CSEI Catania
Centro Studi di Economia applicata all'Ingegneria

Con il contributo di:

Regione Siciliana
Assessorato dei Beni Culturali e dell'Identità Siciliana
Dipartimento dei Beni Culturali e dell'Identità Siciliana

HIGHER EDUCATION FOR SUSTAINABLE FOOD PRODUCTION

1st Joint Meeting of Agriculture-oriented PhD Programs

UniCT, UniFG and UniUD

17-21 June 2019, Catania-Salina

UNIVERSITÀ degli STUDI di CATANIA

UNIVERSITÀ DI FOGGIA

UNIVERSITÀ DEGLI STUDI DI UDINE
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INSTITUTO SUPERIOR DE AGRONOMIA
Universidade de Lisboa

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IMPRESSUM

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HIGHER EDUCATION FOR SUSTAINABLE FOOD PRODUCTION

Abstracts of
"1st Joint Meeting of Agriculture-oriented PhD Programs at Unict, Unifg and Uniud"
Edited by S. Tortorici and A. Priolo
Catania – Salina, Italy - 17th – 21st June

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June 2019

Monday June 17, Catania, Main Hall, Via S. Sofia, 100

15:30: Welcome address

Alessandro Priolo: coordinator PhD in Agricultural, Food and Environmental Science, Unict

Agatino Russo: Director - Department of Agriculture, Food and Environment, Unict

SESSION 1 (Chairpersons: Alessandra Barlaam - Giorgio Gusella)

Main lecture: The use of semiochemicals in biological control

José Carlos Franco, Instituto Superior de Agronomia, University of Lisboa (Portugal)

Pomegranate by-product as feed: effect on lamb meat quality

Antonio Natalello, Unict

Investigating the potential of hyperspectral imaging for early detection of chilling injury in green bell peppers

Farahmand Babellahi, Unifg

A multifactorial approach to honey bees health: from multiple causes to consequences and possible actions

Davide Frizzera, Uniud

Tuesday June 18, Salina

SESSION 2 (Chairpersons: Amanda Vaccalluzzo – Abdul Moiz)

15:45 - 16:00: Welcome address from the mayor of di Santa Marina di Salina *Domenico Arabia*

16:00 - 16:25: Methicillin resistant *Staphylococcus aureus* (MRSA) in raw buffalo milk and its fate along the human gastrointestinal tract

Elisa Spinelli, Unifg

16:25 - 16:40: New Breeding approaches to improve mildew resistance in wine grape cultivars

Loredana Moffa, Uniud - CREA-VE

16:40 – 17:00: Modular modelling of energetic crops in marginal lands on a climatic change scenario

Sebastiano Andrea Corinzia, Unict

17:00 – 17:15: Understanding of meat quality management from “farm to fork” by an “omic” approach

Martina di Corcia, Unifg

17:15 – 17:30: Modulation of Microbiome in dog and adaptative response of microbiome-gut-brain axis

Elisa Scarsella, Uniud

17:30 – 17:55: Gluten-FriendlyTM bread increases mucus production and gut-barrier function in human intestinal goblet cells

Daniela Musaico, Unifg

17:55 – 18:10: Plant suitability to the South American tomato pinworm and pest-induced resistance in wild and commercial Solanum species

Simona Tortorici, Unict

18:10 – 18:30: Development of synthetic methods, namely oligonucleotide aptamers, in order to stimulate the plant defense response in Arabidopsis thaliana

Federico Bosetto, Uniud

18:30 – 18:50: Exploring the role of downy- and powdery-mildew susceptibility genes in Vitis bystudying the genetic variation and the in-vivo function

Carlotta Pirrello, Uniud and Edmund Mach Foundation (TN)

18:50 – 19:10: Focusing on early defence responses in Arabidopsis sieve elements during phytoplasma infection

Chiara Bernardini, Uniud

19:10 – 19:30: Reducing mechanical damage induced by fresh-cut processing

Alessia Incardona, Unifg

20:00: Dinner at “Il Gambero”

After dinner keynote: DNA origami and nanorobots

Giuseppe Firrao, Uniud

Wednesday June 19, Salina

SESSION 3 (Chairpersons: Carlotta Pirrello – Domen Skrab)

9:00 – 09:25: **Keynote** - Predictive modeling to estimate shelf-life of ready-to-eat fresh produce

Giancarlo Colelli, Unifg

09:25 -09:40: Sustaining Low-Impact Practices in Horticulture Through Non-Destructive Approach to Provide More Information on Fresh Produce History & Quality

Abdul Moiz, Unifg

09:40 – 09:55: Characterization of new fungal diseases of Pistachio in Sicily and development of sustainable management strategies

Giorgio Gusella, Unict

09:55 – 10:15: Biochemical, metabolic and molecular characterization of pear-apple hybrids: PEARAPPLE-Omics.

Giulia Pasqualetto, Uniud, Edmund Mach Foundation (TN) and Plant & Food Research Institute (NZ)

10:15 – 10:35: Sustainable durum wheat supply chain: the effects of genotypes and low nitrogen levels on grain yield and technological quality

Federica Carucci, Unifg

10:35 – 10:50: Dietary tannins to improve efficiency and product quality in ruminant extensive production systems

Ruggero Menci, Unict

10:50 – 11:10: Identification of histological and immunological indicators of larvae/juveniles quality in marine fish species

Valentina Pacorig, Uniud

11:10 – 11:30: Hyperspectral Fluorescence Imaging Method for Early Detection of Mature and Immature Green Tomatoes (Solanum Lycopersicum L.)

Danial Fatchurrahman, Unifg

11:30 – 11:45: Sustainable building components from agricultural waste

Monica Parlato, Unict

11:45 – 12:05: Mountain products and ecosystem services: assessment methods and enhancements strategies.

Chiara Spigarelli, Uniud

12:05 – 12:20: DNA barcoding for the identification of fish species in processed products

Ashraf Ali, Unifg

12:20 – 12:35: Utilization of cover crops and biodegradable mulching for the sustainable management in a Mediterranean agro-ecosystem

Stefania Fontanazza, Unict

12:35 – 12:50: Multi-omic signature of the grapevine resistance to Downy mildew (DM) and Powdery mildew (PD)

Ramona Mihaela Ciubotaru, Uniud, Edmund Mach Foundation (TN)

12:50 – 13:05: Cisgenesis and genome editing approaches to improve health-promoting properties on citrus fruits.

Fabrizio Salonia, Unict, CREA

Wednesday June 19, Salina

SESSION 4 (Chairpersons: Monica Parlato – Francesco Ciampi)

14:30 – 14:55: Effect of innovative husbandry strategies on meat quality

Silvia Del Bianco, Uniud

14:55 – 15:15: Investigating the effector-target interaction and its role in Apple Proliferation disease

Mattia Tabarelli, Uniud, Edmund Mach Foundation (TN) and Research Centre for Agriculture and Forestry Laimburg (BZ)

15:15 – 15:30: An alternative approach to assess the effects of clogging on the constructed wetland by testing the pollutants concentration removal efficiency

Alessandro Sacco, Unict

15:30 – 15:50: Printing cereal dough: novel sensory perception of food from plant tissue microstucture

Maddalena Paolillo, Unifg

15:50 – 16:10: Optimization of agronomical and technological parameters of Ribolla Gialla for the production of quality sparkling wine

Domen Škrab, Uniud, Edmund Mach Foundation (TN)

16:10 – 16:30: Novel ingredients and underexploited feeds to improve the sustainability of farmed fish species.

Roberto Cerri, Uniud

17:45: Departure from the dock of Lingua to Panarea and Stromboli on board of the GLENTOR. Social dinner on board

Thursday June 20, Salina

SESSION 5 (Chairpersons: Valentina Pacorig – Andrea Corinzia)

9:30 – 09:55: **Keynote** - Future challenges for a sustainable animal production

Giuseppe Maiorano, Unimol

09:55 – 10:20: *Antioxidant effect of natural extracts on bovine aortic endothelial cells*

Francesco Ciampi, Unifg

10:20 – 10:35: Determinants of frost resistance in fruit crops Vitis vinifera, Prunus persica and Malus domestica

Valeria De Rosa, Uniud

10:35 – 10:55: Heath and eco innovation in food packaging

Antonella Cammarelle, Unifg

10:55 – 11:10: A multidisciplinary approach for citrus rootstocks evaluation

Giulia Modica, Unict

11:10 – 11:30: Investigation on Cyclospora cayetanensis in vegetables and imported soft fruits

sold on Italian market by microscopy and standardized qPCR

Alessandra Barlaam, Unifg

11:30 – 11:50: Detection and identification of insect compounds in fish diet

Enrico Daniso, Uniud

11:50 – 12:05: Omics approach to explore microbial interaction of table olives consortium

Amanda Vaccalluzzo, Unict

12:05 – 12:20: Functional characterization of resistance genes against Apple Scab

Ayesha Yousaf, Uniud and Edmund Mach Foundation (TN)

12:20 – 12:35: Physiology of stress response in grapevine

Marco Vuerich, Uniud

Thursday June 20, Salina

16:00 – 16:45: General conclusions in plenary session

17:00 – 20:00: Discussion groups in parallel sessions

Friday June 21, Salina

09:00 – 10:30: individual feedback

Session 1

Pomegranate by-product as feed: effect on lamb meat quality

Antonio Natalello - antonio.natalello@unict.it

PhD in Agricultural, Food and Environmental Science, University of Catania

Due to the potential benefits of pomegranate fruits (*Punica granatum* L.) on human health, there has been a great increase in the demand of those fruits, and especially for juice production. Consequently, a huge amount of waste is produced after juice extraction. This waste, indicated as whole pomegranate by-product (WPB), contains peels, seeds and residual pulp and could be used in ruminant feeding. To investigate this alternative feed, an *in vivo* experiment with lambs was carried out. Animals were assigned to two dietary treatments (for 36 d): the control group was fed to a commercial concentrate (CON; 8 lambs), while the other group was fed with a concentrate containing 20% of WPB (WPB; 9 lambs). Animals were slaughtered after the feeding trial and samples of rumen digesta, liver and muscle were collected for fatty acid composition analysis. The WPB diet increased the intramuscular concentration of healthy fatty acids (FA), such as CLA, vaccenic acid and polyunsaturated FA. These results could be explained by the presence of both tannins and/or CLnA (conjugated linolenic acids) in the pomegranate. However, it was not possible to attribute our results to one of these two bioactive compounds. Therefore, two *in vitro* experiments were planned and simultaneously carried out. Pomegranate tannins and CLnA (*study 1*) or WPB fractions (peels and seeds; *study 2*) were separately fermented in a system that simulates the rumen functioning. From these results, it seems that pomegranate tannins have an effect on increasing the CLA, while the pomegranate CLnA might manly be converted into vaccenic acid in the ruminal digesta. Moreover, meat from *in vivo* trial was used to investigate the shelf-life and volatile organic compounds (VOC). In brief, colour stability and lipid oxidation were assessed in fresh and cooked meat over 7 days of refrigerate storage. Antioxidant status and vitamin E were also determined. Meat from lambs fed WPB showed less susceptibility to lipid oxidation over the days of storage. Furthermore, lipophilic fraction of WPB muscle displayed a greater antioxidant activity (ORAC assay). These results could be due to the higher vitamin E concentration found in WPB muscle. Indeed, WPB (particularly seeds) is naturally rich of vitamin E which was ingested and deposited in muscle. In conclusion, pomegranate diet improved the fatty acid composition, shelf-life and vitamin content of meat, without compromising animal growth performances and reducing feeding costs.

Investigating the potential of hyperspectral imaging for early detection of chilling injury in green bell peppers

Farahmand Babellahi - farahmand.babellahi@unifg.it

PhD in Management of innovation in the agricultural and food system of the Mediterranean region, University of Foggia

Chilling injury (CI) or cold injury is a physiological disorder which could be occurred by unsuitable temperature at postharvest phase that results in the damage of fruit cell membranes. Early detection of chilling injury (CI) in bell pepper when subjected to low temperature conditions is very important for high marketability, improved produce, and high throughput. The existing methods to detect chilling injury are time consuming, destructive, invasive, and require interaction with chemicals. Therefore, sorting and grading industries demand a non-destructive technique for rapid and accurate detection of CI. This research aims to utilize hyperspectral imaging in the VIS-NIR (400-1000 nm) range combined with chemometrics tools in order to detection of chilling injury in early stage of chilling process. For this purpose, 126 fresh bell peppers in mature green stage were purchased and stored in 4 0C and 12 0C as a chilling and safe temperature, respectively for 18 days. After removing fruits by 6 days interval and leaving one day at room temperature as a shelf-life, image was acquired. PLS-DA was the supervised classifier which applied on data as two steps classification models. At first step, based on whole wavebands, fresh fruits and those were stored at 120C and 40C could be classified with 88% and 84.33% non-error-rate for calibration and classification, respectively after pre-processing data (MSC+mean-centering). In second step, classification model was applied on 4 0C fruits, since bell peppers start chilling process after 14 days at temperatures under 7.5 0C. So, for early detection of chilling process realizing days of storage in improper temperature is crucial. Second model could classify day 6, day 12, and day 18 fruits with 100% and 96% non-error-rate for calibration and classification respectively after pre-processing. The results demonstrate the applicability of the integrated VIS-NIR hyperspectral imaging for early detection of chilling injury in bell peppers.

A multifactorial approach to honey bees health: from multiple causes to consequences and possible actions

Davide Frizzera - frizzera.davide@spes.uniud.it

PhD in Agricultural Science and Biotechnology, University of Udine

Honeybee plays a fundamental role in crop production and biodiversity maintenance. During the last decades, widespread losses of honey bee colonies were reported in the northern hemisphere causing great concern for the beekeeping and ecological implications. It has been shown that losses have a multifactorial origin, with pathogen and parasites playing a key role but also environmental conditions concur to shape this worrying phenomenon. Specifically, temperature is one of the principal abiotic factors that these organisms have to cope with. In this study we investigated the effects of low temperatures on honey bees and their interactions with biotic stressors (i.e. Varroa destructor and Deformed Wing Virus), focusing our attention on the fundamental role of nutrition as an essential pre-requisite for an adequate immune response.

Session 2

Methicillin resistant *Staphylococcus aureus* (MRSA) in raw buffalo milk and its fate along the human gastrointestinal tract

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The aims of the study are: i) to assess the occurrence of MRSA in buffalo dairy farms and buffalo tank milk from Italy ii) to study the fate of MRSA along the human gastrointestinal tract and its interaction with the gut microbiota. Seventy-five bulk tank milk (BTM) samples from farms and 24 nasal swabs from farm workers were collected, respectively. Three (4%) out of 75 BTM samples and 1 (4%) out of 24 nasal swabs were MRSA-positive. The milk isolates showed the following genotypes: ST1/t127/Va and ST72/t3092/V, while the human isolate was characterized as ST1/t127/IVa. No ST398 were found. All the isolates were multidrug resistant but vancomycin susceptible, carrying the icaA gene, while they tested negative for pvl and ses genes. This study demonstrates for the first time in Europe that MRSA might be present in dairy buffalo farms and in raw buffalo milk. Regarding the second aim, a MRSA ST398/t011/V strain, previously isolated from raw cow milk, and a human origin MRSA strain were inoculated into two foods of animal origin respectively. The pH of the matrices was gradually decreased to 2.0 in 2 hours, during which time they were kept at 37 °C and periodically homogenized. The same MRSA strains levels were inoculated within an intestinal in vitro simulator and it was periodically analyzed their fate along the whole transit. Mucin agar carriers replaced the intestinal mucus layer and a basic feed medium represented the intestinal lumen contents. A three-day in vitro study was performed using microbiota from the pooled faeces of healthy individuals that were stabilized simulating colon conditions. The MRSA population survived the decreasing gastric pH levels unharmed, but it was affected by the organic acids produced by the enteric microbiota along the transit into the simulator. It was, in fact, no longer viable after 24 h of incubation with luminal colon microbiota, whereas counts of 4 log cfu/g were still obtained in the mucin agar carriers after 72 h of incubation. Despite the ability of MRSA to overcome human stomach acidic conditions, these results confirm the hypothesis that competitive microbiota may control MRSA intestinal colonization.

New Breeding approaches to improve mildew resistance in wine grape cultivars

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Vitis vinifera sp. is one of the most important economical resource for many countries; in particular, the European grape productions accounts for about 90% of worldwide wine grapes, sparkling, table grape and raisin productions. Worldwide grapevine cultivation is based on the use of elite cultivars that shows low resistance to the oomycete *Plasmopara viticola* (downy mildew) and the ascomycete *Erysiphe necator* (powdery mildew). These pathogens annually produce significant yield losses. To manage these and other grape diseases and pests many pesticides applications are required, making the viticulture one of the agricultural activities with the greatest chemical input, and negative consequences on the environment and consumer health. Recently, the genetic improvement of the grapevine took great advantages from New Plant Breeding Technologies (NPBTs) which combine the traditional breeding techniques with the most recent molecular tools shortening the selection times and costs and limiting the effect on the genome of the cultivar of interest. The two breakthroughs in breeding programs derived from availability of molecular markers and NPBTs such as CRISPR-Cas9: the molecular markers derived from genetic analyses have been integrated in the breeding process to assist both the selection of candidate varieties carrying the desired gene combinations as well as the introgression of a resistance gene; the CRISPR-Cas9 technology has the potential to operate in a targeted way to obtain an accurate editing without altering the genetic heritage of the cultivars. In this context feasible methods for plants transformation includes *Agrobacterium tumefaciens*-mediated transformation and direct gene transfer into protoplasts (direct delivery). The Agrobacterium-mediated gene transfer has emerged as the most widely used method in plant genetic engineering as shows by International Service for the acquisition of Agri-Biotech Applications (ISAAA). In this project molecular markers and NPBTs will be used to obtain *Plasmopara viticola* and *Erysiphe necator* resistance in some economically important *V. vinifera* cultivars.

Modular modelling of energetic crops in marginal lands on a climatic change scenario

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The main aim of the PhD project is to develop a model solution for biomass and energetic perennial crops on marginal lands. This solution would help Mediterranean agriculture to deal with the climate change scenario and extreme climatic events; to identify new exploitable lands for biomass and energy among marginal areas; to define the best management practices suitable for these areas, including the choice of the crop species and to quantify the environmental impact of these crops. Two lines of research can be identified within the project. The first line of research is based on the agronomic approach to collect empirical data from field trials on biomass crops, focusing on the relationship genotype x irrigation management. The trial assesses the productivity, the physiological (leaf gas exchange) and morphometric (leaf area, solar radiation interception and above ground biomass growth) traits of 9 genotypes of 3 species (*Miscanthus* × *giganteus*, *Miscanthus* genotype 9, *Miscanthus* genotype 10, *Arundo donax* ecotype Catania, *Arundo donax* ecotype Morocco, *Saccharum spontaneum* spp. *Aegyptiacum*) under three irrigation levels (rainfed, 50% and 100% of potential evapotranspiration) during three subsequent growing seasons. The second research line is based on the modellistic approach in order to provide predictions for biomass and energy crops production and impact, on climate change and marginal scenarios and to define marginal suitable areas and optimized agronomic management practices for biomass and energy crops grown in these areas. The activities concerns the implementation and validation of biophysical models to simulate the response of crops under different climatic and environmental conditions, and thus the geo-spatial analysis of the results. A biophysical model have been developed to define the optimal sowing date and the seed ripening date for castor (*Ricinus communis* L.) and thus to obtain suitability maps for castor crop in Sicily. The model is based on castor thermal and photoperiodic requirements for seed germination and crop phenology, defined during a field trial.

Understanding of meat quality management from “farm to fork” by an “omic” approach

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Meat has an important part on the human diet, providing proteins with high biological value and essential nutrients, some of which are more bioavailable than in alternative food sources. Although the meat nutritive value plays a remarkable role on the healthiness, the consumer’s decision to purchase meat is guided also by a variety of organoleptic traits including colour, tenderness, juiciness, and aroma or flavour. Tenderness is probably one of the predominant criteria that determines acceptability of meat. Many authors reported that meat tenderness is highly dependent by exogenous and endogenous factors like genotype, age, sex, type of muscle and management factors as diet, stress, pre-slaughter handling and post-mortem aging. The aim of my PhD project will be to evaluate the effect of livestock management from “farm to fork” on animal welfare and on meat quality by using an "omic" approach. The “omic” technologies seeks to understand and discover the mechanisms involved in many biological processes that determine animal productivity, product quality and health as a result of interactions between feed, environment, animal genetics, physiology and its symbiotic microbiota. Particularly, proteomics is applied to study protein modifications that are indicators of meat quality and its defects such as reversible phosphorylation, oxidation, degradation and denaturation in post-mortem meat. Application of proteomics for investigating meat quality is a relatively new approach (< 20 years old), it is mainly used to elucidate the relationship between the muscle proteome and the conversion of muscle to meat, tenderness, colour and water holding capacity. During the PhD period, welfare parameters, microbiota and meat quality, particularly muscular protein biomarkers and oxidative status, will be evaluated on selected animals in order to establish the correlation between animal health and its productivity.

Modulation of Microbiome in dog and adaptative response of microbiome-gut-brain axis

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Dogs intestinal dysbiosis and food sensibilities had several origins and present complex symptoms, but they have in common the increase of the gut permeability and the instauration of a local inflammatory response. These situations often derive from an interaction with the intestinal microbiome, altered by a prevalence of some bacteria taxa within the microbial population, which can also lead to a decrease of the gut microbiome biodiversity. Several studies have been made to find a correlation between the gut microbial community and the onset of these disturbs. At this point, microbiota reference values are still hard to define because of the great individual variability, based on genetic, epigenetic and environmental characteristics. Breed, cross-breed, gender, age, immune-education during the first stage of life and nutrition are some of the factors that can influence the gut microbiome. Moreover, it has been recently underlined the function of the gut microbiota as a neuroendocrine organ, with the production of neurotransmitters that can regulate mood, food habits, energy intake and immune response. Intestinal microbiota data of healthy dogs from different breeds, genders, ages and food habits will be collected, with the final goal to build a microbial database to use as a reference for the clinic practice. These data will be compared with information from dogs with gastrointestinal and dermatological disorders, to see if there is any variation in the composition of microbial community. Furthermore, changes in faecal microbiome due to the modulation of diet with several nutrients will be studied, searching for a relationship – if any - between dog microbiome and adaptative neuroendocrinal response. The research activity will be focused on the characterization of microbiome by analysis of faecal DNA, with the use of High-throughput DNA sequencing, and on the characterization of metabolome, by analysis of microbial end products through Nuclear Magnetic Resonance. In addition, biochemical and endocrinological analysis will be carried on, to characterize the amount and the production of endocrine molecules, such as serotonin and cortisol, in biological fluids.

Gluten-FriendlyTM bread increases mucus production and gut-barrier function in human intestinal goblet cells

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Gluten FriendlyTM is a temperature-based technology that is applied directly to wheat kernels before milling. Based on early in vitro investigations, it has been postulated that the Gluten FriendlyTM process reshapes the tertiary structure of gluten proteins, without altering the secondary or primary structure. The temperature-treated gluten maintains most of its organoleptic and viscoelastic properties. Previous studies have shown that Gluten FriendlyTM bread (GFB) was able to modulate *in vitro* the qualitative-quantitative gut microbiota composition in both healthy and coeliac donors. Additionally, GFB prolonged the survival of *Lactobacillus acidophilus* and had antibacterial effects towards *Staphylococcus aureus* and *Salmonella Typhimurium*. In a symbiotic fermented milk, Gluten FriendlyTM flour (GFF) affected positively the viability of the probiotic *L. acidophilus* La-5. Moreover, in a symbiotic yogurt supplemented with GFF and *Bifidobacterium infantis*, GFF prolonged the viability of the probiotic for 14 days without compromising the technological performances of the starter cultures. This study aimed at investigating in vitro the effects of GFB and Control bread (CB) on intestinal epithelium mucus secretion and barrier permeability in healthy human mucus-secreting goblet cells HT-29-16E. Mucus secretion in cells exposed to digested GFB and CB was preliminarily investigated using staining techniques, Periodic Acid-Schiff (PAS) and Alcian blue (AB). MUC2 and MUC3 were also quantified by ELISA assay. The barrier permeability of the cell monolayer was evaluated by trans-epithelial electrical resistance (TEER) measurements. GFB increased secretion of mucins, expressed as levels of PAS and AB staining in comparison to the control. While MUC3 levels were not affected, higher MUC2 concentrations ($P<0.01$) were observed on cells treated with GFB compared to the control. Additionally, significantly higher TEER values were observed after treatment with GFB in comparison to the CB ($P<0.01$). While further researches are recommended to confirm the *in vitro* outcomes presented in this study, the observed effects have the potential to effectively contribute to a better intestinal functionality.

Plant suitability to the South American tomato pinworm and pest-induced resistance in wild and commercial *Solanum* species

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Among key tomato pests, the South American tomato pinworm, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae), is one of the most dangerous, particularly across the Afro-Eurasian continent since its first invasion in Europe in 2006. This pest has been historically subjected to chemical control since the seventies in South America, however this strategy led to the emergence of multi-resistant populations. As a consequence, alternative control strategies have to be prioritized and, among them, the exploitation of plant resistance could represent a valid sustainable control tool. Within this context, eleven tomato varieties were chosen among those commercially grown in protected crops of the Southern Mediterranean area on the bases of different agronomic traits and fruit types. The suitability of leaves from healthy plants of the varieties as feeding substrate for larvae of *T. absoluta* was estimated. Results showed a strong diversity in plant suitability for the development of *T. absoluta* larvae in terms of larval survival, larval development time, weight of the pupae and eroded leaf area. The potential for *T. absoluta* larvae to elicit systemic resistance mechanisms was also assessed. For this, we used healthy leaves from plants previously infested with *T. absoluta* larvae. The obtained results suggested that few varieties have potential for induced plant resistance, because on these species *T. absoluta* larvae developing on leaves from pre-infested plants suffered a low survival and fed less than those feeding on leaves from healthy plants. Therefore, the varieties showing promising results, together with wild and further commercial species belonging to the *Solanum* genus (e.g., eggplant), are going to be test in further experiments. These are aiming at assessing the susceptibility to the pest attack, the potential pest-induced resistance mediated by resistance genes expressing the synthesis of allelochemicals, both volatile and non-volatile compounds. The results will provide evidences on potential direct antibiosis and antifeedant activities of induced substances on *T. absoluta* behavior and development.

Development of synthetic methods, namely oligonucleotide aptamers, in order to stimulate the plant defense response in *Arabidopsis thaliana*

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Aptamers are small synthetic nucleic acids that fold into a well-defined three-dimensional structure in order to specifically and tightly bind to a variety of diverse molecular targets. Aptamers evolve from random oligonucleotide pools by an *in vitro* selection process called systemic evolution of ligand by exponential enrichment (SELEX). Cell-SELEX is a novel type of SELEX which uses whole cells as enrichment targets. Moreover, it has been reported that several aptamers identified through Cell-SELEX not only bind to the target cells, but they are also intracellular transported. The aim of my PhD project is, starting from a DNA library with a random region of 20 nucleotides, to select proper aptamers by Cell-SELEX that either specifically bind membrane receptors involved during the plant defense response or internalize into cell cytoplasm. The Cell-SELEX methodology was performed on mesophyll protoplasts isolated from Arabidopsis thaliana. To date, the Cell-SELEX protocol was designed to recover potential aptamers either binding the cell membrane or internalizing into cell cytoplasm on protoplasts that are not immune-stimulated. To monitor the convergence of the aptamer species during the selection process a combined strategy dependent on quantitative real-time polymerase chain reaction (qRT-PCR) amplification curve and high resolution melting (HRM) curve analysis was employed. An actual aptamer class selection from the library is expected to be associated with a peak fluorescence shift from lower to higher melting temperatures, implying that convergence of aptamer species is occurring. A first Cell-SELEX was conducted from round 1 to 5. The main fluorescence signal peak at 74 °C, typical of first rounds, was progressively flanked by a fluorescence signal peak at 81 °C. Selected putative aptamers were cloned in an appropriate cloning system and sequenced by Sanger-method. One putative aptamer showed a G-rich sequence that could have the propensity to form a G-quadruplex structure possibly involved in the binding of the target. A second Cell-SELEX was carried out up to the sixth round, and a similar trend seen during the first Cell-SELEX was shown. In both Cell-SELEX experiments it was found that the cell-internalizing oligonucleotides always showed similar melting curves through the different rounds, with the fluorescence peak at 81 °C resulting gradually larger as the selection rounds increased. A similar behaviour was also found for the oligonucleotides which bind membrane cells, but only when a prior dilution of at least 1:10 had been carried out; conversely, when the qPCR-HRM was performed on the undiluted samples the fluorescence peak at 81 °C was never detected. The designed work will permit to develop nucleic acid sequences that specifically recognize defense membrane receptors, and in turn they can readily induce the immune response. Moreover, the possibility to select sequences that internalize cells may have prompt applications in plant biotechnology, such as transfection, carrying and many others.

Exploring the role of downy- and powdery-mildew susceptibility genes in *Vitis* by studying the genetic variation and the *in-vivo* function

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Being one of the most spread cultivations all over the world, grapevine has been studied for several aspects, including its resistance/susceptibility to pathogens. To reduce the use of environmentally-impacting fungicides, breeders have crossed the European *Vitis vinifera* with American and Asian wild species to obtain plants resistant to the most important grape pathogens such as the oomycete *Plasmopara viticola* and the ascomycete *Erysiphe necator* (causal agents of downy and powdery mildew, respectively). However, till recently the wine quality derived from these plant-hybrids was clearly lower than that derived from pure V. vinifera varieties. For this reason, the investigation on resistance loci/genes has been the main trend to understand the interaction between grapevine (*Vitis spp*.) and mildew causal agents in the last decades. Dominantly inherited gene-based resistance has shown to be race-specific in some cases, not to confer total immunity and to be potentially overcome within a few years. Recently, the identification of susceptibility genes in plants and crops, as factors required by the pathogen to infect the host-tissue has recently opened up their exploitation as an alternative strategy to breed resistant plants. On the footprint of research conducted on Arabidopsis and barley, genes associated to downy (DM) and powdery (PM) mildew susceptibility have been discovered also in the grapevine genome. Aim of this project is to investigate the role of recessively-inherited susceptibility genes, whose total or partial inactivation can lead to broad-spectrum and long-lasting resistance to downy mildew. To do so, the investigation has been carried out following two parallel strategies: to explore naturally-occurring genetic variation of DM and PM susceptibility genes throughout 190 grape accessions and studying their *in-vivo* function through transcriptional analysis on mutant plants obtained with genome editing.

Focusing on early defence responses in Arabidopsis sieve elements during phytoplasma infection

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Phytoplasmas are unculturable phloem-limited prokaryotes, discovered only 50 years ago and associated with numerous diseases affecting different crops. They are obligate pathogens with a very small genome, lacking many genes, indispensable for the autonomous life. The phytoplasma reliance on host metabolism causes severe alterations of plant cytology and physiology, resulting in the dramatic yield reduction. As direct methods to control phytoplasma diseases are not available so far, it is important to study in depth the strategies adopted by the plant to face phytoplasma infection. Sieve-elements are able to contrast the stress imposed by wounds or pathogen invasions, by the activation of Sieve Element Occlusion Related (SEOR) proteins or of callose, limiting the mass flow, and thus avoiding sugar losses or blocking pathogen spread. In *Arabidopsis thaliana* two SEOR genes concur to the formation of sieve-element protein filaments (*AtSEOR1* and *AtSEOR2*), through a heteromeric assemblage of the two proteins they encode. Our research group previously demonstrated that in the Arabidopsis line silencing Atseor1 gene (*Atseor1ko*), so expressing 'free' AtSEOR2 protein, phytoplasma titre is significantly lower in comparison with wild-type or *AtSeor2ko* plants. This led to speculate that AtSEOR2 protein, free from its bond with AtSEOR1, could act in plant-defence mechanism. Nevertheless, no explication of this phenomenon was provided. Aim of my research project is to investigate the possible involvement of AtSEOR2 protein in plant reactions against phytoplasmas. The effective defence responses occur precociously during plant-pathogen interaction, so we focused our investigations both in fully symptomatic plants at the late stage of infection and in plants yet asymptomatic, i.e. at an early stage of infection. As literature reports possible interferences of AtSEOR2 with the phytohormone signaling system, in particular with the indol-acetic acid (IAA) and abscisic acid (ABA) pathways, we quantify phytohormones in wild-type, *Atseor1ko*, *Atseor2ko* Arabidopsis lines following phytoplasma infection. Moreover, with the aim to evidence possible site-specific response we performed analyses using leaf laminae or midribs. Interestingly the amount of IAA and ABA are significantly higher in *Atseor1ko* plants compared to the other lines, starting from the first stage of infection, when macroscopical symptoms, sieve-element ultrastructural modifications and modulation of Atseor genes are not yet detectable in infected plants. The project will continue focusing on the second occlusion system activated in injuried sieve elements: the callose deposition at the sieve plates, regulated, in Arabidopsis, by the expression of the Callose synthase 7 (AtCas7) gene. The Arabidopsis mutant line *AtCas7ko* will be infected with phytoplasmas, with the aim to study not only the role of the long-term callose-mediated occlusion in infected Arabidopsis, but also to evidence variations in sugar metabolism, also related to the early plant defense signaling.

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Reducing mechanical damage induced by fresh-cut processing

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A continuous demand for fresh, healthy and convenient food products, especially fresh-cut fruits and vegetables, has risen in the previous decades. Wounding stress causes tissue deterioration in terms of browning and softening. Mechanical damage is function of cutting in case of fresh-cut process as a corollary to which tissue damage occurs, hence impacting the quality of the ultimate product in terms of nutritional value, visual appearance and shelf life. Cutting tool sharpness and morphology of the cut tissue possess a direct correlation with the extent of mechanical damage. The degree of sharpness (defined as the force exerted by the instrument to cut a reference body) is an important factor for an efficient cutting operation and a methodology has been developed in order to formulate a quantitative index (blade sharpness index, BSI). The aim of the present study is to define a methodological approach to evaluate the degree of sharpness of a cutting tool, correlating it with the mechanical damage on the plant tissue (based on biochemical, physiological and morphological analysis) with the objective to define a threshold within which maximum cutting efficiency can be achieved also in terms of shelf-life of the fresh-cut produce. In addition, it will also explore the combination of BSI and treatments on fruit and vegetable products in order to decrease the effects of mechanical damage. Keeping under consideration these general objectives, preliminary trials for method standardization were performed and results are hereby presented.

Session 3

Sustaining Low-Impact Practices in Horticulture Through Non-Destructive Approach to Provide More Information on Fresh Produce History & Quality

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Sustaining quality of production and of the environment by means of low input agricultural practices (LIP) and non-destructive (ND) quality evaluation is a novel approach being used worldwide. ND quality evaluation will be an additional tool for discrimination of fresh products obtained with LIP and estimation of quality and shelf-life of packaged products. Product quality will be assessed measuring conventional quality attributes, including physical, sensorial and nutritional quality with emphasis to biologically active compounds. Each attribute will be correlated with spectral information obtained with NIR and hyperspectral imaging systems with the aim of building a prediction model. The performance of each model will be evaluated. The crops produced with different inputs will be discriminated. Spectra will be analysed in order to study the impact of LIP on spectra variation and to select the spectral range significantly representing the agricultural practices. Discriminant models will be applied to model the differences among the treatments. Quality assessment of packaged products will be Identification of optimal colours for designing packaging that improve the quality evaluation by Computer Vision Systems (CVS) Color correction of images is critical to make meaningful the quality prediction and discrimination by a CVS. The activity will substitute color references (color-charts) normally used for this task with coloured graphical elements introduced in properly designed packaging. For each product, different experimental trails, involving packaged and unpackaged samples, will be carried out. Chemical and physical analysis will be statistically correlated to color parameters extracted from images acquired by a CVS on the same samples with and without packaging. Multivariate analysis and machine learning techniques will be used for quality evaluation through packaging. The obtained findings will be utilized in order to design strategies to ensure better marketing conditions for fresh produce from the LIP. Other than the visual and organoleptic attributes representing conventional quality standards and regulations, the nutritional values using environment-friendly methods like ND has a great influence on the production processes of food commodities. Better crop performance with enhanced nutritional attributes and sensorial quality are key drivers in developing strategies of efficient utilization of resources (fertilizers and water) in irrigation of greenhouse agriculture. The methodology will be designed based on proper LIP and ND quality determination for tomato (*Solanum lycopersicum* L.) and rocket leaves (*Diplotaxis tenuifolia* L.) diffusely produced in greenhouses. The LIP experiments will be first aimed to reduce the agricultural inputs in the greenhouses by optimizing the use of water and fertilizers in soilless and soil cultivation. In second step quality attributes of the production will be assessed by conventional and ND approaches, with emphasis on possible differences deriving from the application of LIP, developing algorithms for discrimination based on spectral information. Then, in the possible influence on consumer choices of certification of LIP practices resulting from ND methods and their willingness to pay will be tested, in order to produce adequate and realistic marketing strategies. Finally, dissemination of optimal execution of the project both from the scientific and financial point of view and coordination activities will be carried out into the scientific community and to potential users.

Characterization of new fungal diseases of Pistachio in Sicily and development of sustainable management strategies

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In Italy the pistachios production is condensed in the southern regions with Sicily considered as the first Italian producer. Concerning the Sicilian pistachio cultivation, we have few and outdated elements to evaluate the current phytopathological situation. Aim of the project is the identification and characterization of fungal isolates associated with branch and twig cankers, discolourations, blight and foliar and fruit lesions of pistachio. This first part needs fields surveys, sampling and laboratory isolations, followed by morphological and molecular methods to properly identify the fungi associated with observed symptoms. Second part of the study is to determine the pathogenicity of the most commonly isolated putative pathogens, their distribution and aggressiveness comparison. This second part is important in order to fulfil Koch's postulates and to determine a scale of pathogen risk for the crop in Sicily. Once ascertained the highest risk for the crop is important to focus on different management strategies. The aim of the third part is to evaluate sensitivity of recovered pathogens to multiple fungicides, and, regarding the integrated pest management (IPM) practices, to some approved commercial biological control agents (BA) and Systemic Acquired Resistance inducers (SAR) through *in vivo* assays, in order to develop alternative and sustainable control strategies. The goal of the entire project is certainly to clarify to the current state the phytopathological situation, regarding fungal diseases, for the Pistachio industry in Sicily.

Biochemical, metabolic and molecular characterization of pear-apple hybrids: PEARAPPLE-Omics.

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Malus and *Pyrus* are closely related, with highly co-linear genomes. However, the two genera are characterized by many specific differences, including disease resistances, secondary metabolites, fruit texture, flavor and shape. Hence, intergeneric hybrids between apple and pear provide a unique germplasm resource for genomic analysis, the application of advanced breeding strategies, and new cultivar development. We utilized two sources of apple-pear hybrid plants, 'Zwintscher's hybrid' (Fischer *et al*., 2014) and its F2 progeny held at Edmund Mach Foundation (FEM, San Michele all' Adige, TN, Italy), as well as 95 putative apple-pear hybrids developed at Plant & Food Research (PFR, Palmerston North, New Zealand). We have determined which of the putative apple-pear hybrid genotypes are true hybrids by characterizing their genomes for hybridity using several approaches, including an innovative bioinformatics method developed during the course of my PhD to identify SNPs between apple and pear, which has enabled application of HRM analysis. These results have been validated by SSR analysis and with two SNP arrays. Furthermore, my PhD thesis will describe the genus-specific metabolite pattern in the putative hybrids and will assess resistance or susceptibility to *Erwinia amylovora, Venturia inaequalis* and *Venturia pirina*. Additional putative hybrids between pear and apple have been obtained by germinating seeds in tissue culture. Crosses of pear onto 'Fast Flowering' apple plants are being used to quickly obtain further generations and carry out advanced breeding more rapidly. The use of genomics and other -omics technologies (metabolomics, transcriptomics) will provide insight into the genomic organization of the hybrids. In addition, it will enhance and accelerate the breeding process for the development of crops with improved characteristics for both breeders and consumers, by introducing desired traits from the pear gene pool into apple and viceversa.

Sustainable durum wheat supply chain: the effects of genotypes and low nitrogen levels on grain yield and technological quality

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As a part of PhD project, a study has been conducted in order to optimize the durum wheat yield and quality response through a sustainable agronomic approach in Mediterranean environment, within a supply chain project. To this aim, four durum wheat genotypes (Marco Aurelio, Quadrato, Pietrafitta, Redidenari) were grown during two growing seasons (2016/2017 and 2017/2018) under different nitrogen fertilization (36, 90 and 120 units of nitrogen: N36, N90 and N120, respectively). At harvest the main yield and quality parameters were evaluated; moreover, the gluten protein aggregation level was analyzed by SE-HPLC. In the second growing season, characterized by higher rainfall and maximum temperature during grain filling period, significant lower yield and higher protein content were observed, probably due to a concentration effect, together with the increase of the monomeric and polymeric fraction and the decrease of UPP (unextractable polymeric protein). As for nitrogen fertilization, in the first year characterized by lower rainfall during grain filling period, as normally occurs in Mediterranean environment, N90 level showed the highest yield value but the lowest protein content, associated with higher monomeric/polymeric ratio and UPP. Finally, the four genotypes differed as function of both growing season and nitrogen levels. In particular, Redidenari showed the highest yield, protein values and gluten index in the first year, especially under N90 for yield and protein content. These preliminary results show the possibility of modulating the choice of the genotype associated with low nitrogen inputs, depending on the growing season, to promote a sustainable agronomic approach for the good quality production of durum wheat in Mediterranean environment.

Dietary tannins to improve efficiency and product quality in ruminant extensive production systems

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Extensive ruminant farming systems are known to respect both animals and environment; therefore, sustainable strategies are required to improve efficiency and product quality in such systems. Plant tannins are natural bioactive compounds able to modify ruminants' metabolism thanks to their bioactive properties (e.g. anti-microbial, anti-parasitic, protein-binding, antioxidant). In this study, we are investigating the effects of dietary tannins on rumen fermentation and biohydrogenation, productivity and quality traits of milk and cheese, in relation to fluctuating pasture availability, typical of the traditional husbandry of dairy cows in the Mediterranean. In a first trial, the rumen content of three cannulated sheep was incubated in vitro with two different substrates (vetch pasture and vetch hay) and 2 different tannin extracts (ByPro and ByProQ), each tested at three different doses (0, 15, 30 g/kg). After 24 h, volatile fatty acids, ammonia and fatty acids (FA) were analysed. In a second trial, in spring, 14 pasture-grazing Modicana cows were divided in two groups and fed a supplemental conventional concentrate or the same concentrate with 150 g/head/day of tannin extract (ByPro) for 22 days. During this period, individual milk from evening and morning milking was collected in 10 dates and analysed for proximate composition, somatic cell count, FA, proteins, vitamins, carotenoids, coagulations properties and cheese yield. Furthermore, at the beginning and at the end of the experimental period, 7 kg of milk from each cow were individually processed into cheese and aged for 28 days at 10 °C and 90 % humidity. At the end of the aging period, cheeses were sampled for chemical composition, calcium, FA, vitamins, volatile compounds, proteolysis and texture analysis. The same trial will be repeated in summer, with no availability of pasture.

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Identification of histological and immunological indicators of larvae/juveniles quality in marine fish species

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Gilthead seabream (*Sparus aurata*, GSB) and European seabass (*Dicentrarchus labrax*, ESB) are two of the most farmed fish species in EU. In recent years, the rapid emergence of non-EU competitor farmers has been leading to an economic stagnation. In consideration of this, my PhD activity is conducted in the framework of an EU Horizon2020 funded project named PerformFISH. The mission of this project is to promote the sustainability/competitiveness of Mediterranean aquaculture and in particular, as it is largely recognized that the success of marine aquaculture depends strictly on the production of good quality larvae/juveniles, my study aims at improving the predictability of quality and sustainability of the hatchery phase, validating prognostic histological and immunological indicators of larval/juvenile quality with a multi-modal approach. Good and bad quality ESB/GSB batches, identified on the basis of Key Performance Indicators provided by the farmers, are collected from 4 European hatcheries at 5 different developmental stages (first feeding, flexion, end of larval rearing, middle of the metamorphosis, early juvenile). Specimens are evaluated according to histological and immunohistological protocols. For histological analysis a multiparametric semiquantitative scoring system (scoring range 1-5) has been originally defined for larval/juvenile evaluation. It includes 35 descriptors related to gills, skin, gastrointestinal apparatus, kidney/urinary bladder, yolk sac, swim bladder. Moreover, immunohistochemical stains are performed to support histological findings related to inflammatory events. Preliminary results will be presented.

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Hyperspectral Fluorescence Imaging Method for Early Detection of Mature and Immature Green Tomatoes (*Solanum Lycopersicum* L.)

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The potentiality of hyperspectral fluorescence imaging was evaluated for the non-destructive detection of mature and immature green tomatoes. Hyperspectral fluorescence imaging with excitation wavelength at 365 nm and UV-VIS CCD camera as the detector was employed over mature and green-immature tomatoes from 200 randomly harvested fruits at the green stage. Soluble Solid Content (SSC), pH, total acidity (TA), color change, and conventional sensorial analysis for tomato seed texture were conducted for maturity conformation. The results suggested that over the storage period SSC, pH and TA changed in mature green tomatoes while no change was observed in these parameters in immature green tomatoes. Partial least squares discriminant analysis (PLS-DA) was used for the classification of mature green and immature green tomatoes yielding correct classification with 100% non-error rate. It was revealed that the fluorescence intensity in the surface area of immature green tomatoes was significantly higher in the red region (690 nm) as compared to those of mature green tomatoes. Conclusively, hyperspectral fluorescence imaging can be utilized as an effective and reliable classification tool for classifying mature and immature green tomatoes.

Sustainable building components from agricultural waste

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The valorisation of agricultural wastes (AW) by their reuse could reduce pollution and cost production, realizing a circular economy process which turns them into a resource. Nowadays, the interest for environmental sustainability, energy and efficiency of construction, generally known as Green Buildings, it is strongly increasing. Procedures adopted for Green Buildings are often based on both the reduction of environmental impact of buildings and the improvement of human and animal wellness. This project stems from the need to reuse AW for producing sustainable materials to be used for the construction or renovation of rural buildings. Since a sustainable reuse of AW depends on their availability and geographical location, a methodology was put forward in order to locate and quantify the wastes considered in this research study i.e., plastic films used for protected crops and sheep wool. Application of plastic films for covering protected cultivation enables a significant advantage for the production system. However, without a correct disposal management of plastic wastes, environmental degradation could take place with serious ecological and economic consequences. By using a Geographical Information System (GIS), land use analysis was carried out in an area with the highest concentration of protected crops in Italy (Ragusa province). A suitable index for computing Agricultural Plastic Wastes (APW) was chosen from literature. This first part of the research provides basic information for analysing the environmental impact due to the localization of the APW collection centres, recycling industries or landfills. Next phases of the research study will regard the development of a Life Cycle Assessment (LCA) to investigate the sustainability of the recycling process. Wool sheep is a special waste for his potential bacterial load, with high disposal costs for breeder. In the context of a circular economy, the reuse of greasy wool could reduce both environmental pollution and energy consumption and, as a consequence, could valorize wool sheep becoming a material fully complying with ecofriendly construction criteria. Among building materials, wool sheep fiber could be used as an insulating building component due to its thermophysical performances. In the first phase of the research study, by using data supplied by National Zootechnical Registry, a dedicated GIS was developed to localize and quantify sheep and breeder's sheep in Sicily. Future studies will regard thermophysical and mechanical tests carried out on new building elements made with wool fibers and their comparison with other natural fibers.

Mountain products and ecosystem services: assessment methods and enhancements strategies.

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The gradual abandonment of traditional activities and the difficulties faced in promoting local productions, are challenging mountain cultural and natural heritage and thus the touristic attractiveness of the area. The project aims to support mountain food chains using the policy instruments provided by the optional quality term “mountain product” as defined by EU Reg. 1151/12 and 665/14. The term has not been implemented yet in the transboundary area and its uptake could be beneficial to both producers and consumers by adding value and traceability to local mountain products. Project actions will target regional case-studies in order to identify and suggest solutions to barriers for the adoption of the optional quality term. The innovative approach consists in empowering the “mountain products” by identifying and quantifying ecosystem services (e.g biodiversity, landscape aesthetics, animal welfare, emissions’ regulations) linked to the natural and cultural assets of the area but which are currently hidden into a broader food quality concept. The identification and quantification of ecosystem services allows not only to support high-quality products also in terms of social and environmental sustainability, but also to meet the expectations of tourists and consumers by adopting effective communication strategies on traditional mountain products that contribute to a lively and attractive transboundary area.

DNA barcoding for the identification of fish species in processed products

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Nowadays demands for fish and fishery products are rapidly expanding in all continents because of the increased health benefits and more acceptance in all religions and cultures. Species substitution, the use of a low value fish in place of a high value fish, is the biggest problem in international trade and the leading cause of commercial and sanitary fraud in the fisheries sector. Species identification is a key problem in fisheries research and control, as well as processed fish products in consumer protection. My study aims to evaluate the applicability of the mitochondrial genes, cytochrome b (*cyt* b), and cytochrome oxidase subunit I (COI) for the identification of around 100 fish and processed fish product by techniques of "DNA barcoding". In the present study, universal primers for mitochondrial cyt b will be used to discriminate fish species in raw and processed forms. The barcode primers will be cross-tested against 100 fish species and processed fish product. For this project 100 samples will be collected from different supermarket and of different companies. DNA will be isolated from all samples and it will be amplified by PCR, the most intense amplified product will be chosen for Sanger Sequencing. For mixed products NGS sequencing will be done. After sequencing, the sequencing can match with NCBI BLAST and FISH- BOL database. After obtaining the results species will be identified and will match with the labelling of the products. It will be very significant work in the area of food safety and detection of food fraud in fish and food industry.

Utilization of cover crops and biodegradable mulching for the sustainable management in a Mediterranean agro-ecosystem

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The research project aims to gain new scientific knowledges in order to define innovative agronomical practices and strategies for a sustainable management of polycultural systems, while maximizing the efficiency in the use of native resources (rainfall, solar radiation, phytonutrients), preserving soil chemical, physical and biological fertility, rebalancing the relationship among fruit trees, weeds, cover crops and arthropods community. The final goal is to significantly reduce both external inputs and cultivation costs, and to improve the quali-quantitative aspects of orchards production and for this reason the species have been selected are annual self-seeding legumes (the most suitable plants for the inclusion agroecosystems).In fact these species fully correspond to the principles of sustainability, since they are able to exploit, add, conserve and recycle nutritive elements, control pathogens and weeds and improve soil physical characteristics. In recent decades, the need to quantify microbial populations is a pressing statement in many areas of microbial ecology and has led the scientific community to an increased interest in the ecology of nitrogen transformation. The project, which involves a comparison among different types of soil management in orchards (tilled and without vegetation; covered with weeds; covered with subterranean clover; biodegradable mulching), will analyze the production process from an agroecological point of view, specifically evaluating: soil quality, relationship between agroecosystems components (weeds-cover crop-orchard), orchard productivity, fruits quality, best cover cropping models in orchards to improve water and minerals utilization, techniques for a sustainable crop protection and comparison of cover cropping system and biodegradable mulching. The research project, which is designed following a multi-disciplinary approach, is structured to generate a dual impact, both scientific and applicative. From a scientific perspective, the aim of this research is a better understanding of the relationships regulating the orchard – cover crop – soil microbial community – insect fauna system – biodegradable mulching, in order to maximize the ecological complementarity and minimize the competition among biotic components (weeds, cultivated plants, arthropods), that leads to an optimization of use efficiency of both native and auxiliary resources. From a technical point of view, the project activities performed in a representative farm, operating within the selected area will allow to propose innovative agricultural strategies for both weeds and cover crops management, proper for orchards cultivation in the inland hilly areas of Sicily and able to enhance its productivity and product quality.

Multi-omic signature of the grapevine resistance to Downy mildew (DM) and Powdery mildew (PD)

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The two most economically important diseases of grapevine are Downy mildew (DM) caused by the oomycete *Plasmopara viticola* and Powdery mildew (PD) caused by the *ascomycete Erysiphe necator*. The majority of cultivated grapevines originate from *Vitis vinifera*, a Eurasian species known for its remarkable flavor. However, this species is highly susceptible to the *P. viticola* and *E. necator* which implies a dependency of the grape production on the frequent use of fungicides. One of the most promising strategies to diminish the use of fungicides is to focus on the selection of grapevine varieties showing pathogen-specific resistance. Several genetic factors derived from *Vitis* species have been identified with resistance to DM and PM, but occasionally the protection offered by these resistance genes can be overcome by virulent strains of the pathogens. Interspecific hybrids of *V. vinifera* and North American species, that showed a better resistance, have yielded cultivars with good wine-grape qualities and greater resistance to the pathogens. The most frequent type of resistance is based on a gene for gene interaction with the pathogen followed by a more promising strategy, the pyramiding resistance where several resistance genes are associated in the same variety. The aim of this project is to pursue a multi-omic approach to comprehensively study the metabolic changes in grapevine hybrids with different resistance strategies to *P. viticola* and *E. necator*. The "omics" approach could integrate various associations between genome, proteome, transcriptome, and metabolome which will help in understanding the genotype–phenotype relationship and could lead to the identification of unknown genes and their regulatory networks involved in metabolic pathways of interest. The outcome of the project is expected to pave the way for better understanding of different resistant mechanisms which underline the hybrids-pathogen interaction affecting the *Vitis* species.

Cisgenesis and genome editing approaches to improve health-promoting properties on citrus fruits.

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The new breeding techniques (NBTs), defined as cisgenesis and genome editing, represent an instrument to induce mutations on a specific genetic region without modify the rest of the genome and reproducing what could be happen naturally or through conventional crosses. Recently there is a huge interest on fruits and vegetables containing nutraceutical, antioxidant and healthy compounds, such as anthocyanin and lycopene. Naturally in Citrus the presence of both is not reported. The goal of the project consists in the use of cisgenesis and genome editing to obtain new citrus genotypes naturally containing, respectively, lycopene and anthocyanins. A preliminary and crucial step for using NBTs consists on the knowledge of candidate genes controlling the trait of interested. Also in citrus anthocyanin biosynthesis is regulated by a complex formed by a Myb-like transcription factor (FT), a basic helix-loop-helix FT and a WD40-repeat proteins. In our case Myb is represented by Ruby, nowadays the only candidate fruit-specific gene controlling the anthocyanin production in blood oranges; bHLH is Noemi gene; the WD repeat is not yet characterized. For the aim of the project we plan to use Ruby for cisgenic experiments. Conscious of practical difficulties strictly depending on the approach, we also hypothesized to edit genes working as competitors of the Myb specifically involved in the control of anthocyanins, as reported in apple, peach and citrus itself. The expression of a subset of candidate genes potentially involved in the negative regulation has been evaluated on eleven genotypes, including common and pigmented (blood for anthocyanins and red for lycopene) citrus varieties. At the meantime, we focused on the expression of a gene involved in the biosynthesis of lycopene (phytoene synthase), on two alleles (one functional and the other one non-functional) responsible of the degradation of lycopene (beta-cyclase) and on a regulatory gene (stay-green), to be used for a genome editing approach on blood citrus varieties. Moreover, preliminary GWAS analysis performed on sweet oranges mutants with and without lycopene showed the statistical association with a single based mutation potentially involved in accumulation of lycopene. These data will be verified by High Resolution Melting analysis to assess allelic status. The potential positive evidence will be used for a gene-editing approach. Finally, the optimization of regeneration protocols of varieties that will be used for cisgenesis, genome/gene editing are under evaluation. This step represents a huge challenge that have been solved for the successful use of the NBTs.

Session 4

Effect of innovative husbandry strategies on meat quality

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Meat and meat products represent an important source of protein in human diet, and their quality varies according to intrinsic and extrinsic parameters that can sometimes be shaped to make a product more desirable. This would allow the meat sector to better satisfy consumer expectations, demands and needs. The general aim of my PhD project is to investigate the effect of innovative husbandry strategies on meat quality, carrying out different analysis from molecular processes to consumer perception. In particular, lamb meat possesses such a peculiar flavor that it is often referred to as “sheep meat” or “sheepy” flavor. This typical odour has been attributed mainly to the presence of some branched chain FAs (4-methyl octanoic and 4-methyl nonanoic acids), skatole (3-methyl-indole) and indole that are compounds deriving from the degradation of tryptophan in the digestive tract. These compounds are found in the adipose tissues of animals and can be controlled through feeding strategies. Particularly, tannins are bioactive compounds able to interfere with protein and lipid metabolism in the rumen, by forming undegradable complexes with dietary proteins and by modulating several bacterial activities, including the biohydrogenation of polyunsaturated fatty acids and are claimed to mitigate the pastoral flavor of sheep meat. This study on dietary supplementation with different tannins is the first research contribution to my PhD project, another topic of which, regards the valorization of by-products as animal feeding. Indeed, food-processing industries produce a multitude of by-products still containing different valuable substances and that could be therefore suitable for animal feeding. Cardoon meal is a by- product obtained after the extraction of oil from cardoon seeds and has been proposed as animal feed because of its favorable chemical composition and phenolic content. In fact, it has high nutritive and energy value due to its valuable source of fibre, protein amino acids and bioactive compounds as polyphenols and unsatured fatty acids. Therefore, the aim of this part of my research was to evaluate the influence of dietary cardoon meal on selected quality parameters of lamb meat.

Investigating the effector-target interaction and its role in Apple Proliferation disease

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Apple Proliferation (AP) is a severe disease widespread in apple-growing areas in Europe. Typical symptoms comprise foliar reddening, shoot proliferation, small leaves with altered shape and undersized, tasteless and colourless fruits. The causal agent of the disease is the phytoplasma '*Candidatus* phytoplasma mali' ('Ca. P. mali'). Typically, phytoplasmas release proteins, called effectors, in the host cells altering the physiological processes and providing a fitness advantage to the bacteria. SAP11CaPM is the only bacterial effector known to date to be secreted by Apple Proliferation phytoplasma. In *Malus* the effector binds and deactivates two transcription factors (TF) of the TCP family, namely MdTCP24 and MdTCP25. To elucidate the role of SAP11CaPM and its targets in the context of resistance against Apple proliferation phytoplasma, the sequences of the target genes from susceptible and resistant *Malus* have been compared. Interestingly, some mutations exclusively present in AP resistant *Malus x domestica* TCP 25 could be observed. The sequences of many other AP-resistant plants were sequenced and a good degree of correlation was found between resistance and presence of the previously detected resistant exclusive mutations. Using a Yeast Two hybrid-based reporter gene assay, the interaction strength between SAP11CaPM and MdTCP25 from resistant plants was then evaluated. However, preliminary data show no visible differences in interaction strengths between SAP11CaPM and MdTCP25 from susceptible or resistant plants. Using Agrobacterium-mediated transformation, MdTCP24 and MdTCP25 are being overexpressed in the cultivar Gala. Two cisgenic lines have already been confirmed and other promising candidates are currently in the regeneration phase and will soon be ready for molecular analysis. Furthermore, in order to determine whether the presence of the effector actually affects the TFs-DNA binding capability and to identify the TFs DNA target regions, a functional assay will be set up by adapting a DNA affinity purification sequencing (DAP-seq) protocol, a transcription factor binding site discovery assay that couples affinity purified TFs with next-generation sequencing of a genomic DNA library. *In vitro* expressed TFs will be used as baits for the purification and sequencing of the DNA regions recognised by the two proteins in presence and absence of the effector, thus providing crucial information for understanding the molecular basis of the interaction and the biological pathways regulated by MdTCP24 and MdTCP25.

An alternative approach to assess the effects of clogging on the constructed wetland by testing the pollutants concentration removal efficiency

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Constructed Wetlands system (CWs) are infrastructure designed to promote wastewater (WW) treatment, simulating the processes taking place naturally in wetlands. CWs require a thorough understanding of hydraulic behaviour for their correct design and efficient operation. With time and as a result of their treatment activities, the beds that make up the constructed wetland system undergo a gradual clogging, due to the characteristics of the effluent, design errors, and bad management. The development of clogging can be detected by water appearance on the medium surface, and its effects could reduce the treatment efficiency, making maintenance operations necessary. Obstructive phenomena can sometimes be complex, such that an in-depth analysis of the whole system is necessary, aimed to revealing interactions between components of CWs and operative modalities. Estimate of the level of clogging, at this point, takes on considerable importance in the operation and management of CW. The suitability of available measurement techniques, in terms of accuracy, repeatability as well as time and skills required, can vary depending on the substrate type, the system design as well as clogging degree and its distribution. To investigate the space-time evolution of the clogging, a monitoring campaign has been started in the full scale plant, working as secondary WW treatment system of the Ikea®, located in the industrial district of Catania, Eastern Sicily, Italy. The research concerns an alternative approach to assess clogging effects on the CW removal efficiency by pollutants concentration (COD test) in different bed districts. With the aim to validate the proposed method, other hydraulic measurement techniques were tested. In particular tracer tests were carried out by pulse-injecting a sodium chloride solution into the inlet pipe of the hydraulic system and then measuring WW electric conductivity at different sites within the bed to assess clogging levels variation. As an alternative comparison test, a measure of hydraulic conductivity of the medium was used, namely the “falling head” test, developed specifically to detect the hydraulic conductivity at saturation (NAVFAC, 1986; Caselles-Osorio and García, 2006; Pedescoll et al., 2009).

Printing cereal dough: novel sensory perception of food from plant tissue microstructure

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3D Printing (3DP) is an emerging technology capable to convert a virtual 3D model in a physical tangible structure by a layer by layer deposition process of different materials. This technology belongs in Additive Manufacturing (AM) that is constantly extending his usage in many fields of research and industrial applications like medicine, architecture, industrial prototypes, electronic systems, jewellery and food manufacturing, as well. With regard to the food sector, the manufacturing of personalized/tailored food in terms of both nutritional and sensory properties is the main vision on which is based the great interest of researchers and industries on 3D food printing technology. This work aims to create novel sensory perceptions, mainly texture, by mimicking the 3D architecture of edible plant tissues. X-ray image of some fruit and vegetables (apple, pear, etc.) have been used to obtain a 3D model easy to be used in 3D printing process. Then, by using a commercial 3D printer, the most relevant printing variables (layer height, nozzle size, print speed, etc.) have been modulated to print thin cereal-based 3D structures inspired by microstructure of the plant tissues. Finally, after cooking in an ordinary oven, these new 3D food structures have been stacked in different number and order by combining the diversity in microstructure and thickness with the main goal to create multi-layered food with innovative sensory properties.

Optimization of agronomical and technological parameters of Ribolla Gialla for the production of quality sparkling wine

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Ribolla Gialla is an old grape variety mostly cultivated today in the Friuli-Venezia Giulia region in north-eastern Italy, Slovenia and Ionian Island where is known as Rebula and Robola respectively. Up to the early 1900s, Ribolla Gialla was widely cultivated throughout both today's DOC areas in the aforementioned region, the Colli Orientali del Friuli and the Collio, but often blended with other local varieties in the winemaking process. Nowadays the area cultivated with Ribolla Gialla is increasing, as well as its potential for the production of quality monovarietal sparkling wine, due to its naturally high acidity. The main research objectives of this project are directed towards viticulture as a principal research line. Therefore, the aim is to study how the geographical location of vineyards (especially hills versus flat areas) affects the ripening of the grapes, and on the other hand, to determine how agronomic procedures can be used in the vineyard, in order to obtain the best ratio between quality and quantity of grape. For instance, cluster thinning is a commonly adopted viticultural technique for selective removal of excessive clusters and allows calibrated vine productivity with increased accumulation of metabolites in the fruit. In order to identify those metabolites the metabolomic approach is applied, focusing on the characterization of volatile compounds, tryptophan derived metabolites and lipids at all stages of the sparkling wine production. For this purpose, analytical techniques such as one-dimensional gas chromatography and liquid chromatography, both coupled with mass spectrometry, are employed in this work.

Novel ingredients and underexploited feeds to improve the sustainability of farmed fish species.

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Aquaculture industry is the fastest growing food production industry world-wide. For a long time, fish meal (FM) and fish oil (FO) were the optimal raw materials in aquafeed for carnivorous fish species in intensive aquaculture, because of their protein content and their amino acid profile. The steady decline in catches of wild fish and the increased demands for livestock and aquaculture feeds have resulted in a rapid decrease in availability of FM and FO and their concurrent price increase, these limit the development of carnivorous fish farms. Over the past several years, intense focus has been trained upon the reduction or elimination of fish meal protein in aquafeeds, especially those designed for high-level carnivorous fish species, as Rainbow trout (*Oncorhynchus mykiss*), Gilthead seabream (*Sparus aurata*) and European seabass (*Dicentrarchus labrax*). This is due to the need to make aquaculture industry sustainable, in particular to contrast overfishing and high cost of fish meal and fish oil. Since 1990, these two raw materials have been replaced with other ones from terrestrial plants, but these ingredients are not sustainable as well because they compete for human and animal food. For this reason, new raw material should be found to be included in aquafeed formulation. The aim of this study is to characterize the nutritional value of novel ingredients and to evaluate the biological response of European seabass, Gilthead seabream and Rainbow trout to new feeds. Raw materials used in this study are Poultry by-product meals, Microalgae dried biomass (*Arthrospira platensis, Tetraselmis suecica and Tisochrysis lutea*) and insect meal (Hermetia illucens). The basic factors to establish the nutritional value of these novel ingredients are the evaluation of the nutrient composition and the nutrient and energy digestibility. The proximate, amino acid and fatty acid compositions of the test ingredients were determined with standard methods. Nutrient and energy apparent digestibility coefficients (ADCs) were estimated with the *in vivo* indirect method and by difference relative to those of a reference diet. Moreover, the extent of protein degradation and the amount of free amino acids released by proteolytic enzymes extracts obtained by fish were evaluated with the *in vitro* assay, used as preliminary alternative test to evaluate the quality of protein-rich ingredients for aquafeeds. Furthermore, first preliminarily data about fish biological responses to novel formulated diet were evaluated (feed intake, growth performances and feed conversion efficiency).

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Session 5

Antioxidant effect of natural extracts on bovine aortic endothelial cells

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Oxidative stress is implicated in a large number of animal diseases and it occurs when cellular production of reactive oxygen species (ROS) overcomes antioxidant capacity needed to prevent oxidative injury. Previous studies in humans demonstrated the positive effect of natural extracts enriched in polyphenols and tannins on mitigating oxidative stress. These positive results encourage the use of such natural antioxidant substances in dairy cattle to help managing oxidative stress. This preliminary study aims to test the antioxidant effect of pomegranate (PG), tara (TA), chestnut (CH) and gambier (GM) extracts on bovine aortic endothelial cells (BAEC). In particular, the effect of experimental extracts on cell viability, ROS production and apoptosis/death were performed. BAEC were isolated from bovine aorta tissue and used at passage 9. Intracellular ROS production and cell viability assays were carried out with a fluorescent and luminescent assay kits, whereas, apoptosis/death cells were determined by flow cytometry using a commercial Yo-PRO kit. The Pomegranate and Gambier extracts tested did not affect the viability of BAECs, though an increase of apoptosis/death has registered in Gambier extract. Furthermore, the extracts showed a greater capacity to decrease the ROS production, particularly in both pomegranate and gambier extracts, showing the lowest intracellular ROS production compared to the other extracts. The in vitro antioxidant activity of this extracts has been attributed to their high polyphenolic compounds. This preliminary study has shown the positive effects of the extracts tested on oxidative stress reduction. These data encouraged further experiments in order to better understand how these compounds can mitigate the metabolic stress and diseases related.

Determinants of frost resistance in fruit crops *Vitis vinifera*, *Prunus persica* and *Malus domestica*

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Crop production needs to adapt to the changing environment by using different genotypes or more resource-efficient cropping systems. Despite the increase of average temperatures, extreme climatic occurrences, such as spring frost, are becoming more frequent. Warm winters also affect buds' frost resistance, as the occurence of thaw during this season results in a loss of hardiness, increasing tree vulnerability to subsequent subfreezing temperatures and to spring frost. The levels of basal stress tolerance vary across species and genotypes, but plants can acquire a tolerance through previous exposure to non-lethal transient level of the same stress. This phenomenon, in which the cellular state of stress-exposed plants is different from that of non-stressed ones, is called priming. This stress memory functions within a generation in *Arabidopsis*, but remains largely unknown whether such a memory is transgenerational. Moreover, the changing environment requires an advancement of our understanding of the molecular and physiological mechanisms linking cold exposure to the expression of genes involved in cold resistance in main cultivated fruit crops in Italy and Europe such as *Vitis vinifera, Prunus persica* and *Malus domestica*. Whether epigenetic mechanisms come into play in these responses also needs investigation. Sugar content, amino acids and protein content will be evaluated during the progression of dormancy in buds of differently cold-resistant varieties of *V. vinifera, P. persica* and *M. domestica*. *Differential Thermal Analysis* (DTA) will be employed to evaluate the response of buds to different freezing temperatures, and to assess if non-lethal freezing conditions may induce a modification at transcriptional level of genes putatively involved in cold resistance mechanism. The study will proceed through the analysis of the epigenome of different varieties to evaluate whether stress-responses are conserved during propagation. An understanding of epigenetic changes underlying genotype/environment interactions could be useful for the improvement of fruit crop performance in unpredictable climates. Analysis of the distribution of epigenetic markers and DNA methylation in relation with gene expression profiles could identify epialleles as important new targets for plant breeding.

Heath and eco innovation in food packaging

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Technological innovations in food packaging area are focused to reduce two important environmental problems: plastic waste and food wasted and lost through the food supply chain. In 2015, worldwide 300 million ton of plastic waste was generated and nearly the 50% was plastic packaging. Moreover, according to FAO (2011) roughly one-third of the edible parts of food produced for human consumption, gets lost or wasted globally, which is about 1.3 billion ton per year. Then, such technological research leads to the development of active and intelligent packages, to improve food quality and safety level and to extend food products shelf-life, as well as the creation of sustainable packaging materials. Bioplastics, for example, are made from plants that are rich in carbohydrate, such as corn or sugar cane, or from ligno-cellulosic feedstocks (plants that are not eligible for food or feed production). Moreover, the technological innovation in this area is focused on the use of organic waste feedstocks, such as the use of food waste processing of the dairy industry. Then, there is one important advantage of biobased plastic products compared to conventional versions: they save fossil resources by using biomass which regenerates (annually). However, innovations in the food sector, including packaging materials, are consumer-driven. Understanding consumer acceptance and willingness to pay (WTP) may guide firms' investment towards those packaging innovations that are more likely to succeed in the market. Then, the objective of this research was, at the first, to summarize the evidence from studies investigating on consumer acceptance and WTP for food products packaged with innovative technologies (active and intelligent packaging, sustainable packaging) and, at the end, create an experimental design (choice experiment) to analyze the consumers' WTP for biodegradable bioplastic packaging for milk, in order to support food companies' investment decisions. The goal was to analyze if italian consumers are WTP a premium price for biodegradable packaging for milk in comparison with the traditional recyclable containers. Moreover, to understand if their WTP is the same for biodegradable bioplastic containers made from different raw materials, such as 100% plant based and 100% food losses and waste. Finally, the treatment information could give indications to food companies about how to drive the successful implementation of these innovations into the market.

A multidisciplinary approach for citrus rootstocks evaluation

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Horticulture is a key sector for the Mediterranean and Italian economy and in particular, citrus is the primary fruit crop in terms of monetary value. The annual worldwide citrus production reached approximately 147 million tonnes in 2017, and their cultivation is concentrated mainly in tropical, subtropical and Mediterranean regions. As well as several other crop species, citrus production is influenced by abiotic stresses such as drought, salinity, flooding and extreme temperature. For these reasons, all citrus species are grafted, and rootstock strongly affects the performance and production. Choosing an appropriate rootstock for a new citrus orchard has an enduring effect that lasts on the life of plantings. For this reason, it is important to select the most suitable rootstock for each growing area, taking into account variables as climate, soil, crop management, scion, pests, diseases, and fruit destination. The aim of the research is the evaluation of some recent and emerging rootstocks in comparison with others dominant and widely spread, in order to investigate their tolerance to water and salt stresses. The study will be carried out combining different approaches, in particular morphological, physiological, trancriptomics and metabolomics, to study the behaviour of the rootstock under both abiotic stresses. In addition, mycorrhiza's influence on physiological and vegetative performances will be investigated. The research will be carried out on different rootstocks, alone and in combination with Tarocco cultivar. Initially, several morphological and physiological assays will be analysed to evaluate the response to water and saline stress, both on rootstock and rootstock/scion combinations. These results will provide useful information in order to select 3 to 5 rootstocks that will be further investigated coupling transcriptomics and metabolomics approaches. The expected result is the indexing of the rootstock(s) showing the best performances under stress conditions and to identify potential candidate genes that (i) can better elucidate the physiological mechanism underlying abiotic stress responses and (ii) can be further employed in novel breeding programmes. Choice.

Investigation on *Cyclospora cayetanensis* in vegetables and imported soft fruits sold on Italian market by microscopy and standardized *q*PCR

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In the USA, large outbreaks of cyclosporiasis have increasingly been linked to various types of fresh produce imported from South-American countries. Despite some outbreaks of cyclosporiasis have been recorded in Germany and Sweden, *C. cayetanensis* has received much less attention as a foodborne pathogen in Europe compared to the U.S. In Italy, the high prevalence of *C. cayetanensis* DNA detected in water, soil, vegetables, and in humans indicate the need to investigate more in depth the public health significance of *Cyclospora* in this country. The aim of this study is to investigate the prevalence of *C. cayetanensis* as a contaminant of fresh produce ready to eat (RTE) mixed salads and berries- sold on the Italian market, using validated methodologies. e chose a pool size of 9 packages of fresh produce each month, for a total of 54 samples per month and 72 pools per one year. After collection, they were washed using the FDA washing procedure and the pellets subjected to molecular and microscopy analyes. Two real-time qPCR assays (18S ribosomal RNA gene), according to the BAM 19b (*Assay 1*) and, a new, specific qPCR assay (ITS1 region) (*Assay 2*), were set up and showed correct amplification from the internal amplification control as well as the positive control, with consistent Ct values across the replicates. So far, from January to April 2019, a total of 108 RTE mixed salad packages, belonging to three industrial brands, and 108 berries packages (blueberries and blackberries imported from Perù and Mexico, respectively, and raspberries grown in Italy) have been collected and washed. Twenty-four pools and a total of 216 aliquots (9 per pool) were obtained. Four samples (two from RTE mixed salads belonging to two different brands and two from raspberries and blueberries) were tested in duplicate in *q*PCR using *Assay 2* molecular analysis. Preliminary microscopy and molecular results will be presented. Once the study is completed, the molecular tools that have been developed cooperatively in different partner labs, will provide both the prevalence of *Cyclospora* in fresh produce in Italy and a shared methodological direction for wider monitoring of fresh produce at European level.

Detection and identification of insect compounds in fish diet

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Meat, fish, milk and eggs are the major protein sources for human consumption. The target of the animal protein sources production is directed toward the use of cost-effective raw materials, the increase of food safety, with a special focus on environmental sustainability using existing and novel protein sources and improving the actual production methods. In a scenario where novel and safe proteins availability is the target, the European Commission recently allowed the use of seven insects species as Processed Animal Proteins (PAPs) to be used in aquafeeds (Reg. 2017/893/EC, 2017). DNA detection and identification of animal materials by means of PCR, thanks to its high sensitivity, is actually considered the official analytical method to: (i) determine the species origin of PAPs; (ii) give evidence of the absence of prohibited ingredients; (iii) confirm the presence of ingredients in animals' feeds. Among the detection/identification of PAPs, insect species identification may represent a new challenge and PCR methods the solution but not the only one. Among the authorized insect species, *T. molitor* (TM) and H. illucens (HI) appear the most promising to be used in feeds according to availability and market price. The primary objective of the present study was to identify species specific PCR primers for the seven authorized insect species in EU while the secondary was to evaluate the sensitivity of the detection of *Hermetia illucens* (HI) and *Tenebrio molitor* (TM) in compound feeds using the PCR and compere this worldwide used method with a new biosensor under development.

Omics approach to explore microbial interaction of table olives consortium

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Table olives can be considered the widespread fermented food in the Mediterranean area and their consumption is expanding in all over the world. Thanks to their high fibre content, antioxidant vitamins, polyphenols compounds and short fatty acids, table olives can be considered a natural functional food, representing a good food matrix to carry active viable bacteria into the gastrointestinal tract. The presence of these compounds is strongly influenced by microbial consortium that is relevant for the development of biotechnological process. The aim of the PhD project is i) to determine the complexity, variability and functionality of microbial consortium during the table olives fermentation; ii) to understand specific mechanisms of proteomic adaptation involved in microbial performances, and iii) to study gene expression and metabolites generated from metabolic pathways, affected by abiotic factors, such as pH, temperature, salt content. Moreover, *in vivo* studies will be carried out to in depth understand the influence of microbial consortium on physiology of human host. For the achievement of the proposed goals multi-omics approaches, such as metagenomics, metatranscriptomics, metaproteomics and metabolomics, will be applied. The use of the culture-independent techniques will allow a higher sophisticated level of microbial and metabolic investigation to identify new biomarkers that could improve the quality of table olives and have beneficial effects on human health.

Functional characterization of resistance genes against Apple Scab

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Apple (*Malus X domestica*) is one of the most popular fruit and major useful food resource, belongs to the *Rosaceae* family. Apple scab is one of the most devastating apple disease, caused by Ventuira inaqualis which reduce fruit yield worldwide. The aim of my PhD Thesis is to manage apple scab by genetically engineered apple with a new resistance gene and study its function to develop durable apple scab resistance*. In vivo* experiments would be performed for the functional characterization of resistance genes Rvi 12 (vb). Currently, the BAC clone from *Malus baccata Hansens Baccata # 2* is used for cloning the Rvi 12 candidate gene with his native promotor. Over-expression of Rvi 12 in the susceptible apple plants are carried out by agrobacterium transformation. New plant breeding techniques such as cisgenisis and CRISPR/Cas9 technologies would be used to manipulate targeted genetic sequences directly. Newly transformed apple would be inoculated with *V. inaqualis* for phenotypic evaluation to check their susceptibility to apple scab. In parallel, the functional characterization of cisgenic plants expressing Rvi 6 (vf) or Rvi 15 (vr2) via Omic approaches (Transcriptomic and metabolomics) would be performed.

Physiology of stress response in grapevine

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In the framework of a more sustainable vineyard, an improved and more efficient irrigation management together with the decrease in the number of pesticide treatments per year represent necessary steps towards a more environmental friendly – low impact strategy in conventional, as well as in organic viticulture. In this scenario, the main goals of this project can be divided into two research lines. From one side, the project aims to improve comprehension about different hydraulic behaviour (namely isohydric and anisohydric) of grapevine cultivars and the role of carbon reserve during and after drought stress. Carbon reserve, also known as non-structural carbohydrates (NSC), besides their role in sustaining plant respiration when photosynthesis is limited, and the resumption of vegetative growth, could play a critical osmotic function, aiming to maintain turgor and long-distance vascular integrity. How water shortage affects NSC assimilation and storage, and what is their role in the physiology of cavitation refilling in Vitis vinifera, will be investigated. The other research line plans to test capability of inducing broad-spectrum resistance against pathogens in grapevine by plant extract biostimulant treatments. Grapevines, as well as other plants, respond to stress by activating a large array of mechanisms at different levels of complexity, ranging from molecular and cellular to anatomical and morphological ones. In particular, the research will focus on the study of the accumulation and/or modulation of pathogen-related (PR) proteins in grapevine plant and the inhibition of pathogenic fungal widespread, caused by bio-stimulant application.

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