually refers to *Cannabis* plants low in THC, which have long been selected for fiber or seeds production. Hemp female flowers, for long time neglected and considered a waste material, are now emerging as real “chemical factories” of bioactive compounds, mostly cannabinoids and terpenoids, as with inflorescences of drug-type *Cannabis* genotypes. Hundreds of different terpenoids with numerous pharmaceutical applications are contained in cannabis resin, and they are also responsible for its different fragrance attributes, determining consumers’ preferences. Many of these metabolites have not been conclusively identified, and terpene composition is still a phenotypic trait showing large variation across different *Cannabis* strains. As a consequence of its large genetic diversity (mostly due to high heterozygosity and wind-pollination), *Cannabis* still needs research efforts to reach a level of standardization similar to that achieved for other crops. A complete set of biochemical data for one or a few genotypes can support the development of reproducible and well-defined varieties, with a special emphasis on European landraces, which are still under-sampled. With this aim, we evaluated Essential Oil (EO) yield and chemical composition from eleven European hemp genotypes, cultivated in the same area for two consecutive years, belonging to three different chemotypes (III, IV, and V). The cultivation in open field included five monoecious (Uso-31, Carmaleonte, Codimono, Futura 75, Felina 32) and six dioecious (Bernabeo, Carmagnola, CS, Fibranova, Fibrante, Eletta Campana) genotypes. The EO was obtained by hydrodistillation and its composition was analysed by GC-MS, then subjected to multivariate statistical analysis. 116 compounds were identified in EOs in both years and the extraction yield ranged from 0.03 to 0.12% w/w for the 2019, and from 0.03 to 0.23% w/w for the 2020 samples. Sesquiterpenes emerged as the main class of compounds. Conversely to the hydrocarbon forms, oxygenated sesquiterpenes were significantly more abundant in all the 2019 samples compared to those of 2020. Cannabinoids were also found, with cannabidiol as the main compound. In general, cannabinoids resulted meaningfully higher in 2020 samples. The statistical distribution of the samples was performed on the complete chemical composition of the EOs and evidenced a partition based on the year of cultivation. Also, EO yield seemed to be affected by years of cultivation, with a significant variation evidenced also among the different genotypes. In general, all the 2020 samples were characterized by a significantly higher EOs extraction yield than the 2019 ones, suggesting that the climatic conditions that occurred in the two growing seasons contributed significantly to the different EO compositions and yields.

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# **ABSTRACTS MEDICINAL & NUTRACEUTICAL PLANTS DIVISION**

**ORAL + KEYNOTE PRESENTATIONS**

**CHAIR**

**DIANA JASSO DE RODRIGUEZ, UNIVERSIDAD AUTONOMA AGRARIA ANTONIO  
NARRO, SANTILLO, MEXICO**

## DATA-DRIVEN MEDICINAL AND AROMATIC PLANT (MAP) VALUE CHAIN

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The Medicinal and Aromatic Plant (MAP) value chain encompasses all production/collection and processing steps to produce herbal medicinal products and sophisticated phytopharmaceuticals: selection of plant raw materials, cultivation and/or wild-harvest, postharvest handling (drying and dry herb processing), distillation or extraction, purification/bio-refining (and/or isolation), product formulation and packaging. It usually consists of a set of interdependent enterprises with complementary activities (e.g. production, processing, trading and end-use industry). The core actors in the MAP value chain typically include input suppliers, farm machinery manufacturers and/or technology providers, primary producers (wild-collectors and/or herb growers), primary and secondary processors (drying and extraction), wholesalers (agents or traders), service providers (quality assurance, certification) and end-use industries. In recent years, the MAP domain, has also entered an era of digitally enabled processes, where data can be generated during all operations along the value chain i.e., on-farm and beyond the farm gate into the distribution and processing sectors, to the end-user and valorization. All these processes have significant benefits, when they follow a data-driven approach under the condition that the systems, tools, and techniques used have been designed to handle the volume and foremost the variety of the data. Rapid advances in computing technologies are leading to radical transformations across a multitude of industry sectors. Data analytics, machine learning, and artificial intelligence offer new solutions to challenges in sectors including agriculture. Internet of Things (IoT) and digital predictive technologies (e.g. cloud computing) as the new frontier that can offer real-time information flow, can be used as a very valuable tool to enable plant material traceability, product verification and implement transport logistics. It also offers new technologies to gather massive volumes of data from vast networks in real-time, transmit these data to central storage (cloud), develop large data banks, undertake data analytics which deliver mined and collated data to the relevant stakeholders in real-time. It also provides historic records that can be very valuable in crisis situations or a plant raw material recall. The further development of data infrastructures (platforms and standards) will enhance the utilization of the data collected to deliver knowledge-based support for the industry, regulators and consumer as well as solutions to provide real time data to industry stakeholders.

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## CURRENT RESEARCH ON DRYING OF MEDICINAL AND AROMATIC PLANTS (MAPs) BELONGING TO LAMIACEAE FAMILY

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In the last decade, there is an increasing demand for the production of medicinal and aromatic plants (MAPs) due to their potential use by both the pharmaceutical and food industries. Lamiaceae is one of the most economically important plant families including about 236 genera and 6900-7200 species. Plants belonging to the Lamiaceae family are cultivated for multiple uses such as teas, herbal medicines, cosmetics, supplements, and functional food. Spearmint (*Mentha spicata* L.), peppermint (*Mentha piperita* L.), oregano (*Origanum vulgare* L.), lavender (*Lavandula spica* L.), thyme (*Thymus vulgaris* L.), marjoram (*Origanum majorana* L.), lemon balm (*Melissa officinalis* L.), basil (*Ocimum basilicum* L.), hyssop (*Hyssopus officinalis* L.), and sage (*Salvia officinalis* L.) are some of the most popular plants belonging to the Lamiaceae family. Due to their high-water content (up to 75% wet basis) MAPs are perishable. Therefore, postharvest processing is required to prolong their storage life and maintain their quality attributes. However, non-optimum drying conditions may result in quality deterioration of herbal drugs. For various MAPs, the minimum quality requirements of dried herbal drugs are postulated in the European Pharmacopoeia (Ph. Eur.). The current study aims to review the most recent studies comparing different postharvest processing methods for drying selected herbs belonging to the Lamiaceae family. The effects of different conventional (e.g., hot air-drying, sun-drying, and shade-drying,) and non-conventional drying methods (freeze-drying, microwave-drying, vacuum-drying, infrared-drying combined with hot air-drying, and ultrasound-assisted hot air-drying) on the physical and chemical parameters of the Lamiaceae family plants have been recently investigated. The drying technique has a significant impact on the processing time, energy consumption, and dried herb quality attributes (e.g., color, essential oil content, and active ingredients). Even though freeze-drying has been known for maintaining the quality of herbal drugs, it is considered an expensive drying technique due to its high energy requirements. Hot air-drying has been traditionally used as the main drying technique for MAPs due to its low capital investment and lack of complexity. The main disadvantage of hot air-drying is the prolonged drying times at high temperatures which may lead to quality deterioration of the dried herb (e.g., enzymatic browning, degradation of heat-, or oxygen-sensitive active ingredients). For instance, studies in lemon balm have shown that low temperatures during hot air-drying facilitated minimal essential oil and color losses, that were greater at higher temperatures. However, loss of lemon balm bioactive compounds (e.g., rosmarinic acid) has been reported even at low drying temperatures and significantly increases as the drying temperature rises. The duration of the hot air-drying can be shorter when combined with infrared-drying or ultrasound power. Microwave-drying is a non-conventional technique which results in significantly reduced drying times and energy consumption compared to hot air-drying. However, microwave-drying may have different effects on the essential oil contents of various herbs (e.g., lavender, sage, basil, and hedge nettle). Future studies are encouraged to optimize and compare the effects of different drying methods considering both physical and chemical parameters of herbs belonging to the Lamiaceae family. Additionally, future techno-economic studies for each drying process (or their combinations) and plant species are encouraged to be conducted since there is a lack of information in the current literature.

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## CHARACTERIZATION OF BIOACTIVE VOLATILE BASED PRODUCTS AND ITS EFFICIENCY IN APHID POPULATION CONTROL ON A PEPPER CROP

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The “farm to fork strategy” for a fair, healthy and environmentally-friendly food system of the European Union states that there is an urgent need to reduce dependency on pesticides. Among the steps to be taken is the one to promote greater use of low-risk pesticides, for instance, those based on botanical extracts. In recent years, aphids have emerged as important pests of pepper in Southeast Spain, causing direct and indirect (due to virus transmission) damages to the crop. In previous work we selected some bioactive compounds against *Myzus persicae* Sulzer (Hemiptera: Aphididae), for having repellent or insecticidal properties, namely: citral, (*E*)-anethole, farnesol, cis-jasmone and lemon essential oil and mixed a combined product based on the five of them in equal amounts. Three formulations were prepared using a high-speed rotor IKA Labor Pilot (10 min, 7940 rpm, 15°C), the first one with Tween80 1:2, the second one with soya lecithin 1:1 and the third one with soya lecithin and sunflower oil (*Helianthus annuus* L.) 1:1:1. The oil in water nanoemulsions at 1% were tested in a laboratory study with Petri dishes, using a computer controlled spraying apparatus (CCSA, Burkard Ltd.) at 5 psi and 6r (equivalent application of 200 l/ha). The mean efficacy was 45%, 71% and 63% respectively with an LSD statistically significant at 5% comparing the first two formulations. A field experiment (in two pepper greenhouses) followed in April 2020 in which a reduction in aphid populations only occurred by spraying the product formulated with soya lecithin. Phytotoxicity was also observed but was less pronounced in the formulation that contained sunflower oil. Nanoemulsions were characterized using a Zetasizer Nano ZS (Malvern Panalytical Ltd.) and it was found a polydispersion of 2-3 populations of particles ranging from 15-341 nm in size with the 1:2 formulation and bigger sizes (250-438 nm) with the 1:1:1 formulation but more stable (Z potential = -28.15 mV). The application of ultrasounds (Digital Branson Ultrasonicator 4500 with a tapered disruptor horn ½” 400 W 15 sec. pulses for 5 min and 30% amplitude) reduces the Z-average to 100 nm in the mixture product with stability at least for 14 days. In addition, it was observed that cis-jasmone gave a good emulsion with Tween80 (1:2), with 15 nm particles and a Polydispersity Index (PdI) of 0,289 (in comparison with 353 nm and PdI = 0,486 of the formulation 1:1:1). Another field experiment was carried out in 2021, in two pepper greenhouses at Torreblanca Exp. Stat. in Murcia (Spain), infested with aphids (initial populations of 20-50 aphids/plant). The experiment was repeated twice, in February and in April, spraying the plants sequentially (every 4 days) with the following treatments: a) mixture product at 0.5% and 1:1:1 formulation applied to the air (not directly to the plants to avoid phytotoxicity), b) cottonseed (*Gossypium*), (oil and soap at 3% (Feb.)-1.5% (Apr.) (directly to the plants) and c) cis-jasmone at 0.25% and 1:2 formulation (directly to the plants). The treatment with the mixture of bioactive volatiles (a) was not better than that of the vegetable fixed oil (b). In February, the Instantaneous Rate of Growth Increase (ri) of aphid populations showed a significant reduction when the plants were sprayed with the cottonseed oil: Wilcoxon test W=64, p=0.000931\*\*\* and ANOVA (F=48.8, p<0.001); in April, there was not a statistically significant difference but it was close to 5%: (W=49, p=0.083 and F=4.12, p=0.0633). In addition, cis-jasmone spraying (c) in February (although with data variability) produced aphid mortality as well (W=7, p<0.0322\* and F=5.28, p<0.05). In conclusion, more research is needed on the formulation and method of application of the bioactive volatile product to produce better results in terms of aphid control and to get rid of the phytotoxic side effect being observed in the formulations tested.

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CONTENTS OF CERTAIN MACRO, MICRO ELEMENTS AND BIOLOGICALLY  
ACTIVE SUBSTANCES IN THE FRUIT OF THE GOJI BERRY VARIETIES  
(*LYCIUM BARBARUM* L.)

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Goji berry, the botanical specie *Lycium Barbarum* L. belongs to the family *Solanaceae*. The plant has a long-standing application in East Asia, and now in many other places because of its medicinal and nutritional properties. At the end of XX century, the need of healthy products makes the cultivation of the plant growing - first in Asia and then in other continents. The fruits have high biological matter confirmed in numerous clinical studies. They are rich of many, important for the humans, minerals (Ca, Mg, K, Na, P, Fe, Zn, Cu, Se, Ge, Pb, Ni, Cd, As, Bi, Hg) and 11 amino acids. Nutritionists describe Goji berry as an "exotic super food" as a result of its high content of polysaccharides, vitamins and carotenoids. A connection has been established between the quantity and quality of fruits and the area of cultivation, as well as the dependence of quality on the genetic benefits of the varieties. The aim of the study was to determinate the content of biologically active substances, macro and micro elements in the fruit of four in vitro propagated goji berry varieties (JB1, JB 2, JB 4, JB 10) of the species (*Lycium barbarum* L.) grown in South Bulgaria. In the Experimental Base, in the village of Brestnik, which is located near to FCM (Factory for colorful metals) are studied four, in vitro multiplied varieties of Goji berry (GB1, GB2, GB4 and GB10). The plants are formed as trees and grown under drip irrigation. The climate in Plovdiv is typical for the temperate-continental climate zone with 3900 ° active temperature sum and with precipitation in the amount of about 515 mm. The fruits are analyzed for biologically active substances, macro and micro elements. There were found differences in the content of some biologically active substances, in the fruits of different cultivars, as well in their antioxidant activity. The obtained results show that the macronutrients (K, P, Mg and Ca) predominate in the fruits of Goji berries, followed by Zn and Fe. The content of Cu and Mn is significantly lower. Goji berries also contain the toxic metals Pb and Cd, but do not exceed the maximum levels of the EU Commission Regulation 1881/2006 for setting maximum levels for certain contaminants in the food.

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USE OF SOME PRODUCTS FOR FOLIAR APPLICATION FOR IMPROVING THE  
PRODUCTIVITY, QUALITY AND ESSENTIAL OIL CONTENT OF CORIANDER SEEDS  
(*CORIANDRUM SATIVUM* L.) UNDER SOUTH-EASTERN BULGARIA CONDITIONS

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Coriander (*Coriandrum sativum* L.) is important medicinal and aromatic crop, cultivated for the fresh green herb, the fruits as well as for production of essential oil. The requirement for the realization of the productive potential of the crop, is the application of suitable agrotechnical practices in its cultivation. The deficiency of nutrients is one of the main factors, which can negatively affect the plant growth. The application of biostimulators improves plant development and growth, increases the yields, as well as the content of essential oil, and enables a more sustainable agricultural production. With the aim to investigate the effect of some foliar fertilizers and biostimulators on the elements of productivity, seed yield and the essential oil content in Lozen 1 coriander cultivar, a field trial was carried out on leached Smolnitsa soil type in the region of the village Zhrebino, South-Eastern Bulgaria in the period 2018-2020. The experiment was arranged according to the block-plot design method in four replications with a plot size of 15 m<sup>2</sup>, after predecessor wheat. The following foliar fertilizers and biostimulators were applied at the respective rates: Variant 1 – Humustim – 0,4 l/ha; Variant 2 – Fertigrain foliar – 1 l/ha; Variant 3 – Tecamine max – 1,5 l/ha; Variant 4 – Masterblend – 2,5 kg/ha; Variant 5 – Poly Plant – 1 kg/ha. The application took place at the end of buttoning and the beginning of flowering stage. The variants were compared to an untreated control to follow out the effect of those products on the elements of productivity, seed yield and essential oil content. The experiment was carried out following the adopted cultivation technology. The structural elements of the yield were determined after analysing 50 plants from one square meter. The experimental data was processed according to the Anova Method of dispersion analysis for the purpose of determining the quantity dependence between the studied indicators and the differences between the variants were determined by means of the Duncan's Multiple Range Test. The basic climatic factors, determining coriander growth, development and productivity were temperatures and precipitation, their combination and distribution during vegetative period. The structural elements of the yield, i.e., the number of umbels per plant, the number of seeds per plant, the seed weight per plant and the 1000 seed weight in the treated variants exceeded the untreated control by 5-16%, 6-16,8%, 4-17,2% and 7,5-13,5%, respectively. The treatment of coriander with leaf fertilizer masterblend in dose of 2,5 kg/ha resulted in an average yield increase of 12,5 % of the crop seed for the investigated period compared to the untreated control. After treatment with the foliar applied products an increase in the essential oil content from 2,9 to 9,6% was established, as the highest values were obtained when using the biostimulator Fertigrain – 1,26% and the foliar fertilizer Masterblend – 1,25%. The used products stimulated the biological potential of the tested cultivar and could be recommended for further investigations by other essential oil crops.

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## ANTIOXIDANT AND ANTIPROLIFERATIVE ACTIVITIES OF *FLOURENSIA* SPP.

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Cancer is a serious public health problem in the world and represents the second leading cause of death. In 2020, cancer occurred in 19.3 million people, killing 10 million of them. Lung cancer in Mexico is the third leading cause of mortality, after cardiovascular diseases and diabetes. At present the chemical treatments used against cancer show little selectivity and adverse reactions. The extracts of *Flourensia microphylla*, an endemic plant of the arid zones of Coahuila, Mexico, have shown antioxidant activity, as well as an anti-inflammatory and antiproliferative effects in HT-29 colon cancer cells, inducing apoptosis. Due to the above and considering that lung cancer is a type of cancer with a great impact on the population of México and the world, the objectives of this research were: to determine the antioxidant and antiproliferative activities of extracts of *Flourensia cernua*, *F. microphylla* and *F. retinophylla* in A-549 lung cancer cells and non-cancer ARPE-19 cells. The ethanol extracts were obtained from the dried and ground leaves of the three species in a soxhlet extractor. The antioxidant activity was evaluated by DPPH, ABTS and FRAP tests. The total phenolic content (TPC) was determined by the Folin Ciocalteu method and the main phenolic compounds were identified by UPLC-MS analysis. The antiproliferative activity was determined by the MTT assay. The extract that presented the highest antioxidant activity by all the evaluated methods was *F. microphylla* (DPPH IC<sub>50</sub>: 20.4 µg / mL; ABTS IC<sub>50</sub>: 6.7 µg / mL; FRAP: 3122.9 µM Fe (II) / g D. W.). Regarding the phenol content, the order from highest to lowest content was *F. microphylla* > *F. retinophylla* > *F. cernua*. The three extracts evaluated presented antiproliferative effect on A-549 lung cancer cells and the ARPE-19 cells, and the order was the same for both cells from highest to lowest: *F. cernua* > *F. microphylla* > *F. retinophylla*. These results provide very promising new information for possible lung cancer treatment applications with extracts of *Flourensia* spp.

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## SEASONAL EFFECT ON THE ANTIPROLIFERATIVE AND ANTI-INFLAMMATORY ACTIVITIES OF *BURSERA MICROPHYLLA*

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*Bursera microphylla* A. Gray (Burseraceae), commonly known as “torote blanco” is a medicinal plant largely distributed in the Sonoran Desert in Mexico. The natives use the different parts of the plant (leaves, fruits, bark and exudate) for the empirical treatment of headache, infections and such as a potent anti-inflammatory. Previous studies on *B. microphylla* resin have allowed the isolation and chemical characterization of diverse antiproliferative metabolites such as terpenoids and lignans; in addition, in our research group we have demonstrated that the leaves and fruits of *B. microphylla* have high antioxidant potential. For these reasons, *B. microphylla* is considered a good alternative for the generation of new products with pharmacological properties. The study of seasonality is essential to optimize the parameters related to the large-scale production of herbal drugs. To date, the antiproliferative and anti-inflammatory effects of stems, leaves, and fruits of *B. microphylla* are unknown; in addition, the seasonality effect on the biological properties of *B. microphylla* have not been investigated yet. Therefore, the main goal of this research was to determinate the seasonal effect on the antiproliferative and anti-inflammatory activities of extracts from leaves, fruits, and stems of *B. microphylla*. Plant specimens were collected in the state of Sonora, Mexico during 2019-2020. Leaves, fruits and stems were finely milled and extracted with EtOH (1:10 w/v) to generate ethanolic extracts. The antiproliferative activity of the ethanolic extracts against the cancerous (HeLa, A549, C33-A and MDA-MB-231) and the non-cancerous ARPE-19 cell lines was evaluated by MTT assay. The anti-inflammatory activity of the ethanolic extracts was measured through the quantification of nitric oxide (NO) levels produced in the supernatants of RAW 264.7 cells activated with LPS. In general, all the extracts reduced the proliferation of cancer cells with IC<sub>50</sub> values <30 µg/mL, which in accordance with the established by the NCI from USA, these extracts can be classified as good antiproliferative agents. In addition, the extracts were able to induce morphological changes in cells; contraction, cell rounding, membrane “blebs” formation and apoptotic bodies were observed after the treatment. The above suggests that the extracts inhibit the cell proliferation through the mechanisms associated with the apoptosis induction and the cell cycle arrest. The NO production was also reduced by the extracts, being generally observed that the extracts from the stems are the most effective. In conclusion, our results suggest that extracts can be considered as a good option for the search for new antiproliferative and anti-inflammatory agents, however, additional studies on the chemical composition of the extracts and the mechanisms of action involved are necessary to have a better understanding.

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## BIOACTIVE COMPOUNDS IN FRUITS OF WILD MAQUI IN DIFFERENT RIPENING STAGES AND ENVIRONMENTS

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The demand for food and natural products that present healthy properties, has increased in the last years. Several studies in wild fruits, particularly in native berries, have shown a high anthocyanin content and antioxidant potential influenced mainly by abiotic stress. Maqui (*Aristotelia chilensis* (Mol.) Stuntz), a Chilean native berry, has been referred as a “superfruits” due to their phytochemical composition and antioxidant activity. Bioactive compound content in wild maqui collections are influenced by collection time, ecotypes and environments (locations). The variation of the polyphenolic compounds, the antioxidant capacity and the fruits quality were evaluated in three stages of maturity (green (unripe), redish (pre-ripening) and dark violet (ripe) stages) and from three locations (Coihueco, Tregualemu and Cayumanque) in the Central Valley in Chile, exposed to a Mediterranean and Temperate climate. Maqui fruits were collected in summer during two consecutive seasons, in 10 selected stands. Significant differences for polyphenolic compounds content were observed according to the state of maturity and environments, reaching the mature state the higher values in the 3 locations (1,096 – 2,291 mg EGA 100 g<sup>-1</sup> FW). Total anthocyanin contents ranged from 1.3 to 1,130 mg eq cyanidin 3-glucoside 100 g<sup>-1</sup> FW for green and ripe stage, from foothills (Coihueco). The antioxidant capacity in Coihueco had a higher value according to DPPH assay in mature fruits (8,767 μmol Trolox 100g<sup>-1</sup> FW); however in Tregualemu and Cayumanque the higher values were obtained in the pre-ripening state (5,230 and 9,961 μmol Eq Trolox 100 g<sup>-1</sup> FW, respectively). In the three locations (different environments), the ripe maqui fruits had a higher pH, ranged from 3.26 to 3.45. The soluble solids content was higher in the mature stage in Tregualemu and Cayumanque (20 and 17.3 °Brix, respectively); however, in Coihueco it did not increase from the redish to mature stage (9.5 to 11 °Brix). Seasonal and environmental variations in bioactive compounds and chemical characteristics in wild fruit, make necessary to regulate and cluster the collected fruit among edaphoclimatic conditions for the food and pharmaceutical industry with homogeneous raw materials and sustainably production. On the other hand, valuable data for establishing production practices are also obtained in order to provide standardized fruits quality according to locations.

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## AGRONOMIC AND NUTRACEUTICAL CHARACTERISTICS OF STINGING NETTLE GROWN UNDER ORGANIC FARMING IN ITALIAN ENVIRONMENTS

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Stinging nettle (*Urtica dioica* L.) is a spontaneous plant very widespread in all temperate regions that is used in many sectors including food. As a multipurpose crop easily adaptable to various agroclimatic conditions, stinging nettle may fit well in the current scheme of crop diversification as an alternative crop for the enhancement of agricultural environments. Due to the eco-sustainability of cultivation (its rusticity makes it particularly suitable for agricultural systems with low environmental impact), the health characteristics (interesting for innovative sectors such as novel and functional foods) and the potential uses of by-products cultivation (as fertilizers and repellents) a complete supply chain, from field production to processing, may be envisaged for stinging nettle. In this work stinging nettle plants, organically grown in three different Italian farms, were evaluated in terms of biomass production, nutraceutical characteristics and organoleptic performance of derived food-products. Trials were carried out at three organic farms of the Emilia Romagna region, Italy, located at Tresigallo (44°49'26"N, 11°54'42"E, -1 m above sea level (a.s.l.)), Ozzano dell'Emilia (44°23'16" N, 11°25'54"E, 110 m a.s.l.) and Lizzano in Belvedere (44°12'53" N, 10°51'50"E, 555 m a.s.l.) during the 2020 growing season. Seeds of stinging nettle were sown in nursery beds at the end of April 2020. After 40 days of growth the plants were transplanted in the experimental fields at a density of 6,66 plant/m<sup>2</sup>. Plants were harvested in the last week of September 2020 and representative samples of leaf biomass were evaluated for: free, bound, total polyphenols and flavonoids, and antioxidant activity. Sensory analyses were carried out on bakery products made with different percentages of wheat and stinging nettle flours. The highest biomass yield was obtained at Tresigallo (3067.8 Kg/ha) whereas at Lizzano (471.2 Kg/ha) and Ozzano (242.1 Kg/ha) yields were much lower. No significant differences were observed among locations as regards the dry matter content. The total polyphenol contents varied from 514.22 to 353.19 mg of gallic acid equivalents (GAE) per 100 g of fresh weight (FW) recorded at Tresigallo and Ozzano, respectively. As regards the total flavonoid content values ranged from 348,22 to 273,84 mg GAE/100 g FW at Lizzano and Tresigallo, respectively. No significant differences were observed among locations for total polyphenol and flavonoid contents. Instead, significant variation in the relative abundance of the polyphenol fractions (free and bound) were observed, with the free fraction accounting for 94.1% (Tresigallo), 87,5% (Lizzano) and 80.1% (Ozzano). The highest values of antioxidant activity (FRAP assay) were observed for plants grown at Lizzano and Tresigallo (105,69 and 78,20 mmol/100 g, respectively), which reflected the highest abundance of free polyphenols. On the other hand, plants grown at Ozzano were richer in bound phenolic components (polyphenols and flavonoids) which concur less at the antioxidant activity of the extracts. As regards the sensory attractiveness of the derived products, the most attractive in terms of taste, scent, colour and texture was the product made with 3% of stinging nettle flour. The preliminary results of the present study showed the possibility of cultivating stinging nettle in Italian organic farming systems. Particularly, the growing location seems to affect biomass yield and nutraceutical composition of the aerial part of the plants. Additional data coming from other two years of experimentation (2021 and 2022) will give major insight on the correlation between environmental conditions, agronomic performance and nutraceutical value of organically-grown stinging nettle.

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MICROWAVE-ASSISTED EXTRACTION OF HEMP (*CANNABIS SATIVA* L.) TO RECOVER  
THREE VALUABLE FRACTIONS (ESSENTIAL OIL, PHENOLIC COMPOUNDS AND  
CANNABINOID): A CENTRAL COMPOSITE DESIGN OPTIMIZATION STUDY

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Industrial hemp (*Cannabis sativa* L.) is a multipurpose crop representing an interesting source of valuable products, which could be exploited in several industrial applications. Among them, hemp essential oil is still under-studied and the search for innovative, effective and green methods for its extraction is growing. On this basis, in the present work, hemp Futura 75 was subjected to microwave-assisted extraction (MAE), using a central composite design approach. After the essential oil distillation, two valuable by-products remaining in the reactor, namely the residual biomass and water enriched in hydrophilic compounds, were recovered and studied. The design variables analyzed were the microwave irradiation power, the extraction time and the amount of water added to hemp. The aim of this research was to evaluate how these experimental parameters can affect the essential oil yield and composition, together with the yield and antioxidant properties of the aqueous residue and finally the cannabidiol (CBD) content in residual biomass. Gas chromatography coupled to mass spectrometry was performed to study the chemical composition of essential oils obtained by MAE, while gas chromatography coupled to flame ionization detection was used to quantify the main terpenoids in essential oils, and the CBD content both in essential oils and residual biomass. The aqueous residues were evaluated in terms of polyphenols, flavonoids content, radical scavenging activity and antidiabetic properties. High performance liquid chromatography-mass spectrometry analysis evidenced the abundance of flavone glycosides in these products. Regarding the essential oil analysis, the responses yield and content of CBD and some other compounds were well described by the mathematical models applied. Also, in the case of the aqueous residues, the yield, polyphenols and flavonoids content and antioxidant activity, as the four responses analyzed, suitably fit the models. Aqueous residues were also interesting from a pharmaceutical point of view as they were able to inhibit  $\alpha$ -glucosidase and the formation of advanced glycation end products. On the other hand, there were no relationships between the CBD content in the residual biomass and the MAE operative conditions, probably due to the samples intrinsic variability. A multiple optimizing procedure, the desirability, was employed to identify the most suitable experimental parameters, able to maximize all the satisfactory responses at the same time. In particular, high microwave power and long extraction time are needed to enhance the yield and chemical profile of essential oils, along with medium-high amount of water, required for the improvement of the aqueous extract yield and antioxidant capacity. The results of this work supported the valorization of industrial hemp by-products as sources of bioactive compounds to be exploited in pharmaceutical and nutraceutical fields.

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## HEMP FUNGICIDE SEED TREATMENTS TO CONTROL DAMPING-OFF

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There is a renewed grower interest in hemp following the passage of the 2018 US Farm Bill, that removed the crop from schedule I of the Controlled Substances Act and allowed for hemp production with <0.3% THC. Hemp (*Cannabis sativa* L.) may be grown as crop for fiber, grain or as a nutraceutical for cannabidiol (CBD) production. There is also a need for labeled seed treatments for efficient early-season pest management, including control of soil-borne pathogens. Organically approved treatments would be ideal so that they could be used for both conventional and organic production systems. The purpose of this research was to conduct preliminary efficacy studies in 2020 on biological, biochemical, and chemical fungicide seed treatments for the management of damping-off caused by several pathogens (*Pythium*, *Fusarium*, and *Rhizoctonia*) and to enhance stand establishment. A dual-purpose (fiber + grain) was used for field trials in 2020 and field data are summarized from New York and North Dakota. In 2020, a dual-purpose, seed lot 'Anka', was treated in the senior author's lab at Cornell Agri Tech using a laboratory-scale rotary pan coater. Five biological treatments were tested: K5, *Trichoderma atroviride* from ABM, Amplitude ST, *Bacillus nakamurai* strain F727 from Marrone Bio Innovations, Bio Seed contained five biologicals from Ag Biotech, Phyter, *Clonostachys rosea* from Endo Plant Health Inc, Varnimo, *Bacillus amyloliquifaciens* from LidoChem. A phosphite treatment, Prudent 44 + Nutrol from LidoChem and ProBio Ultim, an organic copper hydroxide from Germain's were biochemical seed treatments. Chemical fungicides were Apron XL and Maxim 4FS from Syngenta. In general, all biological seed treatments did not improve plant stands compared to the non-treated controls. The organic copper and phosphite seed treatment had acceptable efficacy along with chemical seed treatments. Based on efficacy and registrant willingness to pursue a label on hemp, research was conducted on management of damping-off of transplants in the greenhouse in 2021. Seed treatments tested were Prudent 44 + Nutrol and ProBio Ultim. Another organic copper, Americop 40 DF from Industrias Quimicas del Valles, S.A. was included. Chemical seed treatments were not tested as the registrant was not interested to pursue a hemp label. Preliminary studies were conducted with Anka in 2021 and a laboratory germination test was conducted and the germination of the three seed treatments were not different than the control. A lab bioassay that incorporated naturally pathogen infested soil revealed that all three seed treatments provided protection from damping-off. A CBD variety, Boax 2.0 will be used in continued greenhouse trials. Based on efficacy tests and registrant's interest in hemp, an organic copper seed treatment has great potential for future labeling.

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# **ABSTRACTS MEDICINAL & NUTRACEUTICAL PLANTS DIVISION**

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**DIANA JASSO DE RODRIGUEZ, UNIVERSIDAD AUTONOMA AGRARIA ANTONIO  
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## SAFFRON AQUAPONICS CULTIVATION TECHNIQUES: PRODUCTIVITY EVALUATION

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As the world's population grows, the demands for increased food production expand, and as the stresses on resources such as land, water and nutrients become ever greater. Therefore, it is important to identify mechanisms and solutions that can minimize the problems that may occur in the entire primary sector value chain, from the production, distribution and commercialization, respectively, of food to people. Aquaponics, that integrates Recirculating Aquaculture System (RAS) and hydroponics, represents a solution of developing innovative and sustainable food sources and could improve food security and resilience. Stigma of saffron (*Crocus sativus* L.) is considered to be one of the most expensive spices in the world. Since antiquity, saffron has been used as a food spice, colorant, and to treat various disorders. Currently Saffron is studied for its active compounds, characterized by antioxidant, anticancer, and anti-inflammatory activities and memory-improving properties. The aim of this study was to understand the advantages and disadvantages of saffron production in aquaponic system in association with eels rearing in recirculating aquaculture system (RAS), under different illumination systems (Led and Neon). In the present study, a completely randomized plot trial, composed by two experimental decoupled aquaponics system, was set up in growing chambers located at the University of Bologna. In each system corms, at plant density of 106 corms/m<sup>2</sup>, were planted in October 2020 before flowering induction, and maintained until the end of the crop cycle. At the start of bloom stage, flowers were harvested and analyzed. Number of flowers/corm, flower fresh and dry weight, stigmas fresh and dry weight were collected before and after drying process. Samples collected from flowers parts harvested during the flowering phases were analyzed for polyphenols and flavonoids contents and for their antioxidant capacity, by using FRAP and DPPH assays. According to ISO 3632:2003 drug samples were analyzed for picrocrocin, safranal and crocin content. Plant growth parameters such as shoots number, leaf length, leaf number were analyzed monthly. At the end of the crop cycle corms were harvest and maintained in storage condition. Daughters corms numbers, corms diameter and weight were detected. Results showed that high plant density used allowed a higher saffron dried spice production in aquaponics system in front of land cultivation methods (50-75 corms/m<sup>2</sup> plants density). There wasn't significant differences in Led or Neon production of dried flowers and spice. With regard to quality production, system with Led light showed spice with a major content in polyphenols and flavonoids in dried stigmas and dried flowers. All the products from different light trial showed high content in picrocrocin, safranal and crocin, and thus can be classified in quality category I. In conclusion aquaponics showed interesting opportunity for the production of saffron dried spice and flowers. Waste production in the Eels rearing system shows to be able to fertilize saffron and obtain 0 residue products. However the entire crop cycle needs more research about the nutritional aspect in the vegetative and reproductive phase. With regard to the illumination system, Led light shows higher plants performance and better quality, as well as economic and environmental benefits.

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## COMPOSITION AND ANTIFUNGAL ACTIVITY OF THE ESSENTIAL OILS HYDRODISTILLED FROM THREE ACCESSIONS OF PASTINOCELLO CARROT

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Pastinocello carrot (*Daucus carota* L. ssp. *major*, Apiaceae family) is a wild-growing variety of the commercial carrot, whose natural habitat is located in the Northern Versilia (Tuscany, Italy) mountain areas. It is now listed among the species facing extinction risk, as its production has mostly been discontinued due to the massive focus on the cultivation of the commercial carrot variety, characterized by higher productivity and a sweeter taste. Nowadays, Pastinocello is only cultivated by “guardian farmers”, and its seeds are preserved in the germplasm bank of the Dept. of Agriculture, Food and Environment (DAFE) of the University of Pisa, Italy. The decline of local genetic resources like Pastinocello, mainly due to anthropogenic factors, is at the core of the loss of local biodiversity, which threatens the resilience of the environmental niches to the pedo-climatic changes occurring at an ever-increasing rate. The aim of this study was the re-evaluation of Pastinocello carrot, whose characteristics are believed to be still intact since its first human-driven selection, occurred during the Roman Empire Era. Three Pastinocello landraces (landrace L24, from *ex-situ* cultivation in Rispeccia, Grosseto, Italy; landraces L281 and L305, from *in-situ* cultivation at Cardoso di Stazzema, Lucca, Italy) have been evaluated for i) bio-agronomic characteristics, ii) inflorescence production and essential oils (EOs) yield, iii) EO composition, and iv) EOs antifungal activity vs phytopathogenic and beneficial isolates. All the three landraces were grown at the experimental fields of DAFE. The highest germination rate (91%), and the lowest germination time (7 days) was found for L24, whose seeds had an average weight of 1.4 g. The lowest germination rate (21.3%), lowest mean seed weight (0.84 g), and longest germination time (11 days) were found for L281. Although L281 exhibited the shortest plants (85 cm, vs 98 cm for L24 and 110 cm for L205), its inflorescence yield resulted the highest (20.4 g m<sup>-2</sup> on dry weight basis, vs 17.7 g m<sup>-2</sup> for L305 and 15.1 g m<sup>-2</sup> for L24). The EO yield (% w/w) did not show significant differences among the three landraces (0.56% L281, 0.62% L24, and 0.73% L305). All three EOs, extracted by hydrodistillation, exhibited an  $\alpha$ -pinene chemotype, as this monoterpene hydrocarbon was the most representative (26.2% in L305, 22.3% in L281, and 16.2% in L24). In L281 and L305, monoterpene hydrocarbons were the most abundant chemical group, among which sabinene also showed a noteworthy relative abundance in all samples (7.1% in L305, 6.2% in L281, and 3.15% in L24). L24 showed a composition more similar to that of commercial carrot, with the highest carotol and daucol content (6.1% and 2.69%, respectively; not detected in the other samples), and the lowest geranyl acetate relative concentration (2.7%, vs 5.53% in L281 and 15.07% in L305). The EOs were then diluted in DMSO at 5% and tested against *Colletotrichum lupini*, *Fusarium graminearum*, *F. langsethiae*, *F. oxysporum* f. sp. *lycopersici*, *Monilinia fructigena*, *Trichoderma afroharzianum* T6776, *T. gamsii* T6085 and *Verticillium dahliae*. Only the EO of L24 significantly reduced *C. lupini*, *F. oxysporum*, and *M. fructigena* growth, while no effect was registered vs *Trichoderma* spp. Given the exclusive presence of carotol and daucol in this accession, it could be hypothesized that those compounds are responsible for its antifungal activity. The lack of effect vs the two *Trichoderma* isolates - actually under evaluation as biocontrol agents - suggests a possible combined use of these compounds with the two isolates in crop protection strategies. However, not only further studies are needed for confirmation, but the whole phytocomplex-effect must also be taken into great account, as a degree of interaction and, most probably, synergism of the main chemical compounds with the lower concentrated ones is to be expected.

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PHYTOREMEDIATION POTENTIAL OF VETIVER GRASS (*CHRYSOPOGON  
ZIZANIOIDES* L.)

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Phytoremediation is an inexpensive and environmental-friendly method used for cleaning up heavy metal contaminated soils. This technology can be defined as the efficient use of plants to remove, detoxify or immobilize environmental contaminants in soils through the plants' natural, biological, chemical, or physical activities and processes. Food crops for phytoremediation are not environmentally safe as heavy metals can enter the food chain. Medical and aromatic plants can be a better choice for phytoremediation as these species are grown primarily for processing (essential oils). Thus, contamination of the food chain with heavy metals is eliminated. This research aimed to investigate the potential for using vetiver grass (*Chrysopogon zizanioides* L.) to remediate metal-polluted sites. This study was performed in industrially polluted soils containing high concentrations of Zn, Pb, and Cd, situated at different distances (0.3, 3.5, and 8.0 km) from the source of pollution - the Non-Ferrous Metal Works near Plovdiv, Bulgaria. On reaching commercial ripeness, the vetiver grass was gathered, and the contents of heavy metals in their different parts – roots, stems, and leaves were determined after microwave mineralization. Physico-chemical characterization, total, and DTPA extractable metals in rhizospheric soil samples were carried. Translocation factors (TFs) were also determined. The quantitative measurements were carried out with ICP. The essential oil of the ground vetiver roots was obtained by steam distillation in laboratory conditions. The oil analysis was performed using gas chromatography-mass spectrometry (GC/MS), and about 15 compounds were identified in oil, mainly sesquiterpenes. The main compounds of essential oil were as follows: khusimol (29.40-30,25%), valerianol (9.07-10,33%), guaiol acetate (7,83-8,15%), vetiselinol (7,27-7,84%),  $\alpha$ -vetivone (6,79-7,12%), junenol (5.87-6,36%), (e)-isovalencenol (5,95-6,42%),  $\beta$ -vetivone (5,21-5,62%), 10-epi- $\gamma$ -eudesmol (3,85-4,15%),  $\beta$ -vetivenene (3,38-3,55%),  $\beta$ -vetispirene (3,14-3,39%), 7-epi- $\alpha$ -eudesmol (3,12-3,37%), and  $\alpha$ -Amorphene (2,30-2,44%). The vetiver grass is tolerant to heavy metals and can be grown on the highly heavy metal polluted soils. Bioaccumulation factor and translocation factor values (BAF and TF < 1) were less than one suggesting low accumulation in the shoots. The plants are characterized by a low capacity to absorb and accumulate Pb, Cd, and Zn, showing no signs of toxicity (chlorosis and necrosis) in the content of 36.8 mg/kg Cd, 1158.8 mg/kg Pb, and 1526.2 mg/kg Zn in the soil. The vetiver grass can be successfully used for the phytostabilization of heavy metal polluted soils.

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## COMPOSITION AND BIOLOGICAL PROPERTIES OF *RHUS MICROPHYLLA* AND *MYRTILLOCACTUS GEOMETRIZANS* FRUIT EXTRACTS

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Extracts obtained from plants of arid zones of Mexico present a wide number of biological properties due to their interesting metabolic machinery, associated with their growth conditions (extreme weather, pathogen attack, among others). There are several methods for obtaining plant extracts, being classified into two groups: conventional (e.g., hydrodistillation, agitation, etc.) and non-conventional (e.g., ohmic heating, microwave, etc.), which differ in operating times, yields, use of solvents, among other parameters. The objective of this work was to study the effect of two extraction methods (ohmic heating-OH and conventional agitation-C) and two solvents (water and hydroalcohol solution 1:1, v/v) to obtain extracts from fruit of two arid zones plants (*Rhus microphylla*-Rm and *Myrtillocactus geometrizans*-Mg) in terms of yields, phenolic composition, and their bioactivity as antioxidant, antifungal and antiproliferative agents. Total phenolic content (TPC) was made by Folin-Ciocalteu method, the phenolic profile was identified through ultra-high performance liquid chromatography, and the antioxidant activity was measured by three different assays: DPPH (2,2-diphenyl-1-picrylhydrazil), ABTS (2,2-azinobis-(3-ethylbenzothiazoline-6-sulphonic acid), and FRAP (Ferric Reducing Ability of Plasma). The antifungal activity was carried out by microdilution method against *Rhizopus stolonifer* and *Fusarium oxysporum*, and the antiproliferative activity was evaluated through MTT assay against a non-cancerous (ARPE-19) and a cancerous (HeLa) cell lines. The results showed that the solvent and operational times (5 and 10 min) influenced significantly in the yields using OH extraction, being obtaining the maximum values ( $p < 0.05$ ) with the hydroalcohol solution for both plants. A total of twelve (ferulic acid, gallic acid, p-cumaric acid+epicatechin, catechin, ellagic acid, apigenin, quercetin, resveratrol, kaempferol, taxifolin, naringin, and hesperidin) and seven (ferulic acid, ellagic acid, kaempferol, naringin, o-cumaic acid, rutin, and rosmarinic acid) different phenolic compounds were identified in the Rm and Mg fruit extracts, respectively. Interestingly, some compounds were found in different proportions depending on the solvent and extraction method used; being the extracts of Rm fruit those that presented higher TPC values, and stronger antioxidant and antifungal activities than those of Mg fruit extracts. On the other hand, none of the extract under study showed cytotoxicity against ARPE-19; while three extracts of Rm fruit presented cytotoxicity against HeLa with values of IC<sub>50</sub> in the range of 417.7 to 705.7 µg/mL. These results demonstrated the relationship between the extraction conditions (i.e., extraction method and solvent) and the bioactivity of Rm and Mg fruit extracts. This study reveals the potential of these plants as a new source of bioactive compounds for future applications in various areas, such as agri-food and pharmaceutical industries.

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## STABILITY ENHANCEMENT OF GARLIC-ALLICIN BY ENCAPSULATION IN ORGANIC AND INORGANIC MATRICES

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Alliin is the main metabolite from garlic (*Allium sativum* L.) that confers the biological activity against a series of diseases, such as antimicrobial, anticancer, antihypertensive, and others. However, by light, temperature, and time effects, allicin is quickly transformed to other chemical compounds with less or absent biological activity. Different methods to protect allicin from the environment to avoid its degradation have been studied; among them, encapsulation has considered as environment barrier to promote the stability of allicin. However, most of studied methods involve the use of high temperature (spray dryer), which accelerate the allicin degradation. An alternative is the encapsulation by gelation/precipitation using polymeric natural material or supported on inorganic matrices. In this study, the stability of allicin encapsulated in alginate/nopal-mucilage and titanium and zirconium oxides was analyzed. The nopal-mucilage was obtained from *Opuntia ficus indica* cladodes, titanium and zirconium oxides were synthesized by sol-gel method and allicin extract was obtained from garlic bulbs. The encapsulation in titanium and zirconium oxides was made by impregnation of the materials with an aqueous extract of garlic. The encapsulation in alginate/nopal-mucilage was performed by gelation/precipitation technic. The stability of allicin was monitored by high performance liquid chromatography for three months. The zirconium oxide had the highest encapsulation yield (> 97%), followed alginate/nopal-mucilage with ~90%, while using titanium oxide, a 70% encapsulation yield was observed. The stability study of allicin demonstrated that the inorganic oxides as well the alginate/nopal-mucilage are potential materials to preserve the metabolite from degradation at least three months.

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## SETTING UP PRELIMINARY TESTS TO PROVE THE EFFECT OF COCONUT FATTY ACID ASAN APHID REPELLENT IN PEPPER

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Studies have shown that fatty acids derived from coconut (*Cocos nucifera* L.) oil are repellent compounds against a broad array of blood-sucking arthropods including flies, ticks, bed bugs, and mosquitoes. Taking this as an idea, we decided to test whether coconut fatty acid has repellent activity against sap-sucking arthropods from cultivated plants such as aphids (Hemiptera: Aphididae). Repellency of aphids is relevant in crop protection to prevent the transmission of viruses when the insects feed on plants. The objective of this study was to determine whether coconut fatty acid combined with (*E*)-anethole (nanoemulsion 1:2 or nanoemulsion of the compounds alone) showed a repellent effect on winged aphids (*Myzus persicae*) when sprayed on pepper plants at 0.1-0.2% of the active ingredient. The sample used for the mixture is a waxy starch-coconut fatty acid methyl ester composite, code 19391-122, produced and supplied by NCAUR (ARS-USDA). A preliminary methodology has been developed in culture chambers, by choice assays, with which an average choice percentage of 73% has been achieved. Treated pepper plants were offered to the insects and their distribution was counted after 24 h. The results so far indicate that (*E*)-anethole combined with coconut fatty acid has a higher Repellency Index (R.I.) (59%) than (*E*)-anethole alone (32%) for the winged forms of *M. persicae* on pepper when given a choice with control plants. Furthermore, the model used for the statistical analysis of the data, expressed as a percentage of choice, indicates that there are significant differences between treatments. This indicates that the coconut fatty acid might improve the repellent effect of the bioactive, although more work is needed to confirm this report.

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NEW PERSPECTIVE FOR THE GREEN ROOF SECTOR: *SEDUM* SPP.  
COVERING ACCESSIONS WITH BIOMEDICAL APPLICATIONS

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Climate change and resulting effects are currently becoming a very serious problem, and innovative sustainable techniques are being considered in urban environments as part of a climate change adaptation approach. Green roof areas are promising solutions to face the urban challenges caused by climatic threats. The genus *Sedum* (Crassulaceae) is characterized by a facultative CAM metabolism, permitting a xerophytic adaptation with appropriate eco-physiological characteristics, made these plants perfect for low input green roofs. *Sedum* is also known for nutraceutical and cosmeceutical properties, characterized by high concentrations of polyphenols, polysaccharides and antioxidant capacities in extracts. In the present work, ten *Sedum* genotypes (*S. montanum*, *S. album*, *S. reflexum*, *S. acre*, *S. acre subsp yellow*, *S. sediforme*, *S. spectabile rosa*, *S. herbstfreude*, *S. telephium*) were evaluated for agronomic trials and for bioactive properties of leaf extracts, in order to identify the most promising species for extensive green roof systems along with the best harvesting time for maximum skin healing effects. A completely randomized plot trial was set up on an experimental green-rooftop located at the University of Bologna in October 2018 and monitored until October 2019. Plant growth parameters were measured during the experimental time, included rooting capacity, growth index and ground cover capacity, obtained by the percentage ground cover and Relative Growth Ratio ( $\Delta Area\%/\Delta T$ ). Samples were collected from areal parts of plants harvested at five different phenological phases for the determination of bioactive properties. Samples were extracted and analyzed for total polyphenols and flavonoids content and for the antioxidant capacity (DPPH and FRAP assay). In addition, healing activity of extracts was evaluated with *in vitro* wound healing assay on dermal fibroblasts. All *Sedum* species showed an optimal adaptability to the low input growing conditions tested on the green roof with *S. album* performing the best in terms of coverage percentage. It was observed that the highest expression of antioxidant activity in the total extracts was not correlated with the highest polyphenol contents. This was also evident with the wound healing results, where the biological effect of *Sedum* extracts was clearly not only associated with the expression of polyphenols. In particular, the present study has highlighted *S. album* and *S. acre* as the most promising genotypes with highest coverage index and healing potential. Results of the present study permitted a more profound understanding of the plant growth characteristics and bioactive properties of the investigated *Sedum* genotypes. Hence, the ten genotypes belonging to the genus *Sedum* were qualified from an agronomic and physiological point of view for possible use as ground cover plants on green roofs and for use in the biomedical field.

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THE EFFECT OF THE PREDECESSOR AND THE NITROGEN RATE ON THE PRODUCTIVITY  
AND THE ESSENTIAL OIL CONTENT OF CORIANDER (*CORIANDRUM SATIVUM* L.) IN  
SOUTH – EAST BULGARIA

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Coriander (*Coriandrum sativum* L.) is one of the most important essential oil crops grown in the world. Coriander productivity is determined by genotype, environmental factors, and agronomic practices. In this regard, the aim of the present study is to explore the effect of the predecessor and the nitrogen level on the elements of productivity, seed yield, essential oil content and yield in the variety “Mesten drebnoploden” coriander. A field experiment for evaluating the influence of two precursors (winter wheat and sunflower) and 4 nitrogen (N) norms (0, 40, 80 and 120 kg ha<sup>-1</sup>) on the productivity and essential oil content of the respective variety was conducted. The trial was performed during the period of 2015-2017 in South-East Bulgaria. The experiment was carried out by the block-plot design method in four replications with a plot size of 15 m<sup>2</sup>. The plant height, number of umbels per plant, diameter of umbels, number of umbellets per umbel, number of seeds per umbel, seed weight per plant, 1000 seeds mass, essential oil content, as well as seed and essential oil yield were evaluated. To quantify the relationship between the studied indicators, the experimental data were processed and analysed by the ANOVA method, and the differences between the variants were determined using the Duncan’s Multiple Range Test. The results obtained showed that winter wheat is a more suitable precursor to coriander than sunflower. The highest results for number of umbels per plant, umbels diameter, number of umbellets per umbel, number of the seeds per umbel, seed weight per plant, 1000 seeds mass, as well as seeds yield for the rate of 80 kg N ha<sup>-1</sup> were recorded. The highest essential oil content after the application of 120 kg N ha<sup>-1</sup> was measured. Increasing the N rate from 0 to 120 kg N ha<sup>-1</sup> had a positive and significant effect on essential oil yield, but there were not significant differences between the rates of 80 and 120 kg N ha<sup>-1</sup>.

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*FLOURENSIA MICROPHYLLA*: EFFECTS OF ULTRASOUND ASSISTED EXTRACTION ON PHENOLIC COMPOUNDS, ANTIOXIDANT AND ANTIFUNGAL ACTIVITY OF PHENOLIC EXTRACT

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Currently, there are novel techniques available for use in the extraction process of phenolic compounds from plants, among which the ultrasound-assisted extraction (UAE) stands out, which includes an environmentally friendly process, requires less energy, solvent and extraction time than conventional techniques. *Fusarium oxysporum* is a pathogenic and toxinogenic fungi that represents a serious problem in agriculture in Mexico, which continuously develops resistance to chemical fungicides, for which the research interest has focused on the search for phenolic extracts of plants with fungicidal potential. *Flourensia microphylla*, an endemic plant from the southeast of Coahuila, Mexico, has shown antifungal activity *in vitro* against different pathogens, in the case of *F. oxysporum*, 100% inhibition has not been achieved. Due to the above and considering that the UAE technique can significantly improve the extraction of phenolic compounds with respect to maceration, the following objective was proposed: To evaluate the effects of ultrasound-assisted extraction (UAE) on the content of phenolic compounds, antioxidant and antifungal activity of *F. microphylla*. The extraction of *F. microphylla* compounds was carried out by UAE, at different solvent-to-sample ratios (10, 20 and 40 mL / g), and at different extraction times (10, 20 and 30 min), at 30 °C, for a total of 9 treatments, plus a control treatment, conventional extraction, with four repetitions per treatment. The extraction performance, the total phenolic compounds (TPC), total flavonoids (TF) and the main phenolic compounds were evaluated. Antioxidant activities of the extracts were analyzed using the following methods: DPPH, ABTS, and Reducing Power assays. The antifungal effect of the extract against *F. oxysporum* was also investigated. Considering as a criterion, obtaining an extract with higher ABTS antioxidant activity and a high content of total flavonoids; In addition to reducing the extraction times, the treatment that was selected corresponds to a solvent-to-sample ratio of 40 mL / g and an extraction time of 20 min. The selected extract resulted in an extraction yield of 23.4 g / 100 g D.W; content of total phenolic compounds of 252.8 mg GAE / g D.W; total flavonoids of 435.9 mg QE / g D.W.; DPPH, ABTS and Reducing Power antioxidant activities were: 74.2, 740.4 mM Trolox / g D.W and 33.0 mg ascorbic acid / g D.W, respectively. The phenolic extract obtained by UAE extraction increased the content of phenolic compounds, mainly flavonoids, as well as antioxidants, which potentiates the bioactivity of *F. microphylla*.

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## CULTIVATION OF AROMATIC-MEDICINAL PLANTS, A MEETING POINT BETWEEN RESEARCH AND SOCIAL INCLUSION

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Many products such as essential oils, pharmaceuticals and cosmetics contain or are based on extracts from plants. The need for plant materials used for cosmetic and medicinal purposes, combined with the need to protect plant biodiversity, create an opportunity for farmers and foresters to diversify their production and improve their income. The SPAGYRIA Project (€1.8 mil (\$2.13 mil)- European Union Project) is a cooperation, solidarity and innovation project that aims to create a line of plant extracts for the production of organic cosmetics. The project involves tutelary associations of groups at risk of social exclusion to contribute to improving their employability. The aim is to capitalize on the experience and economic development of partners on both sides of the French-Spanish border, both in terms of the cultivation and packaging of aromatic plants and in terms of sustainable extraction and evaluation of prepared products. This work will present some results of the cultivation of six aromatic-medicinal species in two Spanish localities (Huesca and Pamplona), and one French (Toulouse) for 3 years; the results of yield and chemical characterization of the respective essential oils will also be presented. The cultivated species were *Salvia lavandulifolia*, *Salvia officinalis*, *Salvia sclarea*, *Thymus zygis*, *Thymus vulgaris* and *Melissa officinalis*. The aerial parts of all plants were harvested during the optimal blooming time (70-80% flowering) and distilled at laboratory in a Clevenger-type distiller (1) and in a pilot plant by semi-industrial steam drag (2), when at least 50 kg of dry biomass were available. Essential oil yield varied among different species and locations for the same species, probably because changes in climate conditions for cultivation. Species as *T. zygis* and *S. officinalis* could double yield depending on location. The same variation was observed for chemical characterization. The results suggest aromatic plants can be modulated by different environment conditions, specially their secondary metabolism. Among the most relevant results of SPAGYRIA, it is worth highlighting the activities carried out in matters of transfer of innovation and knowledge that, through courses and workshops, have been carried out by tutelary associations in Spain and France. People involved in these institutions have received courses on productive and processing aspects of medicinal plants, reaching the final product, a cosmetic cream based on plant extracts.

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## WORMWOOD CANDIAL VARIETY ESSENTIAL OIL YIELD UNDER CULTIVATION IN ARAGÓN, SPAIN

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Wormwood (*Artemisia absinthium* L.) has been collected since ancient times, being used by the Egyptians and Greeks. It is a perennial plant, known for its antifungal, parasitic and nematocidal effects. Traditionally, the collection of wormwood has been wild. Due to the increase in demand of essential oils obtained from this plant, it is necessary to cultivate them in a standardized way, with emphasis in the study of the oil quality, in order to ensure the constant and homogeneous production for the industry. An investigation was carried out at Agrifood Research and Technology Centre of Aragón, where the process of the domestication of the species was developed, starting with 2 populations: plants collected at the Sierra Nevada National Park (Gr-75) and the selected population from a pre-trial in Teruel (Te-68). Finally, the Candial variety was registered under Decision N° EU 36714 of 27 January 2014 at the Community Plant Varieties Office. Candial has been extensively (more than 2 ha) tested for possible variations in the essential oil yield and quality. To carry it out, plantations located in Luesia, Bernues and Ayerbe (Aragón, Spain) were compared with the initial trial in Ejea de los Caballeros. The essential oil has been extracted in the laboratory by Clavenger distillation. The yield obtained in Ejea corresponds to the average yield (0.66%) from 2013 to 2018, varying from 0.47% to 1.04%, probably due to meteorological factors. To analyse the cultivation in Luesia, Bernues and Ayerbe, data from 2016 to 2020 have been collected. The maximum yield of essential oil was extracted in Bernues in 2020, exceeding by 78% the average yield of Ejea and the minimum in Luesia in the same year with 27% less. As a conclusion, cultivation decreased essential oil yield from 0.66% to 0.44%, in average. The research continues with chemical characterization of essential oils and their bioactivities.

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ALLELOPATHIC EFFECTS OF JUNIPER ESSENTIAL OILS ON SEED GERMINATION AND SEEDLING  
GROWTH OF SOME WEED SEEDS

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Biopesticides are important for controlling agricultural pests, phytopathogens and weeds as synthetic chemicals are not allowed in certified organic production. Plant essential oils (EOs) are often used as ingredients in the development of new biopesticides. The objective of this study was to investigate the allelopathic effects of EOs of two juniper species against weed seeds. Essential oils (EOs) were extracted of *Juniperus sabina* L. Male (M) and Female (F) and *J. excelsa* Bieb., by steam distillation in semi-commercial extractor. The major classes of the *J. excelsa* EO compounds were monoterpenes and sesquiterpenes, while *J. sabina* was monoterpenes (sabinene). The allelopathic effects of the three EOs on seed germination and on seedling growth of weed seeds were carried out by the De Feo method described previously. Bioherbicidal activities of the three EOs were assayed in four concentrations (0  $\mu$ L, 30  $\mu$ L, 60  $\mu$ L and 90  $\mu$ L) against two weed species; *Melilotus officinalis* L. and *Myosotis arvensis* (L.) Hill. The three EOs used in the experiments exhibited different inhibitory effects on tested seeds. The tested juniper EOs actually stimulated the germination of *Melilotus officinalis* seeds. With the increasing of the EO application rates (concentrations), the germination energy (measured at the 4th day) increased up to 70 - 100%, compared with 15% in the non-treated control. However, after the 7th day, an inhibitory effect occurred, and the seed germination of the treated seeds was reduced to 37 - 12% compared with the 63% in the non-treated control. In *Myosotis arvensis* seeds, the *J. sabina* EO had an inhibitory effect on germination energy and germination, while the *J. excelsa* EO stimulated germination. The sprout and root length varied depending on juniper species and EO concentrations in both weed species. The *J. sabina* (M, F) EOs showed the highest inhibitory effect on seed germination as well as on sprout and root lengths. The tested juniper EOs showed promise as ingredients to be included in new products for weed control in organic systems.

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# OPPORTUNITIES FOR SUSTAINABLE PRODUCTION OF ROSA DAMASCENA THROUGH APPLICATION OF THE PRINCIPLES OF BIOECONOMY – A CASE STUDY FROM PLOVDIV REGION

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The production of *Rosa Damascena* and rose products is traditional for Bulgaria and is of key importance for the family farms in the rural areas where it is carried out. At the same time, this production faces a number of technological, economic and environmental challenges. The latter determines the need to look for opportunities to apply the principles of the bioeconomy through an appropriate use of residues and thus potentially increase the revenues of the participants in the process. In this regard, the aim of the study was based on research and analysis of opportunities for optimal use of waste products in the production and processing of *Rosa Damascena*, to reveal the potential for sustainable results in the following three dimensions - economic, social and environmental. This article discusses: (1) trends and problems in the production of raw material; (2) potential for increasing the economic efficiency of processing enterprises and (3) opportunities for stabilizing the economic results for rose growers. The methodological framework was based on case study method. In the period May - June 2021, in the village of Zelenikovo, Plovdiv region, where one of the largest distilleries is located, in-depth interviews were conducted with key experts and representatives of the stakeholders. The results of the study reveal that in recent years there have been market shocks that lead to a decrease in profits and incomes of participants in the process and a corresponding decrease in the number of producers and processors of *Rosa Damascena*. Potential opportunities for using of waste products from processing, which are currently discarded, have been identified and evaluated.

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## ANTIMICROBIAL ACTIVITY AND ALLELOPATHIC EFFECTS OF ESSENTIAL OILS ON SEED GERMINATION OF BARLEY

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Plant essential oils (EOs) are known for their allelopathic and antimicrobial activities. This study elucidated the antimicrobial and allelopathic effects of the EOs from 10 different plant species; common juniper (*Juniperus communis* L.), creeping juniper (*Juniperus horizontalis* Moench.), Rocky Mountain juniper (*Juniperus scopulorum* Sarg.), lavender (*Lavandula angustifolia* Mill), hyssop (*Hyssopus officinalis* L), English thyme (*Thymus vulgaris* L.), lovage (*Levisticum officinale* W.D.J. Koch) costmary (*Chrysanthemum balsamita* L.), absinthe (*Artemisia absinthium* L.), and cumin (*Cuminum cyminum* L.) on barley (*Hordeum vulgare* L.) seed germination and seedling growth. This was *in-vitro* Petri Dish experiment. The antimicrobial activity of the EOs was expressed as percent inhibition following a previously published method. The 10 EOs had different composition. The major EO constituent of common juniper was alpha-pinene, whereas major oil constituent of creeping and Rocky Mountain junipers was sabinene. The major constituents of EO in other species were linalool and linalyl acetate in lavender, cis-pinocamphone and beta-pinene in hyssop, tymol and para-cymen in English thyme,  $\alpha$ -terpinyl acetate and beta-phellandrene in lovage, trans-thujone and camphor in costmary, thujone in absinthe, cumin aldehyde in cumin seed oil. All 10 oils exhibited allelopathic effect and suppressed barley seed germination and initial seedling development, however, the concentrations that exhibited suppressing effect were different among the species. The 10 EOs were also tested for antimicrobial activity against 10 different microorganisms: for antifungal activity against *Candida albicans*, *C. glabrata*, *C. krusei*, *Aspergillus fumigatus*, *Cryptococcus neoformans* and antibacterial potential against gram +ve bacteria *Staphylococcus aureus*, *aureus* and *Mycobacterium intracellulare* and gram -ve bacteria *Escherichia coli* and *Pseudomonas aerogenosa* at a concentration of 50  $\mu$ g/ml. However, none showed significant activity at the tested concentrations. This study also demonstrated the allelopathic effects of 10 EOs on seed germination of barley. Further research is needed to verify the results under field conditions.

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## IMPACT OF HARVESTING TIME ON PHYTOCHEMICAL CONSTITUENT AND ANTIOXIDANT PROPERTIES OF SWEET BASIL VARIETIES

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Sweet basil (*Ocimum basilicum* Linn.) is a popular herb, valued for its rich and spicy one such herb and has been used as traditional medicine for household remedy against various human ailments from antiquity. In this study, four sweet basil varieties (Lemon basil, Holy basil, Cinnamon basil and Osmin purple basil) were planted into polyethylene bag measured at 45×38 cm which were to be filled with soilless mixture included burnt rice husk and coco peat with ratio 1:1 under glasshouse condition at the glasshouse complex of Universiti Putra Malaysia (UPM). Sweet basil varieties harvested from 1 month to four months after plantation. Compositions and contents of antioxidant components of four sweet basil varieties were identified using ultra-high performance liquid chromatography (HPLC) and the antioxidant activities were evaluated using scavenging 1,1-diphenyl-2-picrylhydrazyl (DPPH) radicals, Trolox equivalent antioxidant capacity (TEAC), ferric reducing power (FRAP) and inhibiting Cu<sup>2+</sup>-induced human LDL oxidation. Sweet basil varieties harvested from 1 month to four months after plantation were determined. The results showed that harvest time and variety were important factors affecting the compositions and contents of phenolic compounds in sweet basil leaves; moreover, phenolic contents (polyphenol, flavonoid, condensed tannin and phenolic acid) of the leaves were significantly correlated with their antioxidant activities. In this study, 14 phenolic acids and 11 flavonoids were separated and identified in sweet basil varieties using high performance liquid chromatography. For each variety, the leaves harvested in months with higher temperature, solar radiation and sunshine duration had higher phenolic contents contributing to better antioxidant properties (ranking: 3months > 4months > 2 months > 1 months). In addition, the compositions and contents of phenolic components and antioxidant capacities for the leaves from various sweet basil varieties were also different. The leaves of Osmin purple basil exhibit the highest concentrations of phytochemicals, as well as the highest biological activity following Cinnamon basil, lemon basil and Holy basil. It may also be recommended for the food industry to use this variety for their products, which are going to compete in the expanding functional food markets.

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*FLOURENSIA RETINOPHYLLA*: AN OUTSTANDING PLANT, FROM NORTHERN MÉXICO  
WITH ANTIBACTERIAL ACTIVITY

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Urinary tract infections (UTIs) represent a serious health problem, since they are the third most common type of infectious disease in the world and the second in the United States and Europe. To control infections, antibiotics are prescribed against Gram positive and Gram negative bacteria, which is the main cause of antibiotic resistance. This has caused great interest in the search for bioactive compounds of natural origin, based on plants as an alternative and supplementary medicine, against the pathogens that cause UTIs. *Flourensia retinophylla* is a plant from the arid zones of Mexico that has shown important biological activities. The objective of the research was to evaluate the *in vitro* antibacterial activity of the ethanol extract of *F. retinophylla* leaves against bacteria that cause UTIs: *Enterobacter aerogenes*, *Escherichia coli*, *Proteus hauseri*, *P. mirabilis*, *P. vulgaris*, and *Staphylococcus epidermidis*. The contents of total phenols and flavonoids were determined, as well as the antioxidant activity by DPPH and the composition of phenols by UPLC. The *F. retinophylla* extract showed remarkable antibacterial activity, generally inhibiting all bacteria 100%. *Enterobacter aerogenes* was 100% inhibited with a concentration of 50 mg / L, with a MIC<sub>90</sub> of 50 mg / L, in this bacterium the control, Penicillin G, presented a MIC<sub>90</sub> of 669 mg / L. In addition, *P. hauseri* and *P. mirabilis* presented a MIC<sub>90</sub> of 69.5 and 87.7 mg/L, respectively. The extract from the leaves of *F. retinophylla* exhibited an important source of antioxidant compounds, including flavonoids and polyphenols. *F. retinophylla* extract could be formulated and developed to be used as a supplement and alternative medicine against bacteria that cause urinary tract infection, and that have increased its resistance to antibiotics.

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## CANNABIS MEDICA NAZIONALE - CAMED: INNOVATION AND ENHANCEMENT OF THE PRODUCTION OF MEDICAL CANNABIS PLANT MATERIAL FOR NATIONAL DEMANDS AND NEW VARIETAL CONSTITUTION FOR PHARMACEUTICAL USE

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The national project CAMED is of great scientific relevance and mediatic impact since it affects the medical *Cannabis* supply chain in Italy. Indeed, *Cannabis sativa* L. is a natural biofactory of secondary metabolites, among which phytocannabinoids, that have proved medical and pharmaceutical properties, are exploited for the preparation of *Cannabis*-based drugs. CAMED is strategically relevant for the country since it will support from a technical-scientific point of view the Agreement between the Ministry of Agricultural, Food and Forestry Policies and the Ministries of Defense and Health, with the aim to guarantee the increased national production of *Cannabis*-derived medicinal products. The CREA – Research Center for Cereal and Industrial Crops (CREA-CI) in Rovigo boasts a tradition of national scientific excellence on medical *Cannabis* breeding programs and is the only one authorized to cultivate high-THC *Cannabis* even for research purposes, as stated by the art. n. 26 of the D.P.R. 9/10/1990 n. 309. In Italy, the unique *Cannabis*-based drugs, namely FM1 and FM2, produced and distributed by Military Chemical-Pharmaceutical Factory (SCFM) in Florence, are based on inflorescences of two Italian medical *Cannabis* varieties CINBOL and CINRO, developed via classical breeding at CREA-CI, patented and registered at CPVO (Community Plant Variety Office). CINBOL is characterized by a prevalence of THCA, while CINRO has a CBDA: THCA ratio of 1.5:1. Mother plants of CINBOL and CINRO are maintained in vegetative state in indoor facilities; cuttings were produced from mother plants and used as starting material for cultivation and production of the pharmaceutical preparations based on inflorescences. In this scenario, the CAMED project addresses two major needs: increase and innovate the production of medical *Cannabis*. Indeed, it will guarantee the multiplication in genetic purity of CINBOL and CINRO and the production of cutting as well as breeding activities to select new genetics improved for the content in major cannabinoids. Furthermore, CAMED will provide advancements in the comprehension of the genetic mechanisms responsible for the synthesis and accumulation of cannabinoids. The CAMED project has a duration of two years starting in 2021 and is organized in 4 Work Packages (WPs). The WPs deal with: (WP1) the infrastructural upgrading of the CREA-CI in Rovigo which is compulsory for the maintenance and breeding of *Cannabis* mother plants and for implementation and optimization of the production of the cuttings; (WP2) the maintenance in purity of the mother plants of the CINBOL and CINRO varieties, and cuttings production and supply to SCFM; (WP3) breeding program for varietal constitution of new lines enriched in cannabinoids for pharmaceutical purposes; (WP4) the analysis of the sector regulations, and the study of new programmatic policy tools for the sector plans of the *Cannabis* supply chain. In the context of breeding activity, the WP3 will characterize also pre-breeding material with the aims to (i) develop new genetic lines for pharmaceutical purposes with specific combinations of cannabinoids in terms of THCA and CBDA; (ii) fill the knowledge-gap on the genetic of cannabinoids synthesis and its regulation; (iii) characterize the cannabinoids-terpenes phytocomplex of the *Cannabis* inflorescence and evaluate the so called “*Entourage effect*”.

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# **ABSTRACTS NATURAL RUBBER AND RESINS DIVISION**

**ORAL + KEYNOTE PRESENTATIONS**

**CHAIR**

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## SUSTAINABILITY ASSESSMENT OF PRODUCING GUAYULE RUBBER WITH COPRODUCTS

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Guayule (*Parthenium argentatum*) is shrub native to arid regions of the southwest United States and northern Mexico. Guayule has long been considered a source of natural rubber due to its relatively high rubber content compared to other plants. However, this rubber content still typically makes up less than 10% of the total plant biomass. As a result, economic viability is largely dependent upon the other 90% of the guayule biomass. This work presents an economic and environmental sustainability analysis of guayule rubber production, including the resin and bagasse coproducts that make up the non-rubber portion of the plant. At the core of this analysis is a process model that tracks all energy and materials required to cultivate, transport, and extract guayule rubber, resin, and bagasse. Using this model, we performed a techno-economic analysis and life cycle assessment to assess economic and environmental performance across a range of scenarios. When considering a baseline scenario, we found a minimum rubber selling price of \$3.08 per kg of guayule rubber, with coproducts of resin and bagasse sold at \$1.00 per kg and \$0.10 per kg, respectively. This baseline scenario resulted in cumulative emissions of 10.3 kg of CO<sub>2</sub> eq per kg of rubber produced. Additional scenarios were evaluated, including increased rubber content, harvest yield, and coproduct selling prices. The uncertainty of coproduct selling prices led to further work including a preliminary market investigation of potential resin products and evaluation of multiple bagasse to energy pathways. Based upon these analyses, several important conclusions can be drawn. First, there is large uncertainty surround resin and bagasse coproducts. This uncertainty greatly influences economic and environmental sustainability results. Second, there is a wide range of potential products that might be suitable for guayule resin. However, the separation and effectiveness of guayule resin in these different applications must be tested to reduce uncertainty. Last, guayule bagasse could be an ideal feedstock for energy production, but the operational costs of producing liquid biofuels are high. Directly combusting or pelletizing bagasse appear to be the more effective energy options in the near term.

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## A ROUTE TO SCALABILITY: GUAYULE LATEX RADIATION ATTENUATION GLOVES

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Disruptions of the global supply chains are making domestic natural rubber production vital to national security and defense. A major scalability challenge remains in that commodity markets, which require low prices and huge supplies cannot be entered by rubber or latex produced at pilot scale from small acreages of alternative rubber crops. Thus, premium markets with high profit margins must be identified and addressed to support commercialization on the small scale at which these crops will be initially introduced. Radiation attenuation gloves (RAG) are one such market. RAG protect and shield health care workers (HCW) from occupational exposure to ionizing radiation. Natural latex RAG are classified as personal protective equipment because the high load of attenuation filler required reduces their mechanical properties to below Food and Drug Administration (FDA) medical glove physical performance requirements and so may not provide a sufficient barrier against potentially infectious materials; therefore, double gloving RAG with medical gloves - examination or surgical - is required to fully protect health care workers. However, RAG are often not regularly worn by health specialists since they are thicker and heavier than regular medical gloves, and so decrease tactile sensation, hand dexterity, and fine motor control in the fingers especially when double-gloving is used. Guayule natural rubber latex (GNRL) is an alternative circumallergenic natural elastomer. GNRL's low protein, relatively high fatty acid and resin content and linear polymers make it softer and more elastic than Hevea rubber, and at the same time, enable a high filler loading while maintaining outstanding physical properties. The objective of this study was to investigate the radiation attenuation and mechanical properties of GNRL films prepared with a radio-opaque material filler, micro-sized Bi<sub>2</sub>O<sub>3</sub>. GNRL/Bi<sub>2</sub>O<sub>3</sub> films were produced with Bi<sub>2</sub>O<sub>3</sub> loads ranging from 50 to 300 parts per hundred of rubber (PHR) and film thicknesses from 0.2 to 0.3 mm. The films were subjected to 60, 80, 100, and 120 KeV ionization energies and tensile measurements (ASTM D412). Attenuation efficiencies of GNRL/Bi<sub>2</sub>O<sub>3</sub> films with 150 PHR Bi<sub>2</sub>O<sub>3</sub> loading at 0.2 mm thickness produced by optimized curing conditions were above the ASTM attenuation requirements and met the tensile requirements for natural latex examination and surgical gloves. RAG produced by GNRL would eliminate the need for double-gloving during radiation-assisted procedures and ensure the safety and security of HCW. Entry of GNRL into specialty markets of this type can pave the way to guayule expansion.

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## ADAPTING INDUSTRIAL CROPS TO URBAN BROWNFIELDS: THE FRENCH GUAYULE CASE

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Urban Brownfields (UB) are a black mark of our landscapes with numerous issues: degraded soils (compaction, lack of fertility, pollutant), limited biodiversity and associated ecosystem services furniture, unwanted species (including humans) and vague future. Producing high value biomasses on these brownfields is a promising leverage to “reuse” these areas economically and ecologically. Cultivating biomasses on urban brownfield will improve soil quality, increase ecosystem services furniture and prevent unwanted species installation. For this purpose, Guayule (*Parthenium argentatum* A. Gray) is a promising crop as it is able to grow on low fertility soils and is expected to be economically profitable on small areas (>1 ha). But numerous technical-economical barriers have to be broke to allow Guayule cultivation on brownfields. This is the aims of the AgroGuayule project (ADEME-GRAINE cofounding). Pilot plantations were made across various French brownfields to address the feasibility of guayule production in various conditions of soils, climate and practices. These urban brownfield scenario will be compared to a more conventional scenario (agricultural context) in terms of technical choices and biomass production.

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## COLD RESISTANCE OF GUAYULE CULTIVATED IN CASTILLA-LA MANCHA, SPAIN

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The guayule plant (*Parthenium argentatum* A. Gray) is undoubtedly the most promising natural rubber producing plant for semiarid locations in the Center and Southeast of Spain, capable of replacing the traditional source. Since 2017 a mixed research team from ITAP (Provincial Agricultural Technical Institute) and UCLM (University of Castilla-La Mancha) have been studying the adaptation of several accessions in this geographical area with good results in a wide variety of agronomic aspects except for cold hardiness. On January 7<sup>th</sup>, 2021, this crop was affected by the "Filomena" squall, according to the Spanish Meteorological Agency (AEMET) the biggest snowstorm in Spain in 50 years that kept the crop covered by a layer of snow thick around 50 cm for two weeks. This unexpected event has been used by researchers to study the survival of 11 different guayule and hybrid accessions planted in 2019 in the field called Marina-03 of almost 1 ha size (Santa Cruz de la Zarza, Toledo). This field has its own weather station which shows that the snowfall began on 01/07/2021, intensifying the next day, and reaching a maximum magnitude on 01/09/2021. Subsequently, the snow remained until 01/20/2021. This station recorded temperature peaks of -18.0°C during these two weeks, accumulating, between 12<sup>th</sup> and 13<sup>th</sup> January, 27 hours of temperatures close to -10°C. The study of the damage caused to the guayule crop was done at two specific times: when the first plants began to resprout on April 27<sup>th</sup>, and a month and a half later (June 10<sup>th</sup>) when the survivors were counted. On the first sampling date, 10 representative plants of each variety were randomly selected and analyzed for presence of green leaves from the previous vegetative cycle, sprouting, level of sprouting from the ground (downside, medium side, upper side), and internal state of the plants by making cuts at different heights of the stem (10, 20 and 30 cm). The results shown a significant reduction in the vigor of the plants, which increases in the AZ-2 or 11693 accessions, with 50% of the plants showing no regrowth. The remaining 50% of the plants only regrow in the lower parts of the plant. Internally, the hybrid varieties (*guayule x Parthenium*) as AZ-3, CAL-1, and CAL-2 showed serious damages in the aerial part, some of them showed fungal rot, with doubts in a future regrowth. However, the guayule varieties such as AZ-5, A-48188 or N-565, shown a greater adaptation, less internal damages were observed and the 90% of their plants showed regrowth at all levels. The presence of fungal attacks led to the decision to cut the field 15 cm above the ground to facilitate regrowth and prevent the spread of fungal diseases. On June 10<sup>th</sup> evaluation, the 81.3 % of the 19023 plants analyzed were alive with a very different distribution among accessions. The result maintains the trend observed previously, N-565 accession was the one with the highest survival rate (92.9%) while AZ-2 and 11693 accessions had the lowest survival rate (51.9 and 50.1% respectively), although it was remarkable a significant improvement in the survival rate of hybrid varieties CAL-1 and CAL-2 with respect to the first analysis.

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## COMPOSITION OF GUAYULE RESIN

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Guayule, *Parthenium Argentatum* Gray, is a semi-arid shrub, native from the Chihuahan desert in Mexico. This plant produces polyisoprene, a polymer that can be extracted under the emulsion form (latex) or under the dry form (rubber). It grows in Mediterranean climates, like in the South of France and it represent an opportunity to make dipped product (like gloves, condoms or medical devices) and vulcanized products (like tires) in Europe and stopped being dependent of the Hevea latex coming from Asia. Today, an aqueous process is used to extract guayule polyisoprene under the latex form and in order to make a sustainable and economical-viable process, co-product have to be valorized. One of the co-products is resin, a mix of compounds here defined by the acetone extractables using ASE (temperature: 40°C, pressure: 10<sup>7</sup> Pa). Some of the constituent have already been described, but there is a need for a complete qualitative and quantitative analysis of the resin. Our team developed methods based on liquid and gas chromatography, coupled with mass and UV spectrometry to analyze the various constituents of the resin: argentatins, guayulins, lipids, waxes, sugars, polyphenols, and monoterpenes.

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## GUAYULE, A PROMISING BIOFUEL AND BIOPRODUCTS CROP

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Sustainable, alternative fuels represent a promising solution to help reduce carbon emissions, expand domestic energy sources, contribute to price, and supply stability, and stimulate economic development in rural communities. The success of biofuels relies in finding inexpensive feedstocks that do not compete with food crops and can be cultivated economically in diverse geographical regions and agricultural production systems. In addition to the commonly known use of guayule (*Parthenium argentatum* A. Gray) as an alternative source for natural rubber and hypoallergenic latex, the native shrub to Southwest Texas desert is a rich source of resins and bagasse that make it an ideal feedstock to use in byproducts, pharmaceutical and biofuel industries. Increasing guayule production could be achieved by incorporating it into the sustainable agricultural system of Southern U.S. marginal land regions, where alternative rotation crops are limited. To produce fuel and bio-chemicals, guayule feedstock is deconstructed and fractionated or converted into intermediates such as sugars, condensable gas, non-condensable gas, or chemical block components that can be further refined. The four major technologies to process feedstock during deconstruction and fractionation steps include hydrolysis, gasification, hydrothermal liquefaction and pyrolysis. Our research indicated that guayule pyrolysis byproducts production includes non-condensable gases, condensable gases (bio-oil) and bio-char. The productions of these byproducts are affected by guayule genotypes and growing conditions. The observed significant phenotypic variations could lay the foundation for genetic improvement of guayule for byproduct production. The significant association between irrigation schedules and guayule pyrolysis byproducts could reduce irrigation requirements without impairing guayule byproducts production. The positive correlations among pyrolysis byproducts suggest the possibility to genetically improve several byproducts simultaneously in guayule breeding programs which could increase the genetic gain of guayule byproducts. This research gives an insight to breed for guayule as an economic crop with high bioenergy potential for sustainable agriculture systems.

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## GUAYULE: ALTERNATIVE CROP FOR SEMI-ARID REGIONS IN SPAIN

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Guayule (*Parthenium argentatum* A. Gray) could be an interesting agricultural alternative for semi-arid areas of central and eastern Spain, where the scarcity of water and the poor fertility of some of its soils lead to low productivity in the cultivation of cereals. Successful trials with this plant were carried out from 1931 to 1960s in southern Spain. At that moment, plantations of more than 4250 ha were established through the region of Andalusia promoted by the Intercontinental Rubber Company. Later, it was also introduced in the region of Murcia (south eastern Spain) within a European project for the search of alternative sources of rubber (EU-Pearls Project). However, those attempts to introduce guayule crop failed due to the lack of rubber extracting industries. Nowadays, the climate change has opened the way for this plant to be grown in more northern and inland locations, which previously did not seem appropriate. Among these new locations is central Spain area, where new tests must be carried out, as the behaviour of each line varies greatly with the environment. Furthermore, it is also necessary to update the studies because after the 1960s trials more productive lines have been developed. So, the objective of this study is to select the lines that best adapt to the agro-climatic conditions of Castilla-La Mancha with larger biomass production. For this purpose, 27 guayule accessions were planted in May 2017 in a 0.5 ha experimental field with a randomized designed with three replications per accession and 33,333 plants/ha. Almost all accessions (26) came from the USDA-ARS National Plant Germplasm System and were germinated and transplanted at three months of age, except accession 11591 (CL-1), which was received from CIRAD (France) as four months of age plants. The vegetative development was studied along five sampling dates during the first 24 months after the establishment of the crop. In each of them four plants were taken from each individual plot and the following parameters were measured: height, width, branches diameter, number of branches, dry biomass per plant, branches dry biomass per plant, total plant biomass per hectare, total branches biomass per hectare, volume and ratio height/width. As resume, the tallest guayule plants belonged to accessions CAL-1 and AZ-2, reaching 76 cm height. Branches were more numerous and narrower with age, with 27 branches as average in May 2019 and diameter between 0.12-1.83 cm for accessions R1092 and 11619 respectively. Concerning biomass, the average production per plant was 308.71 g in May 2019, when branches biomass supposed 78% of the total one. Regarding the yield, 5.48 t/ha of biomass were produced as average. The highest production was obtained from AZ-2, 593, AZ-3, CAL-1 and CAL-2. Full details will be provided during the presentation.

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# IMPLEMENTATION GAPS FOR THE BUILDING OF A SUSTAINABLE BIOECONOMY VALUE CHAIN. LESSONS FROM GUAYULE CASE IN OCCITANIA

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To be sustainable, bioeconomy must be reproducible by future generations without exhausting its own foundations. However, the bioeconomy necessarily impacts ecosystems and their biodiversity. Can a bioeconomy which makes the profitability compatible with a minimization of the disruption of cycles (water, elements, etc.) and the strengthening of ecosystem services, constitute an agroecological lever? That is the question we are asking here, by focusing on the experience of the development of a new Guayule (*Parthenium argentatum* A. Gray) sector in Occitania to draw lessons and analyze, in the light of the concrete implementation (various collaborative research projects), the questions to be solved, the possible solutions, and the challenges to be met. It offers the opportunity to challenge the implementation of the principles of agroecology and bioeconomy in real conditions, taking into account the realities of the territory. This will involve evaluating the technical and economic issues related to the application of these principles upstream of the guayule sector in the Mediterranean Region, and also taking into account agricultural habits in this Region and its specificities. By focusing on the upstream part of the sector (biomass production), we will present the lessons learnt from the technical feasibility of cultivating Guayule according to certain agroecological principles, such as the diversity of cultivated varieties, plant cover, the association of crops, the use of nature-based solutions or the limitation of tillage. It clearly appears that the optimization of the valuation of ecosystem services constitutes a source for the creation of economic value beyond environmental value. Crop associations such as varietal diversity are enhancers of ecosystem services that simultaneously reduce risks (biotic and abiotic shocks), promote biodiversity, manage soil fertility as well as weed control. Geneticists and breeders, for their part, work on varietal diversity and on the reproductive specificities of guayule to secure the quality of the biomass in terms of economic value as well as the stable production of quality seeds, which is a keystone for industrial crops. On the other hand, this critical analysis must take into account the issues linked to the fragmentation of the plots in the Region, versus the need to have sufficiently large surfaces to operate the scaling up of this production to allow the development of a profitable agro-industrial sector. Finally, our presentation will examine the bioeconomic feasibility of this sector by taking into consideration the strategic nature of this crop on the European territory, and highlighting the need to consider the payment for the ecosystem services that may be associated with it. Indeed, the valorization of agricultural and industrial wasteland by Guayule crops rehabilitates the soils functions and contributes to carbon sequestration. All of this stems from a collective learning process that the transdisciplinary team working on the Guayule in Occitania strives to maintain.

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## IMPROVING GUAYULINS SELECTIVE EXTRACTION

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Guayule (*Parthenium argentatum* A. Gray) rubber is not economically profitable for the moment in comparison with *Hevea brasiliensis*' rubber. That is why in recent years the exploitation of guayule co-products is being explored to improve its profitability. One of these co-products are the guayulins, sesquiterpene components of the resin that have been shown to have insecticidal and fungicidal properties, among others. The isolation of these compounds from the resin, generally obtained with acetone, have been carried out with two different strategies in the literature, column chromatography or solid matrix adsorption, but nothing is said about the conditions that maximize their extraction from the vegetable material. In order to optimize the guayulin extraction procedure, 3 years old guayule AZ-5 accession was collected, grounded to 0.5 mm of particle size, and dried to a moisture content of 8.6%. Solvent selection was carried out employing accelerated solvent extraction with three different solvents (ethanol, acetone, and acetonitrile), at three different temperatures (40, 56 and 80°C) and two cycle times (5 or 20 minutes), to narrow down the conditions. Although ethanol extractions showed a higher resin yield by weight than acetone and acetonitrile extractions, the higher guayulin content was achieved with acetonitrile's, followed by acetone suggesting then that acetonitrile extraction is more selective for guayulins extraction, and thus more desirable to prepare the resin for subsequent guayulins' purification. Also, ASE revealed that the variable with the greatest influence on resin and guayulins yield is the solvent, while cycle time and temperature are dependent on this. To simulate the process on an industrial scale without pressure conditions, the solvent extraction (SE) was performed only with acetone and acetonitrile at 5 different cycle times (30, 45, 60, 120 and 180 minutes). Hardly any differences were observed in resin yield, with the acetone extraction at 60 min being slightly better. However, the concentration of guayulins did experience changes according to conditions. While the concentration of apolar guayulins (A and B) remained quite constant, polar guayulins (C and D) performed better in acetonitrile. With both solvents there is higher yield at short times. Finally, the plant material:solvent ratio and number of cycles optimization was carried out to get the best conditions to reach a compromise between high guayulins yield and time and cost effectiveness.

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# OPTIMAL DESIGN OF GUAYULE AND GUAR SUPPLY CHAINS FOR THE AMERICAN SOUTHWEST

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The changing climate and the effects of a growing population are causing an increasing agricultural drought in the American Southwest. As a risk mitigation measure, water scarcity has increased the importance of the introduction to drought-tolerant and low-water-use crops that can yield high-value products, such as guar [*Cyamopsis tetragonoloba* (L.) Taub.] and guayule [*Parthenium argentatum* A.Gray.] Guar is a native Indian legume from which guar gum is obtained its derivatives are essential in numerous applications such as agriculture, batteries, ceramics, cosmetics, food, paints, petroleum, pharmaceuticals, and textiles. Guayule is native to the Chihuahuan Desert, from northern Mexico to the southwestern U.S. regions, and it has been treated as a source of natural rubber. However, the guar and guayule supply chains still require a scale-up to profitable production. In this paper, the optimal design of guar and guayule supply chains for American Southwest, that is, New Mexico, Northwestern Texas, and Arizona, is performed. All farms within this region are evaluated and specific cotton, grains, and oilseeds farm groups are identified as candidates for guar and guayule adoption. The information from the farms, transportation such as roads major highways, and railroads, and water sources, is captured using a Geographic Information System. A stochastic multi-objective optimization model assesses the economic, environmental, and social impacts of the guar and guayule supply chains and addresses the adoption rate uncertainty through stochastic scenarios. To identify the effects of a long-term increase in the guar and guayule demand, sensitivity analyses are performed for multiple optimal facility locations. An additional case study is performed for Arizona while avoiding specific regions of cotton, grains, and oilseeds farm groups that have been identified with a reduced probability for guayule adoption. The resulting model is formulated as a complex large-scale mixed-integer linear optimization problem. and the Benders Decomposition algorithm is implemented for an efficient solution. Results indicate the expected guar and guayule yearly production from each cotton, grain, and oilseed farm group and the corresponding facility to be supplied to. For the case of a single processing facility in the state of New Mexico, the optimal location is identified in Quay County. Cotton, grains, and oilseeds through the entire state of New Mexico are expected to provide guar between the range (0.0, 235.8] metric tonnes per year. For a single processing facility in the Northwestern region of the state of Texas, the optimal location is identified in Howard County. The cotton, grains, and oilseed farm groups with the highest yearly expected productivity are mostly within Howard County, supplying between 2,439 to 2,842 metric tonnes per year. For a single processing facility in the state of Arizona, the optimal location is identified in Pinal County. All the yearly expected production of guayule is being supplied by cotton, grains, and oilseed farm groups from within the same county ranging up to 22,200 metric tonnes per year for some farm groups. The sensitivity analysis assessed the effect of a long-term demand increment and identified multiple optimal facility locations for such purpose.

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## IRRIGATION EFFECTS ON SEASONAL GROWTH AND RUBBER PRODUCTION OF DIRECT-SEEDED GUAYULE

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Understanding guayule's response to environmental factors, such as location, soil type, drought stress, and seasonal growth variation is critical for irrigation management to maximize and estimate rubber and resin accumulation throughout the growing seasons. A study was conducted at two sites with different soil types (sandy loam soil at Maricopa, AZ and a clay soil at Eloy, AZ) to compare plant growth and rubber accumulation among different irrigation treatments during the two-year growing season. The above- and below-ground biomass, biomass growth, rubber/resin content, and rubber/resin accumulation were measured every other month from establishment to final harvest in well-watered treatments, which received 100% replacement of crop evapotranspiration (ET<sub>c</sub>) and irrigated with subsurface drip and furrow (denoted as D100 and F100, respectively), and compared to drought stress treatments (D50 and F50), which received 50% replacement of ET<sub>c</sub>. Drip irrigation with high water input (D100) decreased root mass partition, but leaf, stem, and flower partitions were not significantly affected by irrigation treatment. Biomass yield was higher in the well-watered treatments as expected, while rubber and resin content were lower, indicating rubber and resin dilution by higher biomass. For all treatments, rubber and resin yield increased linearly over the two-year growing season. However, the rates of increase were different among the irrigation treatments. The D100 treatment had a higher rubber yield increase rate compared to F100 and D50 in sandy loam soil at Maricopa, while the D100 treatment had the lowest increase rate compared to the F100, F50, and D50 treatments in clay soil at Eloy. Top branches of guayule plants in the D100 treatment at Eloy lodged in the second year and likely contributed to lower rubber content and rubber yield in the treatment. Root rubber content was 31% to 39% lower than stem. This study indicates that rubber biosynthesis occurred in guayule year-round and that it is possible in clay soils to reduce irrigation without a significant loss in rubber yield.

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# **ABSTRACTS NATURAL RUBBER AND RESINS DIVISION**

**POSTERS**

**CHAIR**

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## IDENTIFICATION OF NEW POLYPHENOLS IN LEAF OF GUAYULE AND ITS HYBRIDS

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The guayule (*Parthenium argentatum* A.Gray) leaves represent a significant amount of the generated biomass to which an application should be sought. Early studies focused their attention on the production of essential and volatile oils while recently more attention has been paid to their polyphenol content as an option for their valorization. A very common approach for plant residues at present because of their beneficial effects on health. To date, the following have been identified: 11 flavonoids and 26 polyphenols more belonging to other families. Within the present work 48 polyphenols were tentatively identified by LC-MS (Q-exactive hybrid quadrupole-orbitrap mass spectrometer) analysis of leaves from 25 accessions of guayule and hybrids, being 26 compounds previously identified by other authors. It was also possible to assign the structure of the three isomers of chlorogenic acid (caffeoylquinic acid) previously found, but whose exact structures were pending assignment. The first of the compounds, the one found in higher concentration, corresponds to 3-caffeoylquinic acid (3-CQA) identified by its m/z signals 135 and 179 in addition to the m/z signal 191 which is the characteristic of the family. The second compound to appear corresponds to 5-caffeoylquinic (5-CQA) acid which only shows the family characteristic signal, m/z 191. The 5-CQA is, of all family members, the one that arouses the most interest for its relevant bioactivity as antioxidant, anti-inflammatory, analgesic, antipyretic, and anticarcinogenic. This compound was found in some guayule varieties as CAL-7, but not in others perhaps because it can be easily transformed by acyl migration into 4-CQA and 3-CQA, even under mild conditions in the laboratory. The third of the isomers corresponds to 4-caffeoylquinic acid (4-CQA) showing values for the m/z 173 and 179 bigger than for the m/z 191 signal. On the other hand, the results obtained for 5 other compounds are not in accordance with the previous identification: galloylquinic acid (m/z 343), ethyl m-digallate (m/z 349), tetrahydroxy trimethoxyflavone (m/z 375), tetrahydroxy tetramethoxy flavone (m/z 405) and isorhamnetin glucuronide (m/z 491), which could belong to the same family of compounds. The new molecular formula proposed for each of them will be presented. In addition to the 31 compounds already described, other 17 new compounds corresponding to various isomers of six phenolic acids: gallic acid, coumaric acid, hydroxybenzoic acid, caffeic acid, ferulic acid and protocatechiuc acid, were tentatively identified. Five compounds were detected responding to the molecular formula C<sub>7</sub>H<sub>6</sub>O<sub>3</sub> with a [M-H]<sup>-</sup> m/z value of 137 which would correspond to the structure of the hydroxybenzoic acid. Five other compounds with the same m/z value of 193 which would correspond to ferulic acid isomers [M-H]<sup>-</sup>, were detected. Finally, it was tentatively identified two isomers of caffeic acid (C<sub>9</sub>H<sub>8</sub>O<sub>4</sub>) with a majority line of fragmentation m/z 135 corresponding to C<sub>8</sub>H<sub>7</sub>O<sub>2</sub><sup>-</sup> [M-COOH]<sup>-</sup>; and other two isomers for protocatechiuc acid with a main line m/z 109 that would mean the same loss of the acid group. The quantification of the compounds identified in the 25 guayule and hybrid accessions was carried out using 10 commercial standards.

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## GUAYULE RESIN AND PORMENORIZED GUAYULIN CONTENT BY NEAR-INFRARED SPECTROSCOPY (NIR)

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Although Guayule (*Parthenium argentatum* A. Gray) is known for its potential to replace natural rubber from *Hevea brasiliensis*, recently another fraction of its composition, the resins, is the focus of many studies with commercial applicability. Belonging to such resin fraction are guayulins (A, B, C and D) which belong to the sesquiterpene family. Usually, guayulins are analyzed by HPLC-DAD and more recently with other analytical techniques including LC-MS, ESI-TOF and MALDI-TOF. The introduction of these techniques in the routine analysis of guayulins is not always possible due to the cost. Near-infrared spectroscopy (NIR) has been successfully used in guayule fresh and dried biomass to assess moisture, rubber and resin total contents. The purpose of the present study is to estimate, for first time, pormenorized guayulin content in guayule dry stems, using NIR spectroscopy. A set of 144 samples grown in Santa Cruz de la Zarza (Toledo, Spain), were analyzed by a Perkin Elmer Spectrum One FT-NIR equipment coupled with a Near Infrared Reflectance Accessory (NIRA). Data collection was acquired over a wavelength range of 750-2500 nm. In addition, guayulins A-D standards isolated in our lab were scanned to generate the best partial least squares regression (PLSR) model. The best correlative PLSR models for resin and guayulins were developed within the range of 1100-2500 nm, showing an excellent calibration correlation ( $R^2_c = 0.91-1.00$ ) and cross validation ( $R^2_{cv} = 0.87-0.91$ ). The residual predicative deviation (RPD) was higher than 3 in the case of resin and guayulins A, B and D, while in guayulin C was 2.8. These high RPD values demonstrated a good prediction power of the model, since values greater than 2 can be applied to rough screening, and RPD greater than 3 can be interpreted as good in control quality of NIR models. In conclusion, the use of NIR spectroscopy for guayule resin and guayulins estimation, is a good option for routine analysis versus traditional techniques that they are time-consuming and labor-intensive.

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## COMPARISON OF ACCELERATED SOLVENT EXTRACTION EQUIPMENT'S FOR RESIN AND RUBBER DETERMINATION IN GUAYULE STEM

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Guayule (*Parthenium argentatum* A. Gray) extraction at laboratory scale is carried out by accelerated solvent extraction (ASE) which employs sequential extraction, first with acetone to extract the resin fraction and then with hexane to extract the high molecular weight rubber, then yields are gravimetrically determined. Most guayule research groups have a Dionex ASE 200 or 350 extractors (Dionex Corp., Bannockburn, IL) which can process up to 24 samples in each worksheet. Recently a Bucchi E-914 equipment, with a four-channel simultaneous extraction system has been proposed to extract guayule rubber and resin. In this study, a comparison assay with both Dionex 350 and Bucchi E-914 ASE equipment have been carried out to check gravimetric results for guayule resin and rubber quantification. Four guayule stem samples (dried and powdered at 0.5 mm particle size) were extracted by duplicate in each equipment using similar extraction conditions: 3 consecutive cycles of 20 min at 100 bar with acetone at 40°C for the resin fraction, and then 3 cycles of 20 min at 100 bar with hexane at 120°C for the rubber fraction. Sample preparation method selected for each equipment was the optimal according to the literature: two grams of guayule mixed with 9 g of sand in a 22 ml cell for the Dionex 350, and one gram of guayule dispersed in 32.5 g of sand in an 80-ml cell for the Bucchi E-914. All extracts were evaporated at 50°C under reduced pressure and the yield of the fractions was calculated. Results indicated that resin extraction was significantly higher with Dionex than with Bucchi equipment, although a pormenorized analysis of guayulins within this fraction by HPLC-DAD showed that there were not significant differences within the total guayulin content. In case of rubber extraction, no significant differences were found between equipment's.

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# **ABSTRACTS FIBERS & CELLULOSICS CROPS DIVISION**

**ORAL PRESENTATIONS**

**CHAIR**

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## BIOBASED PLASTICIZER AND CELLULOSE NANOCRYSTALS IMPROVE MECHANICAL PROPERTIES OF POLYLACTIC ACID COMPOSITES

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Biopolymers are a fundamental part of contemporary life due to their biodegradable and biocompatible nature. Among different biopolymers, polylactic acid (PLA) with high processability is one of the most commercially available biopolymers used in different industries. However, the low ductility and impact strength limit its application in commercial plastic products. In this regard, one of the possible methods to enhance the ductility of PLA is adding different biobased reinforcing agents and plasticizer into PLA matrix. Furthermore, several researchers reported enhancement in PLA ductility through incorporating different fibers such as multi-walled carbon nanotubes, chopped carbon fiber, and graphene etc. The objective of this research was to use biobased materials for improving the mechanical properties of PLA composites. Biobased materials cellulose nanocrystals (CNC) and distillers dried grains with solubles (DDGS) were selected as reinforcing agent and polyethylene glycol (PEG) as a plasticizer. Biocomposites containing distillers DDGS (10%), CNC (0.25-1%), PEG (2%), maleic anhydride (0.25%) and PLA (86.75-87.75%) were manufactured using twin screw extruder. To investigate the effects of CNC, PEG, and maleic anhydride on PLA composites the morphological, ductile, and mechanical, properties of the composites were examined. Morphological changes were characterized by using optical microscopic images, chemical changes using infrared spectroscopy, mechanical properties by differential scanning calorimetry and ductility by tensile testing. The results demonstrate that addition of CNC and PEG to PLA improved its mechanical and ductile properties whereas maleic anhydride helped in dispersion of CNC. The observed improvements were attributed to the increased DDGS-CNC-matrix interactions. Optical microscopy also revealed some non-uniform dispersion of CNC. Overall, the results suggest that addition of CNC, plasticizer and coupling agent can aid in improving the physical, mechanical properties of PLA composites.

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## CHARACTERIZATION OF CHITOSAN BIOFILMS REINFORCED WITH NANOCELLULOSE EXTRACTED FROM DIFFERENT LIGNOCELLULOSIC BIOMASSES

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The search for more ecological and biodegradable materials, and of renewable origin represents currently a key focus of the scientific community to combat the massive use of non-renewable and non-biodegradable resources that are contributing to severe levels of pollution. Wastes from lignocellulosic sources are valuable in cellulose, hemicellulose, and lignin which, when recovered and separated, can give rise to innovative value-added products. Nanocellulose (NC) is a suitable example that has applications in many different areas, ranging from automotive manufacturing, medicine, food industry, and extending to the energetic field. In the food industry, the change from traditional petroleum-based plastics to biodegradable polymer materials is causing some constraints, namely the pauper mechanical, thermal, and barrier properties are restricted the opening of biopolymers to the markets. The insertion of homogeneously scattered nanoparticles, like nanocellulose, as a reinforcement agent, into the biopolymer matrix is seen as a promising possibility to surpass these shortcomings, generating bionanocomposites. Therefore, the aim of this work was to test NC obtained from three different lignocellulosic biomass (*Miscanthus × giganteus* Greef et Deu., *Arundo donax* L., *Hibiscus cannabinus* L.) as reinforcement in chitosan biofilms. The nanocellulose was obtained via an alkaline pre-treatment approach diligent to the different biomasses, pursued by acid hydrolysis. These NC were incorporated in chitosan at different rates (1.5%, 2%, and 2.5% w/w) and the bionanocomposites were characterized: X-ray diffraction, morphological characterization, thermal analysis, FT-IR, mechanical properties, thickness, optical properties (opacity and transparency), surface color, permeability (oxygen and water vapor), solubility, swelling degree, and contact angle. Commercial nanocellulose at the same rates (1.5%, 2%, and 2.5% w/w) were also tested for comparison. A biofilm made with pristine chitosan was used as the control. The results confirmed that the nanoparticles improved the mechanical properties of the chitosan biopolymer as planned. At the rate of 2.5% NC, it was possible to achieve near 40% increment in Tensile Strength (TS), 60% in Elastic Modulus (EM) and reduction of Elongation at break (% EAB) by nearly 75%, compared with pristine chitosan films. Further, bionanocomposites are slightly more saturated and showed greater ultraviolet light block than the pristine chitosan films. No significant differences were observed among the characteristics of the films produced with nanocellulose from giant reed, *Miscanthus*, Kenaf, and the commercial NC. Hence, results indicate that residues from those three lignocellulosic crops may afford a source of NC for the production of bionanocomposites. Additionally, the incorporation of NC at the rate of 2.5% presented the most encouraging results, by improving considerably the mechanical properties of the films and by reducing the oxygen and water vapor permeability, essential features in the use of biofilms by the food packaging industry.

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## GIANT REED HYDROLYSATE FOR SINGLE CELL OIL PRODUCTION BY OLEAGINOUS YEASTS *LIPOMYCES STARKEYI* AND *RHODOSPORIDIIBOLUS AZORICUS*

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Giant reed (*Arundo donax* L.) is gaining interest as an alternative sustainable feedstock for microbial oil production with different industrial applications (biodiesel, bio-lubricants, bio-based chemicals). Single cell oil (SCO) can be obtained in a bio-refinery context from the fermentation of sugar-rich hydrolysates of lignocellulosic biomass by oleaginous yeasts, provided that a pre-treatment step is performed to remove lignin and expose cellulose to enzymatic attack. Giant reed has many favorable characteristics, like very high productivity and low input requirements, however, it is relatively recalcitrant to enzymatic hydrolysis.

This work aimed to assess the suitability of giant reed hydrolysate for SCO production by the yeasts *Lipomyces starkeyi* and *Rhodospiridiobolus azoricus*. A mild alkaline pre-treatment at 120 °C followed by filtration and enzymatic hydrolysis, yielded 489±18 mg of total reducing sugars per g of initial untreated finely milled giant reed dry biomass. The hydrolysate showed a very high C/N ratio (196) and it was used to formulate several media with 27.5 g/L of reducing sugars and different C/N ratios. In optimal condition, after 120 h, *L. starkeyi* and *R. azoricus* reached a lipid content of 71% and 60% w/w and lipid yield of 4.9 and 5.2 g/L, respectively. Oleic, palmitic, and stearic acid content was 42%, 42%, and 3% with *L. starkeyi*, and 43%, 25%, and 21% with *R. azoricus*. Finally, a high biomass-to-lipid conversion was obtained with both strains (8.5% and 9.4% w/w as g of lipids per g of raw giant reed biomass, with *L. starkeyi* and *R. azoricus*, respectively). In conclusion, giant reed appears a suitable feedstock for biodiesel, bio-lubricants or other bio-based chemical productions. This work was supported by the Italian Ministry of Agricultural, Food and Forestry Policies (MiPAAF) under the AGROENER project (D.D. n. 26329, 1st April 2016) - <http://agroener.crea.gov.it>.

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# ENHANCING UV-SHIELDING AND MECHANICAL PROPERTIES OF POLYLACTIC ACID NANOCOMPOSITES BY ADDING LIGNIN COATED CELLULOSE NANOCRYSTALS

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From the perspective of sustainable development, it is desired to develop biodegradable UV-resistant and mechanically robust materials. Lignin nanoparticles (LNP) are renewable and commercially available nanoparticles with promising UV-shielding property characteristics. However, the addition of LNP can deteriorate the mechanical properties of lignin-based composites. On the other hand, cellulose nanocrystals containing lignin can serve as UV shielding reinforcing nanofiller without sacrificing the mechanical properties of nanocomposites. In this work, the UV-shielding performance, mechanical properties, and biodegradation abilities of PLA nanocomposites containing lignin coated cellulose nanocrystals (LCNC) were compared to PLA nanocomposite reinforced with lignin nanoparticles (LNP). PLA nanocomposites with different content of nanofillers (i.e., 1, 3, and 5%) were prepared via masterbatch approach followed by extrusion and injection molding. The morphologies of PLA nanocomposites studied by SEM images, confirmed smooth fracture surface with micro size LCNC aggregates, however, rough fracture surface and uniform nanoparticle dispersion was observed in PLA nanocomposites reinforced by LNP. Compared to neat PLA, the thermomechanical performance of PLA-3% LCNCs was improved by 21%, while 3% LNP improved the storage modulus by 11%. Results from UV-Vis characterization revealed a synergic effect of LNP and LCNC nanostructures in terms of UV light blocking ability. By the addition of high content of 5wt % LCNC into PLA matrix, the highest UV radiation was blocked out by (75.27%) of UV-A and (82.81%) of UV-B. It was found that the maximum weight loss of PLA nanocomposites after being hydrolytic degraded for 20 days increased from 8% for PLA to 14% for PLA-5 wt% LCNC, and 12.7% for PLA-5%LNP. This study is expected to provide meaningful insights into PLA nanocomposite reinforced with LCNC and LNP.

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## GROWING NOVEL PERENNIAL BIOMASS CROPS ON MARGINAL LANDS: CHALLENGES FOR PLANT BREEDING

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The biomass demand to fuel a growing global bio-based economy is expected to tremendously increase over the next decades, and projections indicate that dedicated biomass crops will satisfy a large portion of it. The cultivation of dedicated biomass crops raises concerns though, as it can subtract fertile agricultural land to food production, a situation already experienced during the expansion of first-generation biofuels a decade ago. The EU project MAGIC (Marginal Lands for Growing Industrial Crops) aims at circumventing food-vs-biomass land competition by enabling the use of marginal lands (MALs, currently not used by agriculture and of low natural value) for biomass supply. A set of perennial biomass crops, which includes Miscanthus (*Miscanthus x giganteus*), Switchgrass (*Panicum virgatum* L.), Giant reed (*Arundo donax* L.), Poplar (genus *Populus*), Willow (*Salix alba* L.), Eucalyptus (*Eucalyptus obliqua* L'Hér.), and many others, appears particularly suitable for cultivation on MALs, due to their natural ability of withstanding suboptimal growing conditions. Nevertheless, the biomass yield and quality of current varieties of these crops can still be significantly reduced by severe droughts, salinity, harsh soils, and many other constraints that are often encountered on MALs. Therefore, the release of plant varieties that not only survive, but produce large, constant, and good-quality biomass yield under MALs conditions will be crucial for deploying MALs for biomass production. Plant breeding appears therefore the key to enable this vision, and ideotype breeding could be applied to several novel biomass crops by targeting all the traits that concur to the formation of biomass yield and to its quality. These traits include crop architecture and phenology, efficiency in the use of resources, lignocellulose composition in relation to bio-based applications, and tolerance to abiotic stresses. The quick release of advanced varieties of perennial biomass crops is however currently hampered by the lack of both genetic resources and breeding tools for most of these crops. This bottleneck could be overcome by using the knowledge and tools available in model (biomass) crops for the traits of interest, and by transferring those to novel (orphan) species. In this context, conservation of candidate genes, quantitative trait loci (QTLs) and molecular markers across plant species through genome synteny appears an effective strategy to operate the transfer of genetic knowledge between crops and provide perennial biomass species with effective breeding tools in short time. This talk will discuss the feasibility of this approach, as well as the potential of using “universal” QTLs and markers for breeding orphan biomass crops and enable a sustainable use of MALs for biomass provision.

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## HOW THE IRRIGATION AFFECTS THE MISCANTHUS YIELDS IN THE DRY MEDITERRANEAN REGION

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Miscanthus (*Miscanthus X giganteus*) is a perennial grass with a lifetime 15 to 20 years. It provides lignocellulosic feedstock for advanced biofuels (bioethanol), bioenergy production (pellets and briquettes, etc.) and biobased materials (building materials, etc.). In Europe it was grown in an area around 20.000 ha and it is being investigated in many European research projects with emphasis on marginal and/or contaminated lands. In view of this research work miscanthus growth and yields were evaluated under two factors for a decade in central Greece. The tested factors were: plant density (two plant densities; D1: 50 cm and D2: 100 cm within the rows and 100 cm between the rows) and irrigation effect (four levels of irrigation; I<sub>3</sub>: 100 % of PET, I<sub>2</sub>: 50% of PET, I<sub>1</sub>: 25% of PET and I<sub>0</sub>: no irrigation) in three blocks. The field trial was established by rhizomes in early 2012 (6/3/12). Each year the following measurements had been carried out: a) phenological observations (date of sowing/establishment, emergence date, flowering time and harvesting time), b) growth data (plant height, plant/tiller density, stem diameter), c) yields data (at the end of the first growing period only a final harvest was carried out for yields estimations and samples collection for laboratory analyses) and d) laboratory analyses (dry matter content, proximate and elementary analysis, gross and net calorific value). The maximum biomass yields were recorded from the 2<sup>nd</sup> till the 4<sup>th</sup> growing season, while from the 5<sup>th</sup> growing season the yields were declined and remain at this level till the end of the beginning of the 10<sup>th</sup> growing season. Irrigation plays a key role in the biomass yields with double yields in the fully irrigated plots compared to unirrigated ones. The irrigation effect was stronger in these years that the hot months were extremely dry and less strong on those that some rainfalls were recorded. The dry matter yields, averaged overall irrigation effects, varied from 12 t/ha (establishment year) to 18 t/ha (2<sup>nd</sup> growing season). The high density help the crop to have better weed competition and higher yields at the establishment year but thereafter no effects of plant density were recorded on the yields. It was also found that from the 2<sup>nd</sup> till the 4<sup>th</sup> growing cycle slighter higher yields were recorded in the plots with the low density. At the final harvest, 20-35% of the harvesting material corresponding to leaves. It should be pointed out that the lower the leaf percentage is on the harvested biomass the higher the calorific value is. The ash content of the harvested material varied from 3 to 4% (20-35% leaves; up to 2% on the stems and around 12% on the leaves).

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## SOIL WATER AVAILABILITY ON BIOMASS YIELD AND WUE OF PERENNIAL GRASSES IN A SEMIARID AREA

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Lignocellulose is the most abundant no-food raw material on Earth suitable to advanced bioconversion technologies for bioenergy and added value bio-products. Perennial bioenergy grasses (PBGs) have been studied in a countless environmental and agronomic management conditions, and nowadays have been recommended as ideal crops suitable to areas affected by biophysical constraints. Dryness significantly limits crop growth and yield leading to land degradation and abandonment. In these limiting conditions farmers are not willing to grown food crops since not able to reach a marginal productivity, therefore bioenergy crops with exceptional drought tolerant traits are required mitigate land abandonment while providing raw material for the bioeconomy development. The present work compared six PBGs under three irrigation levels in a semiarid Mediterranean area affected by dryness (as ratio between precipitations and potential evapotranspiration) for three consecutive growing seasons on field. A split-plot experimental design was employed, where the main plot was the irrigation, with 3 levels: 100% (I100), 50% (I50) and 0% (I0) of maximum crop evapotranspiration (ET<sub>m</sub>) restoration during the summer months (June-August), and the genotype the second factor, assigned to the sub-plots within the main irrigation plots. Each combination of irrigation and genotype was replicated 3 times within the main plots. The six genotypes were were: two *Arundo donax* L. ecotypes, named ARCT and ARMO (clone Fondachello and clone Morocco), the commercial *Miscanthus x giganteus* (greef et Deuter) named MxG, two seed-based *Miscanthus* hybrids obtained from the breeding program led by the IBERS, Aberystwyth University (UK) and Terravesta Ltd (UK), named GNT9 and GNT10, and one ecotype of *Saccharum spontaneum* L. ssp. *aegypticum* Willd (Hack.), named SAC. At the first year, the irrigation was not differentiated and 100% of ET<sub>m</sub> restoration was supplied during the summer months to all genotypes. The three growing seasons were characterized by similar air temperature, global solar radiation and reference evapotranspiration during crop growth; on the other hand, precipitation amount and distribution greatly changed being the highest on 2020/21 and the lowest on 2019/20 growing season. Biomass dry matter yield was the highest in SAC at and the lowest in MxG at the first harvest. It increased significantly in all genotypes at the second and at the third year. Averaged the three growing season, ARMO, ARCT and SAC were the most productive. Among *Miscanthus*, GNT9 and GNT10 outperformed MxG. The irrigation effect significantly increased biomass dry matter yield in all genotypes and growing seasons, but to a different extent: the relative yield increase (RYI) was the lowest in the GNT10 and the highest in MxG between I0 and I100. Between I0 and I50, the RYI was the highest in MxG and SAC and the lowest in ARMO, while between I50 and I100 it was the highest in ARMO and the lowest in SAC. The crop WUE also changed among genotypes and irrigation treatments: it increased as the irrigation was raised in those genotypes using a non-conservative growth strategy; on the other hand, WUE was the highest under rainfed conditions in genotypes using a conservative growth strategy.

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## TOLERANCE TO AND ACCUMULATION OF CADMIUM IN THREE BAST FIBER CROPS

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As current environmental pollution is getting more serious, researchers and industries seek for environmentally friendly materials as alternatives for conventional ones. The usage of bio-based materials -such as natural fibers- is on demand, and fiber crops are gaining more and more interest. At the same time, cadmium (Cd) accumulation in soils is of great concern due to its potential toxicity to biota at low concentrations. In the present study, three fiber crops were tested as possible alternative species for the phytomanagement of contaminated land. Industrial hemp (*Cannabis sativa* L., var. Yunma 1), flax (*Linum usitatissimum* L., var. Y2I329) and kenaf (*Hibiscus cannabinus* L., var. GGS) were investigated in a pot experiment for their tolerance to Cd and for their soil remediation potential. The soil used was collected from Yuanjiang City, Hunan Province, China, and two CdNO<sub>3</sub>.4H<sub>2</sub>O quantities were added onto the soil in a form of water solution, mixed evenly, and kept for two months for metal equilibration. The seeds of each crop were sown into the pots on March 4, 2020, and the experimental design was the completely randomized with 29 replicates. The final concentrations of Cd in the pot soils were: Cd0 (control, no addition of the metal), Cd40= 40 mg/kg, and Cd80=80mg/kg. The results showed that plant height for all three fiber crops was significantly reduced in all treated plants as the metal concentration in soil increased. The number of leaves and the stem diameter of hemp and kenaf plants were significantly affected by both Cd40 and Cd80 compared to Cd0. In flax, the number of leaves remained unaffected, while the stem diameter was negatively affected only in Cd80. The flax dry biomass of the aerial plant part was reduced by 33% in Cd40 and 51% in Cd80 compared to Cd0, while the highest reduction was observed in hemp plants treated by Cd80, which measured to be 89%. However, in Cd40 the corresponding reduction for hemp plants was only 1%. In kenaf the dry biomass was slightly increased by 7% and 5% in Cd40 and Cd80 respectively. Cadmium concentrations in aboveground tissues of industrial hemp, flax and kenaf significantly increased with increasing metal concentrations in soil (p<0.001). As expected, the highest Cd concentrations were observed in Cd80 treatment and were up to 279.0 mg/kg for hemp, 289.7 mg/kg for flax and 250.0 mg/kg for kenaf respectively. The corresponding concentrations for Cd40 were 70.6, 186.0 and 188.0 mg/kg respectively. In conclusion, flax and kenaf seems to be more tolerant than hemp when exposed to high Cd soil contents, and for phyto-management purposes of contaminated sites it would be proposed to follow the order kenaf>flax>hemp. However, further investigation under field conditions is needed.

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# **ABSTRACTS FIBERS & CELLULOSICS CROPS DIVISION**

**POSTERS**

**CHAIR**

**EFTHYMIA ALEXOPOULOU, CRES, PIKERMI, GREECE**

## UNDERSTANDING THE POTENTIAL OF KENAF (*Hibiscus cannabinus* L.) IN SOILS CONTAMINATED WITH HEAVY METALS IN MOZAMBIQUE

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The increasing demand for biomass for the production of bioenergy and biomaterials is generating land-use conflicts which might be avoided through the establishment of dedicated industrial crops on marginal land, e.g. heavy-metal contaminated land. Yet, heavy metals contaminated soils might induce the reduction of crop yields and the quality of agricultural products, desertification, and the loss of ecosystem services. Therefore, assessment of biomass from marginal land should take into account constraining factors, such as productivity and biomass quality. Hence, the aim of this work was to study the effects of different types of soils contaminated with heavy metals (Chromium, Copper, Lead and Zinc) on growth and productivity of kenaf, in Mozambique. The study was performed in a pot essay and the plants were tested in sandy soils and clay soils. The soils were artificially contaminated, and the concentrations chosen were based on the limits established by the Decree Law 276 of 2009 (Portuguese regulation that establishes the regime for the use of sewage sludge in agricultural soils) - Zn: 450 mg/kg; Cr: 300 mg/kg; Pb: 450 mg/kg and Cu: 200 mg/kg. Results indicate that all the contaminated soils did not affect the growth and yields of kenaf in clay soils. In sandy soils, the pattern observed was completely different. The yields in sandy soils were much lower than what was observed in the clay soils (*circa* 60% less than yields obtained in clay soils). Also, in sandy soils, kenaf showed less tolerance to the contamination with Cu, Cr and Pb. In fact, in soils contaminated with Cu and Cr there was no germination of the seeds, and in Pb contaminated soils, the yields were much lower than in the control (a reduction of 40% was observed). Kenaf showed also in sandy soils tolerance to Zn once yields remain similar to the ones obtained in control. In terms of biomass quality, the ash content and the fiber content of the plants obtained in contaminated clay soils was similar to the ash content and fiber content of plants harvested from clay control soils. The same was observed for the plants collected from the Zn contaminated sandy soils. But in the Pb contaminated soils, the ash content and the fiber content was higher than what was quantified in the plants harvested from the control sandy soils. This means that the technological exploitation of the biomass collected from Pb contaminated sandy soils may be limited. In terms of phytoextraction capacity, the plants extracted more Zn than Pb, Cu and Cr, which are much less mobile in the plant. But, for all the metals studied, kenaf showed phytoremediation capability.

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## LIFE CYCLE ASSESSMENT OF SUPERCRITICAL EXTRACT OBTAINED FROM POPLAR BIOMASS

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The aim of this study was to determine the environmental impact of the production chain of a supercritical extract from poplar biomass by means of an attributive life cycle assessment and to locate "weak links" (with significant environmental impact) in the bioproducton chain. The main function of the product system was the production of poplar (*Populus tremula* L.) biomass extract together with heat from pellets produced from post-extraction biomass. The functional unit was 1 kg of packed dry extract. The system boundaries adopted in the analysis was from the cradle to the gate of the conversion plant and included the following production subsystems: (1) poplar biomass production in 20 annual harvest cycles; (2) logistics and transport, including raw material pretreatment, (3) obtaining supercritical poplar extract and co-products in a biorefinery. The results showed that the poplar biomass extraction stage with the use of supercritical CO<sub>2</sub> had the highest share in all but two analyzed categories of environmental impact of the ReCiPe 2016 midpoint method. It should be noted that the production of heat from pellets produced from post-extraction biomass reduced the environmental impact in almost all impact categories. The use of the ReCiPe endpoint method also showed the highest environmental impact of the supercritical extraction process in all three categories of this method (human health, ecosystems and resources). The normalized results showed that the production of dry extract caused the greatest damage in the human health category, even though its impact was lowered by avoiding heat production from pellets. The life cycle assessment of the entire extract production chain showed that subsystem 1 (plant production) and 2 (biomass logistics) had a slight environmental impact compared to subsystem 3 (production of dry packed extract). This last stage had a very high environmental impact, mainly due to the use of fossil fuels for the production of electricity and steam. A positive aspect of the tested production system was the use of post-extraction biomass for the production of pellets for heat generation. In this way, an avoided product is created, i.e. instead of producing pellets from wood biomass, pellets from post-extraction poplar biomass for heating purposes were used. As a result, the environmental impact is reduced. Definitely, the use of fossil fuel energy still remains the "weak link" in the production chain. Therefore, in order to reduce the environmental impact of extract production, it is proposed to use renewable energy sources in the energy mix supplied to the biorefinery. Of course, such a production system should also undergo an economic analysis in order to validate its economic feasibility.

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## LASER RADIATION TO STIMULATE TOBACCO GROWTH

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During the last 50 years the progress of chemical technology provoked production revolution in the agriculture. Many chemicals are used for fertilizing crops, controlling pests and helping to develop a highly successful farm system. The long-term use of chemicals in agricultural production resulted in a decrease in plant resistance, plant yield and soil yielding capacity. The negative effects of chemicals on the agricultural produce and on the environment are commonly known. Therefore, many scientists believe that this century is the century of biophysical methods in agriculture. One biophysical method for plant growing stimulation is laser irradiation of seeds before sowing. The subject of this study were three genotypes of Virginia tobacco (*Nicotiana tabacum* L.) irradiated with a red laser ( $\lambda=655$  nm,  $P=0,7$  mW) with different duration (30 s, 60 s, 90 s, 120 s). The aim of the present study was to trace the period of seedling formation and to establish the effect of laser irradiation on the duration of the phenological growth stages. Mathematical and statistical processing methods are applied to the database of experimental data. It was found that pre-sowing laser irradiation of tobacco seeds with a red laser led to an acceleration of germination, bigger plant height and higher plant weight. The results show that the wide application of pre-sowing laser irradiation of tobacco seeds will enable intense and more qualitative tobacco leaf production, as well as the protection of the environment.

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## POTENTIAL SOC ACCUMULATION OF TWO FIBER CROPS

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Industrial hemp (*Cannabis sativa* L.) and kenaf (*Hibiscus cannabinus* L.) are two high yielding annual fiber crops (stem cellulose and hemicellulose content of 76% and 54% for hemp and kenaf, respectively) with great potential for their multiple biorefinery applications. The products from their fiber have already been used in automotive, gardening and construction sectors for their unique characteristics such as breathability, thermal and electrical insulating, fire and soundproofing. Including them into common food crops rotation might significantly increase the sustainability of cropping systems through crop diversification, biodiversity and weed suppression properties. Hemp and kenaf aboveground biomass productivity have been extensively studied, whereas little information is available on their potential in increasing soil carbon storage in soils. Soil respiration and root biomass are responsible of the stability of the SOC and of the increase in the soil carbon content, respectively. The objective of the present study was to determine hemp and kenaf potential to increase SOC compared to fallow. A plot trial was set up at the experimental farm of Bologna University (Italy, 44°30' N, 11°21' E) in spring 2018 in a randomized complete block design ( $n=4$ ). Kenaf average soil respiration during the growth cycle registered 0.32 g CO<sub>2</sub> m<sup>2</sup> h<sup>-1</sup> whereas hemp 0.24 and the control fallow 0.43. The higher soil respiration on fallow is probably due to a generally higher soil temperature (24°C) that increases the mineralization of the SOC, whereas hemp and kenaf recorded an average soil T 1.5 °C lower. The measured root biomass was 0.68 for hemp and 1.02 Mg ha<sup>-1</sup> for kenaf (+ 50%). Even though the relatively high respiration rates, it could be said that kenaf and hemp thanks to the large root biomass produced (and the relative distribution) can potentially contribute to a greater permanence and accumulation of SOC in the subsoil, thus significantly contributing to physio-chemical characteristic of the soil compared to a fallow situation.

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## COULD RAMIE BE AN ALTERNATIVE CROP FOR REMEDIATION OF CONTAMINATED SOILS IN SOUTH EUROPE?

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Ramie (*Boehmeria nivea* (L.) Gaudich.) is a perennial herbaceous fiber crop belonging to the Urticaceae family. It can grow 1 to 2 m height, producing high shoot biomass and having multiple harvests under favorable environmental conditions. It is an ancient plant with vigorous growth, high nutritional value and multipurpose applications in textiles, livestock feed, environmental conservation and medicine. The production of ramie fiber is about 500,000 tonnes per year in China, accounting for 96–97% of world production. It has been reported that ramie can grow and colonize diverse heavy metal contaminated lands, possessing a certain degree of tolerance to Pb, Zn, Cd, and As, and accumulating a certain degree of heavy metals in all plant parts, especially in roots. The aim of this work was to investigate the potential of ramie to grow in a long term and multi elemental contaminated mining site in Greece contributing to soil remediation. Surface (20 cm depth) soil was used, taken from a mining site located 50 km S-SE of Athens. This soil was mixed with an uncontaminated soil in three ratios: 100% of uncontaminated soil (treatment T0-control), 50% of both soils (treatment TI), and 100 % of contaminated soil (treatment TII). The aqua regia (3:1 mixture of hydrochloric acid and nitric acid for total) and DTPA extractable metal concentrations were measured. Ramie plants were grown for 9 months in pots containing the three soil mixtures. The pots were arranged according to a completely randomized design with three replicates. After harvest, the concentrations of heavy metals in plant tissues were determined by atomic absorption spectrometry (AAS). The results of this work confirm that ramie is generally tolerant to heavy metals and especially to high soil contents of Cd and Zn. It is an excluder of Ni and Cr since their concentrations in plant tissues were below the detection limits of the AAS. The crop could accumulate Cu and Mn, but in concentrations within the normal limits as the higher concentrations measured were up to 13.1 and 139.6 mg/kg respectively. Concentrations higher than normal limits were measured in both TI and TII for Cd, reaching the 4.9 and 13.7 mg/kg respectively, while the corresponding concentrations of Zn were 348.7 and 490.5 mg/kg. Therefore, ramie could be cultivated for phytoextraction purposes in Cd and Zn contaminated soils contributing to the attenuation of these elements in mining sites. In addition, this crop could be used for the phytomanagement of multimetal contaminated areas since possesses both high biomass and high economic value.

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# **ABSTRACTS OILSEEDS DIVISION**

**ORAL PRESENTATIONS**

**CHAIR**

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## SCREENING OF SPECIALIZED METABOLITES IN SIX CAMELINA VARIETIES

Barbara Alberghini<sup>1</sup>, Federica Zanetti<sup>1</sup>, Stephanie Boutet<sup>2</sup>, François Perreau<sup>2</sup>, Grégory Mouille<sup>2</sup>, Loïc Lepinieć<sup>2</sup>, Massimiliano Corso<sup>2</sup>, Angela Vecchi<sup>1</sup>, and Andrea Monti<sup>1</sup>

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Camelina (*Camelina sativa* L. Crantz) is an ancient oilseed crop gaining attention in recent years thanks to its adaptability to various environments and growing conditions, its resistance to pests and diseases, and its peculiar oil composition, characterized by an unusually high content of n-3 polyunsaturated fatty acids (PUFA). Moreover, camelina seeds also contain specialized metabolites (SMs, formerly called “secondary metabolites”) that are retained into seed meal after oil extraction. Many SMs are well-known antioxidants with positive effects on human health (e.g., flavonoids). The present work aimed at comparing SMs content of six camelina varieties from different breeding programs, grown in five consecutive years. In particular, Midas was supplied by Agriculture and Agri-food Canada (Saskatoon, Canada); 789-02, Cypress and Pearl by Smart Earth Camelina (Saskatoon, Canada); Omega was provided by Poznan University of Life Sciences (Poznan, Poland); and WUR by Wageningen University and Research (Wageningen, The Netherlands). The trials were carried out at the experimental farm of Bologna University at Cadriano (Italy, 44°30' N, 11°21' E, 32 m a.s.l.) between 2015 and 2019. The site has a mean temperature of 13.2°C and an annual rainfall of 712 mm and it is characterized by a silty-clay-loam (37% sand, 22% clay, 41% silt) soil. Seeding rate (500 seeds m<sup>-2</sup>) and plot dimension (10.5 m<sup>2</sup>) were maintained during all years. Trials were sown in March and harvesting took place between mid-June to the beginning of July. Fertilization was manually supplied at stem elongation stage at a 50 kg ha<sup>-1</sup> rate, as urea. Weed control was performed manually, and the trials were rainfed. Representative seed samples were collected from each plot and analyzed by LC-MS metabolomics to determine SM content at the IJPB-INRAE institute (France), as well as SM classes identified. SMs detected in camelina were annotated as belonging to the main SMs classes of flavonoids (flavonols, flavan-3-ols (tannins), isoflavones, flavanones, and flavones), amino acids, cinnamic acids, glucosinolates, alkaloids, benzoic acids, terpenes, carboxylic acids. Regarding agronomic traits, the ANOVA showed that Cypress was the best performing cultivar, with significantly higher seed yield (1.78 Mg DM ha<sup>-1</sup>), TKW (1.49 g) and seed oil content (39.4%) among tested cultivars. SM content in camelina was highly influenced by the growing year. In particular, the minimum SM signal accumulation (1930 relative units) was reached in 2019, which was characterized by the highest cumulative precipitation and the lowest mean temperature during crop growing cycle. Furthermore, SM pattern was strongly affected by the year and in some cases also by the cultivar. Namely, 2016 showed the maximum annotated accumulation of alkaloids (43.5 relative units), amino acids (214.1 relative units) and glucosinolates (106.6 relative units); while in 2019 the highest signal of flavanones (0.07 relative units) and isoflavones (1.5 relative units) was reached. Concerning cultivars, among all years Omega presented the highest signal of alkaloids (35.2 relative units); while Cypress showed the top signal levels of flavan-3-ols (56.3 relative units), flavanones (0.05 relative units), flavonols (309.7 relative units) and isoflavones (1.13 relative units). Finally, the Pearson correlation test revealed a significant positive correlation between protein content and cinnamic acids ( $r=0.99$ ) and between protein content and flavones ( $r=0.89$ ) in the cultivar 789-02. Significant negative correlations were found in the cultivar 887 between TKW and flavanones ( $r=-0.91$ ), and in cultivar Omega between oil content and flavan-3-ols ( $r=-0.90$ ). The presence of SMs in camelina can be a further launching ramp for its use in nutraceutical applications, improving the interest and the importance of this emerging crop. Anyway, the influence of growing conditions should be taken in strong consideration and further research is needed to predict the final composition of SM classes in camelina seeds, at least to some extent.

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## LONG-TERM FIELD SCREENING TRIALS FOR CAMELINA IN GREECE

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Camelina (*Camelina sativa* L.) is a novel oilseed crop for the European biobased industry. In South Europe, it can be grown both as winter and spring crop. It has a short growing cycle (90 to 120 days as spring crop) and it is considered a great candidate as cover and/or catch crop. The purpose of this research work was to compare the adaptability and productivity (seed and oil yields, residual biomass) of several camelina varieties for a period of five subsequent years (2015-19) in central Greece. A total number of twenty-four varieties/lines (789-02, 787-05, 787-06, 787-08, 787-09, 787-15, 787-22, 787-27, 886, 887, 886-02, 886-05, 886-08, 886-09, 1040-0, 1040-04, 1043-01, 1043-04, 1043-05, 1047-01, Midas, Omega, WUR2015-001, Joelle) have been tested. Apart from Omega, Joelle and WUR2015001, all the other varieties had been provided by Smart Earth Camelina Corp (Canada). The variety WUR2015001 had been provided by Wagenigen Research Institute. In one of them the sowing took place in winter (13/12/17), while in the others in spring (6/4/15, 21/3/16, 20/3/17, 19/3/18 & 15/3/19). The experimental layout was a randomized complete block in three replications. The size of the experimental plots was 1.5x6 m<sup>2</sup> and the distances between the rows were 15 cm. In all trials, the plant density and the plant height had been measured at the end of the growing period. The harvest of the trials carried out from the second half of May to late June depending on the year and the tested variety. At the end of each growing cycle a final harvest of 5 m<sup>2</sup> (5x1) per plot took place and the harvested biomass had been first weighted and then the seeds were separated and weighted. Samples from both plant fractions (seeds and straw) had been collected for laboratory analyses (moisture content, oil and protein content of the seeds and biomass characterization for the straw). Camelina seed yields were strongly affected by the variety, the sowing date and the specific climatic conditions of each year. The maximum seed yields for all varieties recorded when the sowing took place in winter. Among the five subsequent spring field trials the least yields were recorded when the sowing was delayed till the beginning of April and the maximum when the sowing took place in the middle of March. Among the tested varieties the best performing, as spring crop, was 787-22 (1548 kg/ha), which as winter crop was Omega (2545 kg/ha). As a mean of all trials, the oil content of the seeds varied from 36.3% (787-27) to 39.8% (787-15) and the protein content of the seeds varied from 27.8% (787-15) to 30.1% (787-27). It should be pointed out that half of the tested varieties have mean seed yields around 1600 kg/ha, averaged overall trials, while the rest between 1300 and 1500 kg/ha. Although WUR2015001 had the smallest growing cycle, had quite high mean seed yields (~1600 kg/ha). The ratio between the seed yields and the residual biomass varied from 1:2 to 1:2.5. The ash content of the residual biomass was 6.96% [varied from 6.26 % (787-08) to 7.70% (887)] and the Gross Calorific Value 4341 kcal/kg (varied from 4232 kcal/kg 4402 kcal/kg). It can be concluded that camelina as spring crop can be an excellent catch crop with seed yields between 1.5 to 2.2 t/ha and oil content from 36 to 40%. Higher yields can be expected in areas with South Mediterranean climate if camelina will be grown as winter crop. Its residual biomass has lower ash content (from 6 to 7%) than other oilseed crops (like crambe, rapeseed) and can be further exploited as feedstock for bioenergy production.

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## SWATHING AS A SUITABLE ALTERNATIVE FOR HARVESTING CAMELINA

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Camelina (*Camelina sativa* L. Crantz) is an oilseed crop that has gained interest during the last decade as excellent feedstock for low carbon emission biofuel and other industrial uses. Seed oil content is higher than 35% (w/w) and the required low agricultural inputs make camelina a good candidate as rotation crop in cereal-based crop production systems. Autumn sowing in Mediterranean region implies the harvesting time ranging from the end of May and end of June. However, the period can be anticipated or delayed according to climate conditions. In areas, where double cropping with typical summer crops (i.e. soybean, sunflower, etc.) is feasible, the anticipation of camelina harvesting could represent an interesting opportunity for farmers significantly improving the establishment success of the second crop. Therefore, as the oil accumulation in camelina seeds peaks at 7 to 15 d before full ripening, a comparison in terms of harvesting cost, machine performance and seed loss were performed between swathing at maximum oil accumulation (H2) + combining and direct combining at full maturity (H1). Moreover, the study aimed to provide data on the possibility of anticipating the sowing of the following crop of camelina without affecting harvesting performance, seed losses and seed quality. A large plot trial was established at the experimental farm of the University of Bologna in autumn 2020. The camelina variety Alba, supplied by Camelina Company (Spain), was used in the trial. The agronomic management was defined as low input, consisting in minimum tillage for establishing the crop and a top-dressing N fertilization of 50 kg ha<sup>-1</sup>, as urea. The experimental design was a randomized complete block with 4 replicates. Swathing was carried out using a GS mower with 1.80 m working width. Cutting height was set up at 0.25 m, in order to ensure proper drying of the swathed plants. Harvesting was performed in both treatments with a combine harvester equipped with a 3 m wide cereal header. Concerning H2 treatment, camelina was cut on May 27 2020, when residual seed moisture was about 35%, and swathing was performed one week later, when seed moisture was below 7%. While the direct combining was performed when the seed moisture content was at about 12%, and it was performed on June 10 2021. Seed yield was 1.17 Mg DM ha<sup>-1</sup> and 1.53 Mg DM ha<sup>-1</sup>, in the H1 and H2 treatments, respectively. Results on working performance evaluation showed that double passage harvesting increased harvesting cost of only 16 %, but no increase in seed loss was observed. Considering what previously reported it is possible to harvest camelina with double passage harvesting, shortening the growing season and increasing the success of the establishment of the summer crop in double cropping systems. Nevertheless, the surveyed yield reduction should be carefully evaluated in further studies, even if the lower residual seed moisture in the H2 would improve the processing phase, since it will positively affect the efficiency of the mechanical seed pressing to obtain camelina oil and cake.

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## A LONG TIME COMING: DEVELOPMENT OF THE HERBICIDE RESISTANT CAMELINA CULTIVAR SES1154HR

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Camelina (*Camelina sativa* L. Crantz) is being developed as an alternative oilseed crop. The unique properties of its seed oil and favorable agronomic attributes make it an attractive option for farmers, particularly in the drier regions of the Canadian Prairies. Despite this, limited herbicide options for managing broadleaf weeds and camelina's susceptibility to Group 2 herbicide carry-over in the soil are major challenges that limit its adoption. Group 2 herbicides affect plants by inhibiting acetohydroxyacid synthase (AHAS), an enzyme involved in branched chain amino acid synthesis. In order to enable in-crop broadleaf weed control and re-cropping where soil residual Group 2 activity remains, camelina cultivar SES1154HR with resistance to the sulfonylurea thifensulfuron-methyl was developed at Smart Earth Camelina Corporation in collaboration with AAFC. To this end, camelina EMS mutant lines 12CS0365 and 12CS0366 with increased resistance to thifensulfuron-methyl were crossed, followed by repeated selfing, leading to homozygous double mutant line 13CS0786. Subsequently, the herbicide resistance (HR) trait was introgressed into cultivar SES0787LS via backcrossing, resulting in cultivar SES1154HR. A comparison of the AHAS gene sequences of 13CS0786 to those of wild-type camelina identified the same single point mutation in orthologues *CsAHAS1* and *CsAHAS3*, causing a Pro197Ser substitution in the resulting enzymes. Dose-response trials in the greenhouse showed that compared to the wild-type, doses of approximately 1000 times more thifensulfuron-methyl were required to reduce plant growth of 13CS0786 by 50%. Interestingly, the single mutant lines 12CS0365 and 12CS0366 expressed different levels of HR, despite carrying the same point mutation in their respective HR *CsAHAS* orthologues, which may be explained by differences in gene expression. Despite the hexaploid nature of the *C. sativa* genome and great sequence similarity among orthologues, KASP markers were developed that are specific for HR *CsAHAS1* and *CsAHAS3*, respectively. In field trials conducted in Saskatoon, SK, in 2018 and 2019, SES1154HR tolerated a 2x rate of thifensulfuron-methyl with little to no herbicide damage and no significant reduction in seed yields. Commercial seed of this cultivar will be available to producers in Canada and the US in spring of 2022 for contract production from Smart Earth Camelina Corp.

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## WINTER WEED SUPPRESSION CAPACITY OF CAMELINA [*CAMELINA SATIVA* (L.) CRANTZ]

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Camelina has proven to be a useful oilseed crop with diverse options for its products, oil and meal. Under the ongoing climatic change, its tolerance to drought is a highly desired characteristic in semiarid cropland areas, such as the Ebro basin in North-Eastern Spain. Despite this, it is a relatively unknown crop for farmers in these regions, who are also quite reluctant to include crops they do not feel secure with. In this work we demonstrate the value of this crop in the most important problem for farmers for field management, weed control, so that we add a strong argument, apart the value of the crop itself, to convince farmers to include camelina in their crop rotation systems. Camelina (cv. GP204) and barley (cv. Meseta) were sown in the experimental field of the University of Lleida at a rate of 8 kg/ha and 200 kg/ha each in November 2018 and with three replicates. In each plot, three 0.5 x 0.5 m<sup>2</sup> quadrates were selected and five individuals of *Papaver rhoeas* were left. The rest of the experiment was hand-weeded, and controls without the crop were left nearby the plots. The previous day to harvest, weed individuals were removed, brought to the laboratory, and fitness parameters were measured: height, number of branches, number of fruits, vegetative biomass and reproductive biomass. The experiment was repeated in November 2019. Results affected by climatic conditions each year: season 2018-19 resulted much drier than 2019-20, but in both they show that the capacity of camelina to reduce the growth of the winter weed *P. rhoeas* was significant, and similar to that obtained by barley in 2018-19. In contrast, in 2019-20 barley resulted more effective than camelina influencing the growth of *P. rhoeas*. The average height of the weed was similar in the three situations (25-30 cm) in the first year, but plants were higher in the control in the second one (95.5 cm > 50 cm and 57 cm in camelina and barley, respectively). The dry weight was significantly reduced from the control to competition with camelina in both years. The fruits of those *P. rhoeas* individuals grown in competition with barley and camelina were half size of the controls; consequently, the potential seed production was also smaller. Potentially the controls produced 3338 seeds/plant in 2018-19 and 164877 seeds/plant in 2019-20. Plants competing with camelina would have produced 966 seeds/plant and 2133 seeds/plant in 2018-19 and 2019-20, respectively; while plants competing with barley produced 741 seeds/plant and 38 seeds/plant in 2018-19 and 2019-20, respectively. Although the results with barley were potentially better in season 2019-20, the shorter life cycle of camelina permitted its harvest three weeks before than in barley. Consequently, more than half of the reproductive organs in plants competing with camelina were still at flowering stage, and those that already presented capsules were still immature. This means that the seed rain of *P. rhoeas* competing with camelina was completely avoided, while the few seeds produced by plants competing with barley were mature at harvest time. These aspects, reduction of the weed size and seed rain avoidance, make of this crop very interesting for its use in rotation with winter cereals, and a higher capacity to reduce the weed population because of sowing date flexibility, and generally earlier harvest.

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# ANALYSIS OF MOLECULAR MECHANISMS ASSOCIATED WITH LOW TEMPERATURE INDUCED FREEZING TOLERANCE AND FLORAL COMPETENCE IN *CAMELINA SATIVA*

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*Camelina* (*Camelina sativa* L., Crantz) is an oilseed crop consisting of spring- and winter-biotypes. Cold-acclimation of winter-biotypes allows for greater freezing tolerance compared with spring-biotypes. Winter-biotypes also require a low temperature treatment (vernalization) to induce flowering processes, while spring-biotypes do not. In this study, a winter-biotype of camelina (Joelle) exposed to a low temperature (5°C) treatment for 0, 1, 2, 4, or 6 weeks and subsequently exposed to a freezing treatment (-15°C for 4 h) demonstrated 0, 0, 20, 36, and 72% freezing survival, respectively. Plants that survived the freezing treatment (42 d post under greenhouse conditions) also flowered; plants receiving six weeks cold treatment flowered first, followed in order by plants exposed to four- and two-weeks cold treatment. No spring-biotype (CO46) plants exposed to the low temperature treatment (up to 8 weeks) survived this freezing treatment. To determine molecular components associated with regulation of low temperature acquired freezing tolerance and floral competence in camelina, we compared transcriptomes of the spring- and winter-biotype exposed to 0, 1, 2, 4, 6 and 8 weeks low temperature (5°C). A program called SCION was used to run network analyses on expressed genes (FPKM >2 for all replicates of at least one time point), which identified 21 and 22 coordinately regulated gene clusters across all time points in the spring- and winter-biotype, respectively. The SCION program also identified transcription factors (TFs) likely regulating genes within each cluster. Among 71 strongly down-regulated genes unique to the winter-biotype (FDR <0.05 and fold change in expression >4 between biotypes at any time point), bZIP, GRAS, and WRKY TFs were identified as potential dominant regulators. Among 168 strongly up-regulated genes unique to the winter-biotype, ERF, MYB and bZIP TFs were identified as the most prevalent regulators. Gene set enrichment analysis of individual gene clusters identified ontologies associated with biotic stress and defense responses as over-represented among clusters of down-regulated genes (from time 0) in both biotypes, and ontologies associated with growth-related processes were uniquely down-regulated in the winter-biotype. Likewise, among clusters with up-regulated genes, ontologies associated with abiotic stress, light quality responses, and photosynthesis were observed in both biotypes, and ontologies associated with secondary metabolism and sugar transport were more prevalent in the winter-biotype. Functional confirmation of candidate genes and TFs regulating freezing tolerance and flowering time will provide new knowledge for improving these traits in oilseed crops with industrial and agricultural importance.

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GENETIC DIVERSITY AND POPULATION STRUCTURE OF *USDA*  
*COLLECTION BRASSICA JUNCEA POPULATION*

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Sustainable, alternative fuels represent a promising solution to help reduce carbon emissions, expand domestic energy sources, contribute to price, and supply stability, and stimulate economic development in rural communities. The success of biofuels relies on finding inexpensive feedstocks that do not compete with food crops and can be cultivated economically in diverse geographical regions and agricultural production systems. Mustard (*Brassica juncea*), a native crop of the western and central Asia regions, has recently become the object of increasing interest due to its high oil content with high saturated fatty acid compositions that can be refined into fuels that meet the specifications of petroleum-based fuels. *Brassica juncea*' agronomic advantages including low requirement for water and nutrients, and adaptability to adverse environmental conditions make it an ideal crop for sustainable agricultural systems and marginal lands. However, the lack of genetic and genomic resources and information has slowed the progress of developing this into a viable oilseed crop. To build a *Brassica juncea* genomic resources, a panel consists of 340 of USDA brassica juncea accessions was genotyped to explore their genetic diversity, relatedness, and population structure. A total of 99030 high-quality single nucleotide polymorphisms (SNPs) were identified using genotyping-by-sequencing (GBS) technology. Those SNP were distributed over the 18 chromosomes with an average polymorphism information content (PIC) values of 0.29 indicates moderate genetic diversity for *Brassica juncea* panel. Population structure and principle coordinates analyses (PCA) based on identified SNPs revealed five distinct subpopulations. The result shed the light on how plant breeding and selection may have affected the formation and differentiation within mustard natural populations and how their genetic diversity can be used in future breeding efforts. the genotyped panel coupled with high dense identified SNP markers is a great resource for allele/gene identification using genome-wide association analysis studies (GWAS) and marker-assisted selection (MAS) approaches. These information provide a tool to enhance genetic gain in *Brassica juncea* breeding programs for biofuel related traits.

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## OILSEED BREEDING PROGRAM AT NORTH DAKOTA STATE UNIVERSITY

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North Dakota State University (NDSU) has a moderate sized oilseed (canola (*Brassica napus* L.) and flax (*Linum usitatissimum* L.)) breeding program. North Dakota led the nation in the production of both canola and flax with over 85% and 92% of the U.S. production, respectively. North Dakota State University initiated the canola breeding program in 2006. Crossing and backcrossing have been made among genetically diverse spring x spring, and winter x spring types canola to develop new breeding lines with desirable traits. Interspecific crosses were also made among *B. napus*, *B. rapa*, *B. juncea* and *B. carinata* to increase the genetic diversity. A total of 366 wide diversified *B. napus* accessions were partially sequenced using genotyping by sequencing (GBS). Canola double haploid production and molecular marker technology are already in place. The breeding program obtained a licensing agreement with INRA, France to utilize Ogura-CMS and restorer (R-2000) system to develop inbred lines for hybrid production. Greenhouses and plant growth chambers are utilized to grow canola in a controlled environment. The seed quality lab is equipped with nuclear magnetic resonance for seed oil and seed protein analysis. Disease screening facilities are available both in greenhouse and in field conditions. The field-plot testing program is located at seven locations in North Dakota with about 5,000 research plots per year. The breeding program is fully equipped with planter, swather, combine, tractor, cultivator, truck, trailer etc. Contra-season nursery is located in Temuco, Chile that significantly reduced the breeding cycle. Although the breeding program is comparatively new, a high oil open-pollinated (OP) roundup ready cultivar (NDSU-662c) was released in 2012, and high seed yield OP conventional type cultivars NDOLA-01 and NDOLA-02 were released in 2017 and 2021, respectively. Furthermore, NDSU has a moderately sized flax breeding program that is mostly focused on the release of high yielding, high oil varieties for production in North Dakota and surrounding states. This is the only flax breeding project in the U.S. and is one of only two in North America. The program is giving emphasis to increase the genetic diversity of NDSU flax. For the first time a genome based genetic diversity study of U.S. flax germplasm accessions has been done at NDSU. Parents were selected from diverged groups for diallel cross to increase genetic diversity in the population. The field-plot testing program is located at seven locations in North Dakota with about 4,000 research plots per year. The breeding program is fully equipped with all necessary field equipment. The program has been releasing superior cultivars on a regular basis. Recently, we have identified two breeding lines, which were outperformed over all commercial flax cultivars in 2019 and 2020 at North Dakota Flax Variety Trial and in progress to release in 2023. The NDSU-Plot 30 was established as part of a flax *Fusarium* wilt study, and flax germplasm has been grown in this plot every year since 1894.

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## IMPROVING PENNYCRESS ESTABLISHMENT THROUGH EARLIER CORN HARVEST

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Pennycress (*Thlaspi arvense* L.) is a new oilseed being developed as feedstock for biofuels and bioproducts. As a cash cover crop, it can be grown between summer annual corn-soybean production to diversify US Corn Belt cropping systems and provide ecosystem services. However, because of corn's long growing season, establishing pennycress after grain-corn harvest is challenging, especially in the northern Corn Belt. A multi-institutional multiyear study was initiated in 2020 to evaluate using early corn relative maturity (CRM) hybrids to hasten harvest and allow earlier direct-drilling of pennycress while determining yield and establishment tradeoffs of the two crops. Study sites from south to north included Lexington, IL, Hoytville, OH, Rosemount, MN, and Morris, MN. We hypothesized that earlier than full-season CRMs may sacrifice some yield, but allow for improved establishment, and hence, yield of pennycress. Five different CRM hybrids were grown at each site. The CRMs used in IL and OH ranged from 95 to 113 days, with 113 days representing a common full-season hybrid for the region. The two MN sites used a CRM range of 76 to 95 days corn, with the 95 days representing full-season. Indeed, earlier corn harvest (i.e., early CRMs) generally resulted in greater pennycress plant densities in the fall, indicating improved establishment. Fall establishment varied considerably across locations. For the three earliest CRMs harvested, pennycress plant densities in OH, IL, Rosemount, MN and Morris, MN averaged 951, 49, 178, and 266 plants m<sup>-2</sup>, respectively, using the same seeding rate of 11 kg live seed ha<sup>-1</sup> direct drilled. Generally, spring stands were less than fall except at IL where they were greater and increased with later sowing, indicating spring rather than fall emergence. For CRMs eight to nine days earlier than full-season corn, there was generally no loss of grain yield. Use of early CRM corn to facilitate earlier harvest appears to improve pennycress establishment without greatly limiting corn yield. Pennycress yields will be reported but have not been processed at this time.

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## CASTOR MEAL FOR ANIMAL FEEDING AND CONTROLLING NEMATODES – STUDIES IN PROGRESS

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Castor (*Ricinus communis* L.) is an important oilseed crop providing raw material for the chemical industry. Castor meal is the by-product of castor oil extraction, which is produced in the proportion of 1.2 times the weight of castor oil. Castor meal has a high protein content, but it also contains a highly toxic protein (ricin) which hinders its use as feed. Castor meal has been traditionally used as organic fertilizer or burned for producing heat. The use of castor meal as feed is a long-term goal which could add value to the product, and benefit the whole castor supply chain. Some progress toward that objective has succeeded. It was observed that when the industrial processing of castor oil includes solvent extraction, the toxicity of the meal is substantially reduced. It seems that in the regular industrial process, the temperature applied for evaporation of the solvent (hexane) from the meal damages the ricin, and most of its toxicity is lost. The residual ricin in the meal is still toxic for monogastric (preliminary tests made in broilers), but ruminants seem to tolerate the remaining toxicity without detectable detrimental effect. It was also demonstrated that solvent-extracted castor meal can be easily sieved and separated into fractions with contrasting properties. After sieving, the fibrous components are retained in the larger particles, while the protein components get concentrated in the fine particles. The fractions with the finest particles can reach protein content higher than 70%, although this fraction correspond to only 3.5% of the integral castor meal weight. Castor meal provided by the industry, without any additional detoxification procedure, was added as protein source to the diet and used to feed 16 male lamb for two months. No symptoms of intoxication was observed in blood analysis, in behavior, or in the performance compared with lamb fed soybean meal. The study is being continued in larger scale and longer duration trials feeding ewes with industrially-detoxified castor meal. Castor meal was also found an interesting option as additive for sugarcane silage because it has high-protein content and high capacity for absorbing moisture from the silage. Castor meal improved the quality of fermentation and the nutritional composition when added to sugarcane silage, and another study is in progress to test the performance of small ruminants feeding this roughage. Castor meal is also being explored as a protein source for feeding small ruminants with properties to manage the adult stage of *Haemonchus contortus* (a very prolific bloodsucker worm that parasite the true stomach of ruminants). Additional studies are testing the use of castor meal as special organic fertilizer with properties for suppression of root-knot nematode (*Meloidogyne incognita*) and of the free-living stages of the gastrointestinal nematode (*H. contortus*) that occurs in the pastures. These studies are a collaboration with the main industry of castor oil extraction in Brazil (Azevedo Óleos Vegetais) with the aim of adding value and developing new uses for castor meal.

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## SAFFLOWER (*CARTHAMUS TINCTORIUS* L.) A MULTIPURPOSE OILSEED CROP FOR THE MEDITERRANEAN REGION

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Mediterranean farmers mostly rely on winter cereals, this making weed management highly challenging due to the selection of herbicide-resistant weeds and the increasing pressure on soil-borne diseases. Crop diversification represents a valid strategy for sustainable weed management and it is essential for an agroecological transition and for ensuring sustainable and resilient farming systems. The need for crop diversification has also to meet the market requests, with particular attention to the domestic shortage of vegetable oil and proteins. In this scenario, new perspectives are open for innovative winter oilseed crops, such as camelina (*Camelina sativa* L. Crantz), carinata (*Brassica carinata* L.), and more recently safflower (*Carthamus tinctorius* L.). The latter is of particular interest in view of its high suitability to Mediterranean climate, low input needs, high plant vigor, also in marginal soil conditions, and tolerance to low temperature. Thus, in such conditions, safflower could be grown with a winter cycle, differently than sunflower (*Helianthus annuus* L.), despite belonging to the same botanical family (i.e., *Asteraceae*). As for sunflower, also for safflower high oleic hybrids are now available on the market tremendously enlarging the applications of its oils, which could meet the needs of the domestic biobased industry. Aiming at evaluating the feasibility of high oleic safflower as a winter oilseed crop in the Mediterranean region, a multi-year and multi-location study has been carried out, across three growing seasons (2019-2021), in several locations in Emilia-Romagna and Tuscany regions (Italy), traditionally devoted to cereal cultivation. In each region, the locations were chosen as representative of optimal, mean and marginal conditions. The high oleic safflower variety “CW99OL”, supplied by MAS Seeds (Italy), has been grown at all locations. At each test site, safflower was sown in winter and spring (January-March) and harvested in summer (July-August). The trials were arranged as open field, to define safflower suitability to available farm equipment and practices. All trials were rainfed and carried out under low input agronomic management and using mechanical weed control. In Emilia-Romagna region, the safflower seed yield ranged between 3.4 Mg DM ha<sup>-1</sup>, in the optimal site, and 0.79 Mg DM ha<sup>-1</sup>, in the marginal one, while in Tuscany, it varied from 2.70 Mg DM ha<sup>-1</sup> to 1.25 Mg DM ha<sup>-1</sup>, in the optimal and marginal site, respectively. In Emilia-Romagna seed oil content was found not affected by the test location, and it was 35% DM, also fatty acid composition seemed only slightly influenced by growing conditions with oleic acid always exceeded 74% DM. Conversely, in Tuscany, a significant effect of test site on seed oil content was observed, ranging from 39 to 43% DM, as well as on fatty acid profile with oleic acid reaching > 74% DM (from 74.3 to 78.8% DM). High oleic safflower confirmed to be an interesting opportunity for Mediterranean farmers who are willing to differentiate their rotations while producing an oilseed crop with several biobased applications and able to increase local production of vegetable oil and protein. Even in marginal conditions, safflower demonstrated to be a versatile oilseed crop, able to provide satisfactory seed yield, and interesting advantages compared with sunflower, such as higher resistance to bird predation and diseases, greater weed competition and an early soil cover, when sown in winter reducing the risks of N-leaching and soil erosion.

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## AMELIORATIVE BIOSTIMULANT APPLICATION AT SUNFLOWER HYBRIDS TREATED WITH INAPPROPRIATE HERBICIDES

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Sunflower (*Helianthus annuus* L.) is among the most important crops grown in Bulgaria. For achieving high and stable yields it is necessary to perform effective weed control strategies. Herbicidal products should be applied in the optimal growth stages of the crop before the critical period. Otherwise that may lead to disturbing crop's physiological processes and growth parameters. In some cases sunflowers from different systems (IMI-SUN or ExpressSUN) are sprayed by mistake with the unappropriated herbicidal product for the growing system – IMI- or Express SUN and reverse. Then, acute or chronic phytotoxic effects (even death of the plants) may occur. In some situations, the plants mistakenly treated with the wrong herbicidal product may recover completely, or to different extends by ameliorative treatment with plant biostimulants. By this reason, two field trials in the growing seasons from 2017 to 2019 were conducted. The experiments were performed in the experimental field of the department of General agriculture and Herbology at the Agricultural University of Plovdiv, Bulgaria. The aims of the experiments were to evaluate the ameliorative effect the plant biostimulant Amino Expert® Impuls after imitation of "mistaken" herbicide application. In the first experiment the ExpressSUN sunflower hybrid "P 64 LE 25" was treated with imazamox, and in the second, the IMI-SUN Clearfield Plus sunflower hybrid "Bacardi" CLP was sprayed with tribenuron-methyl. The treatments included untreated control plots, application of imazamox (Pulsar Plus – 2.00 l ha<sup>-1</sup>) or tribenuron-methyl (Express 50 WG 0.04 kg ha<sup>-1</sup> + Trend 90 - 0.1 %) alone and variants with ameliorative supplication of plant biostimulant four days after the herbicidal intervention. Several physiological and growth parameters were evaluated: Chlorophylls (a and b), carotenoids as well as net photosynthetic rate seven days after herbicidal treatment; aboveground mass, number of leaves and total leaf area in the beginning of flowering stage; full flowering time; plant height at the end of the vegetation; sunflower head diameter, seed yield, hectoliter and absolute seed mass, as well as seed oil content. After the application of the plant biostimulant Amino Expert Impuls for ameliorative treatment the plant from both cropping systems overcome the herbicide stress to different extend. The Chlorophylls (a and b) and carotenoids as well as net photosynthetic rate were higher for the untreated plants from both technologies. The "mistaken" herbicidal application of Express 50 WG + Trend 90 showed higher inhibiting effect on the sunflower hybrid "Bacardi" CLP than Pulsar Plus to the hybrid "P 64 LE 25". The full flowering stage was recorded for the treatment of Express 50 WG + Trend 90 alone, while no significant differences for this indicator at the other hybrid were found. For the treatments where plant biostimulant application was used, increase of the parameters sunflower head diameter, plant height, seed yield, absolute and hectoliter seed mass, as well as seed oil content were recorded. The obtained results showed that the significant results are in favor of ExpressSun technology. In the event of errors in the application of herbicides in these two cropping systems, the consequences are much more corrective in cases where tribenuron-methyl-tolerant sunflower is treated with Pulsar Plus than if IMI-SUN sunflower is treated with the herbicide Express 50 WG + Trend 90.

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# **ABSTRACTS OILSEEDS DIVISION**

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## CRAMBE – WHICH VARIETY FITS BEST IN GRECE?

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Crambe (*Crambe abyssinica* L.) is a non-food oil crop offering feedstock for the European biobased industry. In Greece can be grown as catch spring crop with a short growing cycle (<120 days). The purpose of this research work was to compare in total 10 varieties/lines for a period of four subsequent growing periods (2015-19) in central Greece. The varieties/lines compared were: Galactica, Nebula, Elst-2007-2, Elst-2007-3, Elst-2007-7, Elst-2007-8, Elst-2007-9, Elst-2007-16, Pri-9104-71 and Pri 9104-100 in three blocks. The size of the experimental plots was 1.5x6 m<sup>2</sup> and the distances between the rows were 15 cm. In all trials, a number of measurements had been carried out (number of plants in one meter row in the beginning and at the end of the growing season, plant height, etc.). Each year the sowing took place from the middle to the end of March and the harvesting from late May till mid of June. At the end of each growing cycle a final harvest of 5 m<sup>2</sup> (5x1) per plot took place and the harvested biomass had been first weighted and then the seeds were separated and weighted. Samples from both plant fractions (seeds and straw) had been collected for laboratory analyses (oil content and profile for the seeds and biomass characterization for the straw). The most productive variety was Pri 9104-100 (2090 kg/ha) and the least one was Nebula (1384 kg/ha), averaged over the four trials. Quite high seed yields were recorded by Elst-2007-9 and Elst-2007-16 (>1900 kg/ha) and Galactica (>1800 kg/ha). The yields of the residual biomass was 3.58 t/ha (averaged overall varieties and years). The ash content of the residual biomass was around 10% with calorific value of 4142 kcal/kg.

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## COMPOSITION IN FATTY ACIDS AND TOTAL POLYPHENOLS IN DIFFERENT GENOTYPES OF *CANNABIS SATIVA L.*

Giulio

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Hemp (*Cannabis sativa L.*) is a multipurpose crop traditionally cultivated as fiber crop and since few years rediscovered as natural biofactory of high-value products and bioactive molecules. The seeds of *C. sativa*, named hempseeds, are the edible fruits and can be utilized for food, feed and cosmetics/health care. They can be consumed as such (whole) or dehulled, and the oil obtained by cold press can be used in food and cosmetics. Also defatted flour can be used as feed. Hempseeds have a peculiar composition, different from other parts of the plant, as they do not produce nor accumulate cannabinoids, the typical terpenophenolic molecules that gives peculiar properties to *Cannabis*. Fresh seeds content is typically 20 - 25% proteins, 25 – 35% oil and 20-30% carbohydrates, as well as vitamins and minerals. Hempseed oil has unique properties and features compared to other vegetable oils, with a unique and balanced fatty acids (FAs) composition. It is a rich source of Essential Fatty Acids (EFAs), which cannot be synthesized endogenously by humans and must be provided by diet. The most important and relevant FAs are linoleic acid and  $\gamma$ -linolenic acid, belonging to the omega 6 ( $\omega$ -6) class, and  $\alpha$ -linolenic acid and stearidonic acid, belonging to the omega 3 ( $\omega$ -3) class. The ratio between  $\omega$ -6 and  $\omega$ -3 in hempseed oil, variable according to the genotype, is usually close to the 3:1 considered as optimal for promoting an anti-inflammatory state and other beneficial effects on the consumer, according to the OMS recommendations. Also, polyphenols and tocopherols, known anti-oxidant compounds, are abundant. All these qualities make hempseed oil nutritionally interesting for human consumption. This study was conducted at Rovigo and Bologna experimental stations of CREA – Research Center for Cereal and Industrial Crops. A total of 28 genotypes, comprehensive of hemp registered varieties and experimental lines conserved within the *Cannabis* germplasm collection of CREA-CI, were cultivated in 2020 in Rovigo until seeds were harvested (from end of August to end of September, according to the different flowering time). Genotypes were analysed for the chemical profile of cannabinoids (chemotype) with GC-FID to verify the levels of main cannabinoids THCA (Tetrahydrocannabinolic acid) and CBDACannabidiolic acid) and assure the legacy of the material (<0.2% w/w for THCA). Then, seeds were analysed for oil composition, protein and polyphenols to assess their antioxidant potency. The FA profiles were different, depending on genotype, as already reported in literature. A prevalence of linoleic acid (from 51.1 to 56.4%) and  $\alpha$ -linoleic acid (from 11.05 to 20.2%) was found, and 17 out of the 28 genotypes showed an optimal ratio between  $\omega$ -6 and  $\omega$ -3, ranging between 2.9 and 3.7. The polyphenol content also showed a great variability, varying from a minimum of 2.03  $\mu$ g GA/mg up to 8.21  $\mu$ g GA/mg depending on the genotype. The average protein quantity was around 24%, from 10.87 to 26.9 %. Major variability was found in CREA experimental hemp accessions than in varieties. Considering the optimal  $\omega$ -6/ $\omega$ -3 ratio and the high total polyphenols content, we identified three accessions, namely CREA\_CI\_RO\_S460, S323 and S435 interesting as pre-breeding materials to select new varieties for hempseed production, suitable for Italian environment, with the aim to solve the lack of availability of new Italian varieties for seed / oil production.

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## CAMELINA A CASH COVER CROP FOR THE MEDITERRANEAN REGION: PRELIMINARY RESULTS FROM THE 4CE-MED PROJECT

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Mediterranean dry-farming systems mostly rely on cereal production (mainly wheat (*Triticum aestivum* L.) and barley (*Hordeum vulgare* L.)), often as sole crops, due to a lack of alternatives. In this scenario, winter camelina (*Camelina sativa* L. Crantz) has been recently identified as a suitable alternative as a cash cover crop to be grown in northern USA, in relation to its short cycle, and ability to grow also on marginal soil. Nevertheless, the inclusion of spring camelina, as a cash cover crop, within conventional Mediterranean farming systems, will be also possible matching the environmental benefits, related to its cover crop “attitude”, and additional revenue for farmers, who will get profit from selling camelina seeds for oil and protein. At this scope, in the framework of the 4CE-MED project, funded by the PRIMA program, among concurrent cover crops, spring camelina was chosen as the most suitable for the Mediterranean climate, being extremely drought tolerant, suitable to different sowing times, and able to improve soil and water conservation among smallholders. With the scope to identify the most suitable camelina variety for different Mediterranean countries, a screening trial was established in autumn 2020 in seven countries (i.e., Algeria, France, Greece, Italy, Morocco, Spain, and Tunisia), at one or two locations in each country. The different camelina varieties were all supplied by Camelina Company (Spain), and they were selected to better match the climatic peculiarities of each test site. At all locations six different camelina varieties were compared. Three varieties (CCE26, CCE29, and CCE42) were in common to all the locations, then in Algeria, Tunisia, Morocco and Spain the varieties CCE43, CCE44, and CCE117, characterized by prolonged vegetative cycle and late maturity, were also grown; while in Greece, Italy and Southern France the extra varieties were CCE27, CCE34, and CCE40, characterized by shorter vegetative cycle and early maturity, possibly enabling double cropping in these countries. Sowing times were adjusted to optimize camelina growth cycle, in relation to local conditions, and to promote its cover-crop attitude. Camelina sowing ranged between end of October (Italy) until end of December (Morocco), while harvesting started in early May in the African countries (Morocco and Tunisia) and it end at the end of June in some of the European ones (i.e., France). In order to collect a common set of data, during camelina development and at harvest, the 4CE-MED partners agreed to survey: emergence rate, days to flowering and maturity, height at maturity, plant density at harvest, seed yield, thousand seed weight, seed oil content, and fatty acid profile. Besides being the first time ever that camelina is grown across such a different number of Mediterranean pedo-climates, from the preliminary results it is possible to highlight how this oilseed crop adapted well to all conditions. Also, in the African countries, where 4CE-MED partners were completely new to camelina, the results are very promising and camelina confirmed its wide environmental suitability and capacity to adapt also to sub-Saharan climate. In the next months the surveyed parameters will be analyzed in order to define the differences between the considered varieties in response to different environments.

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## CAMELINA GERMINATION UNDER OSMOTIC STRESS - TREND LINES, TIME-COURSES AND CRITICAL POINTS

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Camelina [*Camelina sativa* (L.) Crantz] is native species of Eurasia, which is gaining interest worldwide due to its better cold, heat, and drought tolerance, and less susceptibility to disease and pests than oilseed rape (*Brassica napus* L.). Furthermore, studies conducted in Canada and USA reported that as far as yield is concerned, camelina could be competitive against other Brassicas. Water shortage during germination is one of the major constraints that induces irregular and delayed seed germination and emergence, leading to poor plant establishment. Camelina has relatively low water requirement and high tolerance to drought, at all stages of development, even at germination and early seedling growth. Aiming at understanding the response of camelina germination under osmotic stress and the identification of critical soil moisture levels for successful establishment, two spring cultivars, developed at the Institute of Field and Vegetable crops Novi Sad, were compared (NS Slatka and NS Zlatka) under 9 levels of osmotic stress, ranging from 0 MPa to -1.6 MPa. Seeds were kept at 20°C and 8/16 h light/dark cycle. Osmotic potential of solution was obtained by using polyethylene glycol. Seeds were considered germinated when radicle was at least 2-mm-long. Germination was surveyed daily, while final germination was determined when no germinated seeds were recorded for 3 consecutive d, or after 15 d of incubation. Results showed that that both cultivars did not decrease germination under mild and medium osmotic stress levels of (i.e., <-0.6 MPa). Higher levels of osmotic stress induced significant germination decrease in both cultivars, with NS Zlatka being the most sensitive one. Significant increase on germination speed was noticed at -0.4 MPa. A significant interaction G x OS interaction was surveyed with NS Zlatka having quicker germination in the control (0 MPa) and under the mildest level of osmotic stress (-0.4 Mpa). When osmotic stress was increased a significant bi-linear trend for both cultivars was surveyed. The inflection points were detected at -1.15 and -1.18 MPa, in NS Slatka and NS Zlatka, respectively, with trend in germination rapidly declining after this level. Furthermore, the estimated osmotic potentials for completing stopping germination were -1.45 and -1.46 Mpa, for NS Slatka and NS Zlatka, respectively. MPa. Time to 50% germination showed also a significant bi-linear trend in response to osmotic potential, but in the opposite direction than the one observed in germination. Inflection points were recorded at -0.77 MPa and -0.78 MPa for NS Slatka and NS Zlatka, respectively. After those points, time to 50% of germination increased rapidly. Number of d for initiating germination progressively increased with the decrease of osmotic potential. In control condition (0MPa) and under the mildest level of osmotic stress (-0.2 MPa), germination began the first day after incubation, with NS Zlatka having a more rapid start. Under more severe stress level (-0.8 MPa) germination began after 2 d, and only NS Slatka fulfilled its germination potential, but both cultivars reached 90% of germination. Under higher osmotic stress levels germination was postponed for a few days and maximum germination was obtained later. At -1.6 MPa camelina didn't germinate. Camelina confirmed to withstand high levels of drought stress at germination and could be considered a more suitable option than oilseed rape on marginal lands, or areas with irregular precipitation.

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## TOWARDS AUTOMATING EARLY VIGOR RATINGS FOR BRASSICA PLANTS

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Early vigor is an important trait of crops. Defined as an increased amount of foliage in the rosette stage, early vigor can lead to numerous benefits. High early vigor can result in better water uptake, making the plants more tolerant to water-limited conditions and more foliage can lead to a higher production of photosynthates, resulting in higher biomass production and seed yields. Increased early season growth further strengthens a plant's ability to compete with weeds, an important trait particularly for crops with limited in-crop weed control options. Currently, rating early vigor relies on tedious, visual, and, most problematic, subjective methods that require a person to observe each individual plot in a field trial. In this study, we aim to develop an automated field imaging workflow for estimating early vigor in Brassica plants with the purpose of increasing speed and objectivity of vigor ratings. Two trials were used as test cases, a diverse *Brassica carinata* nested association mapping (NAM) population and a winter x spring *Camelina sativa* breeding nursery; both set up as modified augmented designs (MAD2). The *Brassica carinata* trial consisted of 150 ranges, each containing 23 double row plots. The *Camelina sativa* trial had 37 ranges, each containing 29 double-row plots. Overhead images of the trials were acquired by GoPro cameras, mounted on a tractor, pointed down, moving perpendicular to plot rows. These images were then processed to obtain binary images of the individual rows of each plot. To do so for the *C. sativa* trial, one image was chosen for each plot with the criteria being that the plot was roughly centered in the image. The plant pixels in each image were segmented from the background using an excess green algorithm. The image was rotated so that the rows would be horizontal, which was done by finding the centreline of each row in the plot using a clustering algorithm with linear clusters. In order to get a consistent distance scale for each image, the average distance between the rotated centrelines was computed and each row was re-cropped to be the rotated centreline  $\pm 0.5$  the previously calculated distance. Using the generated binary images, two metrics were calculated for estimating early vigor: plant area and distance from centreline. Plant area was computed as the number of white pixels in a binary image. Distance from centreline was computed as the Euclidean distance from the centreline of the row to the edge of the plant. We compared these image-based metrics to visual early vigor ratings obtained by two trained technicians for *B. carinata*. The correlation coefficients were found to be 0.560 and 0.754 for plant area and 0.572 and 0.736 for distance from centreline, for each technician respectively. The moderately strong correlation for image-based vigor metrics is promising. Visual early vigor ratings have not yet been obtained for *C. sativa*. Qualitative inspection of image-based vigor metrics shows that plots with low vigor metric exhibited sparser rows than those with a higher vigor metric. Our preliminary results suggest that automated vigor ratings may aid breeders in faster and less subjective identification of which plots to ignore and which to advance.

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# IDENTIFICATION OF A FAD2 POINT MUTATION PUTATIVELY RESPONSIBLE FOR HIGH OLEIC SEED OIL PHENOTYPE IN AN EMS-MUTAGENIZED MILK THISTLE POPULATION

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For several decades, modification of oil seed yield and composition have led researchers working toward genetic improvement of “major” crops. As vegetable oils demand is still increasing, it is worth to also consider so far neglected and underutilized species. Indeed, alternative crops have been considered as convenient oil feedstock, for their nutritional properties and adaptability to marginal environments. *Silybum marianum* (L.) Gaertn., also known as milk thistle, is an herbaceous member of the *Compositae* family. The species is mostly cultivated for the production of silymarin, a high-value complex of flavonolignans with health beneficial properties mainly used for the treatment of liver diseases. Its oil productivity and composition lead to consider the species interesting also for the food and nutrition field, as well as in industry applications. Increasing the fruit quality traits is one of the main targets for breeding in *S. marianum* and high oleic acid content is an important characteristic of fruits for food and industrial applications. For these reasons, an Ethyl Methanesulfonate (EMS)-induced mutagenesis programme was implemented and the screening of a mutagenized population by GC-FID analysis led to the identification of a High Oleic (HO) mutant line with a high oleic oil content. *Fatty Acid Desaturase 2* (*FAD2*) genes are responsible for the determination of oleic acid levels in vegetal oil. Therefore, our research focused on this gene family as a possible site of the obtained mutation. We report for the first time the identification, cloning and transcriptional analyses of twenty-one complete and distinct *FAD2* orthologous genes in two *Silybum marianum* genotypes, to our knowledge the second largest *FAD2* gene family examined to date since a twenty-six-member family has been described for *Artemisia sphaerocephala*. The sequencing and comparison of all the *SmFAD2* genes highlighted a Single Nucleotide Polymorphism (SNP) in a seed-specific gene, resulting in a nonsynonymous substitution in high oleic genotype. The same SNP was identified in the F2 progeny derived from HO X WT backcrosses, segregating with the HO phenotype, thus hypothesizing the direct implication of this gene in the synthesis of linoleic acid. The characterization of this large *FAD2* gene family in *S. marianum* and the newly identified mutation highlight interesting novelties and will provide useful information for directing further breeding programmes towards new varieties for the production of high oleic oils, with improved oxidative stability, suitable for biofuels production or industrial applications.

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FATTY ACID COMPOSITION OF THE OIL FROM EXPRESS-SUN® SUNFLOWER HYBRIDS,  
DEPENDING ON SOIL FERTILITY.

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The oil content of the seeds is the main quality indicator of sunflower (*Helianthus annuus* L.). Increasing consumption of vegetable fats is the reason for the displacement of animal fats, as vegetable oils have an antioxidant role for the human body. In addition to the oil content in the seeds, the main quality indicator of sunflower is the composition of the oil. One of the main valuable qualities of sunflower oil is the higher amount of unsaturated fatty acids compared to saturated. Apart from the ratio of saturated: unsaturated fatty acids, another main quality indicator of sunflower oil is the ratio of the two main unsaturated fatty acids (linoleic and oleic), which determines the respective batch of oil as linoleic or oleic type. According to the standards adopted for oil, the standard sunflower oil is linoleic type. High oleic is considered to be one in which the oleic acid content is over 75% (Codex Stan.) and according to the latest standards of the National Sunflower Association the oil containing more than 82% oleic acid is defined as high oleic. A three-year (2018, 2019 and 202) split-plot trial was set up at the experimental field of Agricultural University of Plovdiv (Bulgaria, 42°13' N, 24°80' E), comparing five ExpressSun® (tribenuron-methyl resistant) sunflower hybrids - P64LE25 – Pioneer; LG 59.580 SX – Limagrain; Subaro HTS – Syngenta; ES Arcadia - Euralis and Magma SU (CSF 17902 SU) - Caussade semences (subplots) in conditions of two different soil nutrition levels (main plot). The fatty acid composition of the oil is determined by the gas chromatography (ISO 12966-2:2017) method. Sunflower hybrids contain an average of 15% saturated and 85% unsaturated fatty acids. Soil fertility has an ambiguous effect on the ratio of fatty acids in the seeds. The highest content of linoleic acid is the LG 59.580, while the highest oleic is Magma. In drought conditions during seed formation, the content of oleic and palmitic acid increases, and the content of stearic and especially linoleic acids decreases.

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## OIL CONTENT AND YIELD OF TRIBENURON-METHYL RESISTANT SUNFLOWER HYBRIDS IN CONDITIONS OF DIFFERENT SOIL NUTRITION

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Sunflower (*Helianthus annuus* L.) is the most widespread and important oil crop in Bulgaria, as well as in other countries in Southern Europe. This is because sunflower oil is the traditional vegetable fat that has been consumed in these countries in recent years. Sunflower production is constantly growing due to improved cultivation technology, the introduction of hybrids resistant to diseases, pests and broomrape, as well as the introduction of hybrids resistant to herbicides. A three-year (2018, 2019 and 202) split-plot trial was set up at the experimental field of Agricultural University of Plovdiv (Bulgaria, 42°13' N, 24°80' E), comparing five tribenuron-methyl (ExpressSun®) resistant sunflower hybrids - P64LE25 – Pioneer; LG 59.580 SX – Limagrain; Subaru HTS – Syngenta; ES Arcadia - Euralis and Magma SU (CSF 17902 SU) - Caussade semences (subplots) in conditions of two different soil nutrition levels (main plot). The study aimed to determine the oil content of the seeds and the oil yield of the five sunflower hybrids under the conditions of two different soil nutrition. Determination of oil contents in the sunflower seeds was done by ISO 659 method. The oil yield was calculated depending on the oil content and seed yield per plot. Analysis of variance was performed to establish the independent action of the factors using a two-way (ANOVA) statistical package. It has been found that the increased content of macronutrients in the soil leads to a decrease in the fat content in the seeds, but due to the strong influence of higher soil fertility on seed yield, it leads to higher oil yield per unit area. The results obtained on the influence of the "soil nutrition" factor on the oil content of sunflower seeds are statistically significant. Although the effect of the "hybrid" factor on the oil content of the seeds was not significant in the analysis of variance, the sunflower hybrids can be ranked in the following descending order of average seed fat content: P64LE25> Subaru HTS> LG 59.580 SX> Magma SU> ES Arcadia.

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## MUTAGENESIS AS TOOL FOR ENHANCEMENT OF FATTY ACID COMPOSITION OF RAPESEED (*BRASSICA NAPUS* L.)

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The rapeseed is the second largest oilseed crop in Bulgaria. Its biological and economical value is due to high content of proteins and essential fatty acids. Seeds from various genotypes can have above 40 – 45% oil, used in food industry, as biodiesel and methyl esters industrial oils, in detergents, soaps and rapidly degradable plastics. A common tendency of lower content of saturated and higher percentage of unsaturated fatty acids in the oil is observed in the present varieties. This contributes to increasing the oxidation stability, both in the process of feeding and in intensive heating. Differences between seed oil quality are primary in the unsaturated fatty acid content. Improvement of the content and quantity of C18 unsaturated fatty acids is one of the main tasks in the rapeseed breeding. AU Plovdiv and National Research Program Foods financed the field trials and laboratory experiments during 2012 - 2020. Irradiation with gamma rays 100 Gy and 150 Gy of seeds from variety Trabant and Abacus provokes genetic and biochemical changes in the mutant generations of rapeseed. Characterization of the induced genetic diversity by ISSR and SSR markers is reported in our previous publication. Quantity and quality of the fatty acids have been screened by gas chromatograph and NIRS (Near Infrared reflectance Spectroscopy). The biochemical analyses of the mutants show lower content of monounsaturated fatty acids (MUFA) and higher content of the polyunsaturated fatty acids (PUFA) as linoleic (C18:2) and linoleic (C18:3) acids, which makes them appropriate for technical uses. A tendency for increase of the total content of crude fats is observed, related to lower total content of unsaturated fatty acids in the mutant seed oil. The changes in the fatty acid composition interferes with shorter plant height and better branching of the plant. Meantime the irradiation with 100 Gy induces larger and heavier seeds of the mutants and thus increases the production potential of the plant. The mutants could be used for further plant breeding procedures for enhancement the productivity and quality of the rapeseed oil as valuable source for food, feed and industry purposes.

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## DIVERSITY OF PRODUCTION POTENTIAL AND QUALITY OF CAMELINA (CAMELINA SATIVA) FOR HEALTHY FOODS AND BIO-ECONOMY.

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One of the underestimated oil crops in Bulgaria gains interest recently by its modest requirements to the environmental factors and multipurpose uses of its products. Camelina (*Camelina sativa* L. Crantz) can be considered as healthy food based on the high content of essential polyunsaturated fatty acids in the seed oil, good forage crop, primary source for the polymer or cosmetic industry, or even for production of biofuels for enhancement of the bio-economy. Despite advanced researches in many countries this crop remains outsider for the local farmers. Introduction of more than 150 accessions with different origin and level of plant breeding improvement, collected from gene banks from all over the world have been made in 2019. Field trials and laboratory experiments were done during 2019 – 2021. Regeneration in field trials in AU – Plovdiv and IPGR Sadovo in 3 successive years – from 2019 to 2021, revealed great variation in the productive potential and vegetation period in our climate conditions. Diversification of the morphological characteristics and elements of the productivity, oil content of seeds, fatty acids composition and oil quality presented by amounts of conjugated dienes and trienes were determined for the most perspective genotypes in our collection in the last year 2020. Germplasm polymorphism on morphological level shewed variation in plant height, branching, number and size of the pods and grain yield of one plant. The low requirements of the crop to the inputs suppose its cultivation on poor, sandy and unfavorable fields with low nutrition value, where other oil crops will hardly grow. Unfortunately, no one of the investigated accessions, sown in the spring, with insufficient rainfall for the vegetation, has significant yield potential (up to 300 kg/ha) without adequate mineral fertilizing and weed control, which impose the necessity of optimization of the crop production. These measures were taken during the last vegetation period and results were shown as references to the control with zero inputs. The oil content of the seeds varied in some degree (27.5 to 34.9%). The fatty acids composition of all genotypes manifested good quality for food purposes, especially for production of functional foods with high content of essential omega-3 and omega-6 fatty acids. Larger variation in fatty acids proportions was found for the main components alpha-linolenic acid 9, 12, 15-18:3 (26 to 34%), linoleic acid 9, 12-18:2 (18 to 26%) and oleic acid 9-18:1 (15 to 22%). The results reveal necessity of improvement of the crop production methods and introduction of new varieties of *Camelina sativa* for field production in order to provoke larger interest of the local farmers and producers.

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## CAMELINA ON MARGINAL LAND: A CULTIVATION TRIAL ON STEEP SLOPE IN ITALY

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The European Union is stimulating the replacement of fossil-based products with bio-based surrogates. Oilseed crops play a crucial role, thanks to their suitability to synthesize molecular structures which could substitute petrol-derived compounds. Worldwide production of vegetable oil is given for 75 % by few crops, such as soybean, oil palm, cottonseed, rapeseed and sunflower; while the remaining 25 % is obtained from other minor oilseeds. Some minor oilseed crops have particular features, which make them particularly suitable to the concept of bioeconomy. Camelina (*Camelina sativa* (L.) Crantz) is a very promising oilseed crop for multiple reasons. Beyond the oil composition, camelina is interesting because its crop management is performed with common machineries and the crop cycle is compatible with typical crop rotations of Mediterranean areas. A further advantage could be the possibility of cultivating camelina on marginal land, such that with slope higher than 12% with the aim of reducing iLUC effects. Currently the majority of these fields are in the process of abandonment since they are poorly productive for typical food crops, or left as grasslands. The present study offers new insights on the possibility of cultivating camelina on steep slope fields, and it represents one of the first documented trial of growing camelina in such marginal conditions.

Field survey was carried out to evaluate seed yield of the spring camelina cultivar Alba, supplied by Camelina Company (Spain), and to assess seed loss, when harvesting the crop with a combine harvester. A large plot trial (1000 m<sup>2</sup>) was established in January 2021 at the experimental farm of University of Bologna in Ozzano (Bologna). The experimental field is characterized by a slope of about 15%. The agronomic management was defined as low input organic. Harvesting was performed with a New Holland TC 5080 self-levelling combine harvester with 6 m working width and conventional cleaning shoe. Seed yield of camelina cultivar Alba reached on average 1.52 Mg DM ha<sup>-1</sup>. Results of the work performance analysis showed that seed loss was substantially lower than what found in current literature for flat land, i.e. 0.53 %, mostly as a consequence of the lower working speed. Therefore, it is possible to harvest camelina via self-levelling combine harvester also in steep slope conditions, confirming the suitability of such crop to grow on marginal lands.

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## WHO'S BEST? A FIELD TRIAL WITH 10 CAMELINA VARIETIES TO STUDY THEIR ADAPTATION TO SEMIARID MEDITERRANEAN CONDITIONS

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Crop diversification is a key element in the integrated management of crops because it provides soil and crop management variations, and contributes to maintain pest populations (weeds, pest and pathogens) under threshold levels. Unfortunately, crop diversification in semiarid rainfed Mediterranean Ebro Valley areas is limited by the water availability. For this reason, drought tolerant crops, also resistant to winter cold temperatures, like camelina (*Camelina sativa* L. Crantz) are valuable options to be considered for using in rotation with winter cereals. Camelina seeds contain high oil and protein levels, with unique fatty acid composition and important quantities of antioxidants. Its main use has been for biofuel production, but the oil has been demonstrated to be healthy for human consumption, and the meal has been successfully included for animal feed. But the limited camelina average yield of 1500 kg/ha, prevents farmers from sowing it more frequently. A solution for its wider implementation would be the increase of the yield price, for which more oil and meal could be destined for other purposes than biofuel, and among these animal feeding rises as the most promising option. On the other hand, despite its interesting characteristics, this crop still is understudied in the semi-arid climatic conditions of the Ebro basin (North-East of Spain) that represents typical Mediterranean conditions. With the aim to search the most appropriate variety that could adapt to these semi-arid conditions, an experiment with 10 varieties was established at the University of Lleida. Sowing was performed on 19 November 2020, in 1.5 x 10 m<sup>2</sup> plots, with three replicates, adjusting the sowing rate for each variety to achieve a density of 500 plants m<sup>-2</sup>. In February 2021 cletodim was applied to control grass weeds. Just before harvest, the biomass of camelina was taken to measure the total, vegetative and grain weights and to calculate the harvest index for each variety. For yield quantification, harvest at all plot was done with a Wittenberg harvester on 17 May 2021, except for the winter variety *Joelle*, which lasted one more week. Preliminary results show that the highest yields were obtained by varieties *Swiss landrace* (2079 kg ha<sup>-1</sup>), *Calena* (1935 kg ha<sup>-1</sup>) and *CO46* (1914 kg ha<sup>-1</sup>), while lowest yields were obtained by *GP204* (1441 kg ha<sup>-1</sup>), *Vera* (1555 kg ha<sup>-1</sup>) and *Omega* (1669 kg ha<sup>-1</sup>), being only significant differences between the *Swiss landrace* and *GP204*. Despite these differences further analysis must be done to consider the best choices of varieties and which are the best physiological models of camelina to be grown in the semiarid climate of Lleida. This study will be completed with oncoming oil and meal analysis.

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## CAMELINA (*CAMELINA SATIVA* L. CRANTZ) A NEW OILSEED CROP FOR MEDITERRANEAN AND BALKAN EUROPEAN CLIMATES

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Nowadays in Europe the new multipurpose oilseed crop, camelina (*Camelina sativa* L. Crantz), is not yet widely cultivated but in the last decade it has gained interest among farmers and other stakeholders in the value chain, in relation to its satisfactory yield, low input requirement, and suitability to different pedo-climates. If until now camelina has been grown as a spring crop in northern Europe, more recently southern environments in the Mediterranean basin and in the Balkan region have been targeted as suitable growing areas, either in autumn, winter or spring sowing. Nevertheless, the limited number of winter genotypes available, together with the typical winter season in those areas which is not very harsh, make the possibility to grow spring camelina genotypes with autumn cycle a feasible option. Aiming at defining the most suitable genotype and the optimal sowing date in the Mediterranean and Balkan regions a common trial has been established in autumn 2020 comparing four camelina genotypes (3 spring + 1 winter) and two sowing dates (early vs. late) across three locations in Italy (Bologna, 44° 30' N, 11° 23' E), Serbia (Novi Sad, 45° 15' N, 19° 51' E), and Spain (Lleida, 52° 10' N, 4° 29' E). The tested camelina varieties were as follow, for the spring genotypes: Alba (supplied by Camelina Company Spain), Calena (supplied by Saatbau, Austria), Sonny (supplied by KWS, Germany), for the winter genotype: Joelle (supplied by USDA-ARS, USA). All the three locations are characterized by clay loam soils. While in Bologna (Italy) and Leida (Spain) the two sowing dates were established, in Novi Sad (Serbia) due to adverse environmental conditions sowing took place only in late winter 2021. At all the locations similar seeding rate, plot size, and experimental design have been adopted. Additionally, a common set of parameters was agreed to be surveyed for easy data comparison, including: emergence rate, soil coverage (by means of the CANOPEO app), winter survival (only for the early sowing), phenology, final plant height, final plant density, seed and straw yield, 1000-seed weight, seed oil content, fatty acid profile. From the preliminary results it was possible to highlight a significant ( $P \leq 0.05$ ) location effect for emergence rate, with Italy reporting the lowest was the lowest value (29%) and Spain the highest (68%). Interestingly the spring variety Calena showed the highest emergence rate in Italy in autumn, while in spring it was the lowest among tested genotypes. Analyzing the data for soil coverage in Italy and Spain, the winter variety Joelle had the highest value in Spain, while in Italy it reported the lowest one. From the analysis of the complete set of surveyed parameters during camelina cycle across the three different environments will permit a through evaluation of its suitability to different pedo-climates in the Mediterranean and Balkan regions of Europe.

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## PHYTOREMEDIATION POTENTIAL OF DIFFERENT OIL CROPS IN HEAVY METALS CONTAMINATED SOILS

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Cultivation of industrial crops in heavy metals contaminated soils is an option once it contributes to reduce land use competition with food crops and the development of a vegetative cover contributes to reduce soil loss and degradation by erosion processes and water surface runoff. Therefore, the aim of this work was to study the effects of different heavy metals (Ni, Pb, Zn and Cd) on growth and yield of different oil crops, namely *Camelina sativa* (L.) Crantz, *Brassica carinata* A. Braun, *Thlaspi arvense* L. and *Crambe abyssinica* Hochst. The soils were artificially contaminated and the concentrations chosen were equivalent to the limit established by the Decree Law 276 of 2009 (Portuguese regulation that establishes the regime for the use of sewage sludge in agricultural soils) - Zn: 450 mg/kg; Pb: 450 mg/kg; Cd: 4 mg/kg and Ni: 110 mg/kg. Results from 3 year growing seasons indicate that all the tested oil crops can be considered tolerant to the heavy metals in this study (tolerance index, yields in contaminated soils/yields in control soils, higher than 0.50) except *T. arvense* that showed a tolerance index lower than 0.5. Among the different heavy metals, all the oil crops showed higher tolerance to Pb, followed by Cd and Zn, and less to Ni contamination. Higher phytoextraction potential was observed for Zn, Cd and Ni and less for Pb, with *B. carinata* showing higher accumulation potential and *T. arvense* the lowest potential. Although contamination reduced significantly the siliquae production, which may hinder the economical viability of the oil crops, the content in heavy metals in the siliquae fraction was low, indicating that the oil characteristics may still be valuable for the oleochemical industry.

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## COMPARATIVE PROCESSING OF WILD-TYPE PENNYCRESS AND LIGHT-COLORED COVERCRESS™ SEEDS

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Pennycress (*Thlaspi arvense*) is a winter annual oilseed crop that can be grown between corn and soybean growing seasons in most of the Midwestern US. The oil can be used as feedstock for conversion into biofuels and the meal could be used as feed. Protein concentrates and isolates obtained from wild-type pennycress (WT-PC) seed meal have good functional properties that are useful in food applications. CoverCress (CC) Y1126, a yellow-seeded pennycress, has been the focus of seed grow-outs recently in preparation for scaling up production. Laboratory-scale oil and protein processing were conducted to determine if the qualities of the products obtained from CC-Y1126 differ from those of WT-PC. Oil was extracted first by prepressing and then with hexane. Protein was extracted from the defatted meal using 0.1 M NaCl at 1:10 w/v meal-to-liquid ratio. CC-Y1126 seeds were smaller (0.89 g/1,000 seeds) than WT-PC seeds (1.07 g/1,000 seeds). The WT-PC and CC-Y1126 seeds have 33.1.3% and 36.1% oil contents (dry basis, db), respectively. Screw pressing resulted in similar residual oil contents in the cake (15.2 and 15.8 %, db) but the oil yield was higher for CC-Y1126 (24.1% versus 21.1%) because of its higher initial oil content. Hexane extraction produced defatted meals with <1% oil content. The color of CC-Y1126 press oil was lighter than that of WT-PC oil (8.8 versus 10.0 Gardner) but the hexane-extracted oils were about the same color (12.4 versus 12.7 Gardner). Both oils have similar fatty acid composition, with erucic and linoleic acids as major components at around 30% and 22%, respectively. CC-Y1126 seeds had 33% less crude fiber than WT-PC (12.9% versus 19.3%). CC-Y1126 defatted meal had slightly higher crude protein content (37.6%) than WT-PC (35.2%). Protein yield from CC-Y1126 (49.0%) was much higher than from WT-PC (36.3%) despite the small difference in crude protein content in the defatted meal. The protein extract obtained had 71-75% crude protein (qualifies as concentrate). The functional properties (solubility, foaming, and emulsifying properties) of CC-Y1126 protein concentrate matched those of the WT-PC, indicating similar potential uses in dairy substitutes, whipped toppings, dips, beverages and meat substitutes. The difference in the seed physical characteristics between WT-PC and CC-Y1126 had no effect in oil extractability, but the lighter color of oil from CC-Y1126 will reduce the cost associated with bleaching in oil refining, if required.

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## DOES SOWING PATTERN AFFECT THE COMPETITIVE ABILITY OF CAMELINA [*Camelina sativa* (L.) CRANTZ] AGAINST WEEDS?

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Crops are usually sown in rows. It is the easiest way of sowing, for which the machinery is adapted to, and enables easier way for weeding, either by post-emergence herbicides or by hand, or desiccation of the crop so that they are less affected by fungi. On the other hand, reduction of crop row distance has been demonstrated to be useful to enhance their competitiveness. But spatial uniformity is the most effective way of enhancing the competitiveness of a crop is maximizing the distance between crop plants (reduction of the intra-specific competition), maintaining the density, and reducing the distance with respect to the weed plants (increase of the inter-specific competition). This is impossible to achieve in large scale, and the best way to approximate to this distribution is the random one. Due to its characteristics, like the development of a rosette and high growth rate, camelina could benefit from this distribution. To test this approach, a preliminary experiment was set in a commercial organic field, where camelina was sown in December 2019 in rows with a 3-m wide no till disc drill, and randomly by releasing the tubes from the hopper and leaving the seeds fall directly into the ground. Results show that, in rows, the distance between plants within the row was of 4.1 cm on average, while between rows was maintained at 15 cm; on the contrary, mean distance between randomly sown plants was reduced to 3.6 cm. This supposes an increase of the crop coverage from 15.7% in average up to 23.2%, and a reduction of the crop-weed distance from 5.7 cm to 4.4 cm, as it is reflexed in Figure 1. Besides the competition, the increase of the crop coverage can also reduce the weed seed germination, contributing to a better weed control. These preliminary results must be confirmed in future research.

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UNCOVER MORPHO-PHYSIOLOGICAL DIVERSITY IN CAMELINA (*CAMELINA SATIVA* L. CRANTZ) UNDER DIFFERENT ENVIRONMENTAL CONDITIONS IN EUROPE

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Camelina (*Camelina sativa* L. Crantz) is an underexplored oilseed crop with great potential to become a staple crop in the near future, being able to meet the needs of different end-users in food, feed and non-food applications. Camelina has been identified as suitable for being grown under different soil and climate conditions, even in marginal land. Furthermore, it is characterized by intrinsic resistance against abiotic and biotic stresses when compared with other widely cultivated non-food oilseed crops, such as oilseed rape (*Brassica napus* L. var. *Oleifera*). One of the principal limitations for camelina development is the scarcity of genetic material, characterized by different morpho-physiological traits. To address this challenge, the UNTWIST project (Uncover and promote tolerance to temperature and water stress in *Camelina sativa*), funded within the H2020 framework research program, has identified a set of 54 camelina genotypes (i.e. UNTWIST core collection) selected for their genetic diversity and response to different climatic conditions. The UNTWIST core collection includes European commercial cultivars, that are currently grown in Europe or have been tested in previous field trials by UNTWIST partners (e.g. *Céline* and *Calena*), as well as diverse landraces from contrasting geographic and climatic regions. The landraces have been obtained from IPK/Germany, PGRC/Canada and external collaborators (i.e. AAFC/Canada, ALARC/USA, IFVNS Novi Sad/Serbia, Poznan University/Poland, University of Minnesota/USA, and University of Natural Resources and Life Sciences/Austria). Furthermore, Camelina Company has provided improved lines selected from their own breeding program in Spain. Genetic diversity, growth cycle length, performance under different climatic conditions, yield and lipid profiles (impacting temperature and drought tolerance) are distinguishing features of the core collection. Four similar field trials have been established across Europe during 2020/21 growing season. The test locations were: Bologna (Italy), Rothamsted (UK), Versailles (France), and Alcalá de Henares (Spain). At each test location similar plot size ( $\approx 1\text{-}2\text{ m}^2$ ) and seeding rate ( $500\text{ seeds m}^{-2}$ ) have been adopted. Sowing time was adjusted to that typical for camelina in that region, corresponding to the end of October in Italy, mid-November in Spain, end of March in France, and beginning of April in the UK. The local agronomic management practices, fertilization and weed control, were defined by region and recognized as optimal for each environment. Surveyed parameters during crop development and at harvest have been agreed among UNTWIST partners to allow for easy comparison across locations (i.e., rate of emergence, days to flowering and maturity, height at maturity, SPAD, plant density at harvest, seed yield, thousand seed weight, seed oil and protein content, fatty acid profile). From the preliminary results surveyed in Italy and Spain, where camelina was sown in autumn, the core collection demonstrated significant levels of diversity for traits such as days to flower, which differed by 25 days between locations, from the earliest and the latest genotypes. Also plant height greatly varied in Italy with about 45% height difference between the tallest and the shortest genotype. The collection and analysis of all data from the four test locations will permit the selection of four camelina genotypes, defined as UNTWIST focus lines, that will be thoroughly studied both in open field and in controlled environments to determine the mechanisms of plant plasticity.

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## PHENOLOGY OF *BRASSICA NAPUS L.* FROM REMOTE SENSING DATA

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Accurate phenological information is essential for monitoring crop development and predicting crop yield. Multispectral remote sensing is one of the advanced and effective techniques to discriminate crops phenology, plant health and different plant stresses. This study aims to exploit the possibilities of multispectral imaging for observation of phenological changes of oilseed rape (*Brassica napus L.*, hybrid - ES “Decibel” CL), treated with different herbicides applied for imitation of herbicide drift and untreated controls. The normalized difference vegetation index (NDVI) has been widely used for remote measurement of vegetation for many years. This index uses the reflections from the red region and the near infrared regions of the electromagnetic spectrum. Higher NDVI values reflect greater vigour and photosynthetic capacity of vegetation leaves, whereas lower NDVI values for the same time period are reflective of vegetative stress. NDVI, which is the normalized difference between the near infrared (NIR) and visible red reflectance, is responsive to changes in both the chlorophyll content and the intracellular spaces in spongy mesophyll of plant leaves. This study employed the NDVI to discriminate between two different phenological growth stages of *Brassica napus L.* in several variants (treated with different herbicides and untreated). The field plot trial was carried out at the experimental station of the Department of “General Agriculture and Herbology” at the Agricultural University of Plovdiv, Bulgaria. The investigated phenological growth stages were BBCH 51-54 (budding stage) and BBCH 69-71 (flowering, beginning of fruit development). The results showed a proven difference in the NDVI between the variants in the field trial in the two different phenology growth stages. Thus the multispectral remote sensing proves to be a sensitive and useful tool for discriminating of oilseed rape’s phenological phases. It can also be employed to evaluate the selectivity of different herbicides on crops and their effect on the plant health.

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