



Research and Innovation in Agricultural, Livestock and Food Sciences

Campobasso, May, 9-12, 2022











# INTERNATIONAL PhD STUDENT CONFERENCE

Research and Innovation in Agricultural, Livestock and Food Sciences

> Campobasso, May, 9-12, 2022

Organized in the framework of

International PhD in Agriculture Technologies and Biotechnologies

&

EcoSET. Ecology, Science, Education and Technology. Ways to Internationalise the Bydgoszcz University of Science and Technology in Areas of Research and Education

**Book of Abstracts** 

Guest Editors: Prof. Giuseppe Maiorano Prof. Siria Tavaniello

# INTERNATIONAL PhD STUDENT CONFERENCE Research and Innovation in Agricultural, Livestock and Food Sciences

### ORGANIZING COMMITTEE

Giuseppe Maiorano (President)

Magdalena Kolenda

Maria Cristina Messia

Fabio Pilla

Andrea Sciarretta

Elena Sorrentino

Siria Tavaniello

Patrizio Tremonte

Marek Bednarczyk (President) Jakub Biesek Raffaello Castoria Mariasilvia D'Andrea Antonio De Cristofaro Giafranco Panfili Katarzyna Stadnicka

SCIENTIFIC COMMITTEE

Secretary Marisa Palazzo

# INTERNATIONAL PhD STUDENT CONFERENCE

Research and Innovation in Agricultural, Livestock and Food Sciences

Campobasso, May, 9 – 12, 2022

Venue University of Molise Department of Agricultural, Environmental and Food Sciences Room: F. Silvestri Via de Sanctis snc Campobasso

# Conference programme

# Monday 9<sup>th</sup> of May

Arrival of participants

T I doth Chd	
Tuesday 10 <sup>th</sup> of May	
09.00 - 9:30	Registration
09.30 - 10.00	Welcome and greetings
	<ul> <li>Prof. Luca Brunese – Rector of University of Molise, IT</li> <li>Prof. Elena Sorrentino – Head of Department of Agricultural, Environmental and Food Sciences, University of Molise, IT</li> <li>Prof. dr. hab. Marek Bednarczyk – ECOSET Project Manager</li> <li>Prof. Giuseppe Maiorano – PhD Coordinator, Department of Agricultural, Environmental and Food Sciences, University of Molise, IT</li> </ul>
10.00 – 10.30	Prof. Katarzyna Stadnicka – Programming the future in chicken egg- research and emerging applications. Prof. Nicolaia Iaffaldano – Recovery of S. macrostigma: Application of innovative techniques and participatory governance tools in rivers of Molise.
10.30 - 11.00	Coffee break
11.00 – 12.30	I Scientific session: Sustainable Plant Production and Protection Chairman: Prof. Antonio De Cristofaro, Prof. Raffello Castoria
14.30 – 15.45	II Scientific session: Welfare, Biotechnology and Quality of Animal Production Chairman: Prof. Siria Tavaniello, Dr. Jakub Biesek
16.30 – 18.00	III Scientific session: Welfare, Biotechnology and Quality of Animal Production Chairman: Prof. dr. hab. Marek Bednarczyk, Dr. Aleksandra Dunisławska

## Wednesday 11<sup>th</sup> of May

09.30 – 11.30	<i>IV Scientific session: Food Science, Technology and Biotechnology</i> Chairman: Prof. Maria Cristina Messia, Prof. Patrizio Tremonte
11.30 – 11.50	Coffee break
11.50 – 12.20	Poster Session
12.20 – 12.40	Closing ceremony Prof. Giuseppe Maiorano – PhD Coordinator, Department of Agricultural, Environmental and Food Sciences, University of Molise, IT

Thursday 12<sup>th</sup> of May

Organized trip in Molise

### Agenda for oral presentations

### Tuesday 10<sup>th</sup> May 2022

### *I Scientific session: Sustainable Plant Production and Protection* Chairman: Prof. Antonio De Cristofaro, Prof. Raffello Castoria

- 11.00–11.15 AMT-mediated insertional mutagenesis in Papiliotrema Page 1 terrestris PT22AV for assessing the role of extracellular polysaccharide (biofilm) production in biocontrol activity. Farhan Ali, Raffaello Castoria, Davide Palmieri, Giuseppe laniri
- 11.15–11.30 Transcriptomic approach to unveil the interaction between a biocontrol yeast and a postharvest fungal pathogen on the *"2 host fruit.*Giuseppe Barone, Davide Palmieri, Filippo De Curtis, Giuseppe Lima, Giuseppe Ianiri, Raffaello Castoria
- 11.30–11.45 Evaluation of essential oils bioactivity on honey bee and "3 Varroa mite.
   Dalila Di Criscio, Sonia Ganassi, Cosimo Tedino, Antonio De Cristofaro
- 11.45–12.00 First results of a microclimatic monitoring in a hydroponic greenhouse. " 4 Michela Orsino, Claudio Perone, Pasquale Catalano, Biagio Bianchi, Ferruccio Giametta, Giovanna La Fianza
- 12.00–12.15 The agrarian reform in Albania: history, impact and "5 implications. Arlinda Hoti, Angelo Belliggiano
- 12.15–12.30 Monitoring soil quality of urban ecosystems in Naples " 6 (Southern Italy).
   Pasquale Napoletano, Anna De Marco, Erika Di Iorio, Claudio Colombo

### *II Scientific session: Welfare, Biotechnology and Quality of Animal Production* Chairman: Prof. Siria Tavaniello, Dr. Jakub Biesek

- 14.30–14.45 Changes in hepatic gene expression and methylation upon in Page 7 ovo modulation of the intestinal microbiota with bioactive substances in chickens.
   Aleksandra Bełdowska, Elżbieta Pietrzak, Maria Siwek, Aleksandra Dunisławska
- 14.45–15.00 OVOBIOM: In vitro study on the new probiotics, prebiotic and synbiotic formulations to in ovo modulate the intestine health " 8 and organism performance.
  Niloofar Akhavan, Katarzyna Stadnicka, Katarzyna Hrynkiewicz
- 15.00–15.15 Methods of acquiring a larger number of chicken primordial germ cells.
   Agata Szczerba, Marek Bednarczyk, Takashi Kuwana
- 15.15–15.30 Mechanisms underlying the function of PGCs in "10 intergenerational and transgenerational epigenetic effects of in ovo chicken embryo stimulation.
   Mariam Ibrahim, Marek Bednarczyk, Ewa Grochowska, Katarzyna Stadnicka
- 15.30-15.45Growth and meat features of chickens fed with halloysite." 11Sebastian Wlaźlak, Jakub Biesek, Mirosław Banaszak

### III Scientific session: Welfare, Biotechnology and Quality of Animal Production Chairman: Prof. dr. hab. Marek Bednarczyk, Dr. Aleksandra Dunisławska

16.30–16.45 Genome-wide association study based on EBV ranking in Comisana sheep reveals loci involved in resistance to "12 gastrointestinal strongyles. Christian Persichilli, Stefano Biffani, Gabriele Senczuk, Marika Di Civita, Antonio Bosco, Silverio Grande, Fabio Pilla

- 16.45 17.00 Preliminary results of a machine learning approach to identify Page 13 informative markers in trout populations.
   Giovanna Salvatore, Valentino Palombo, Stefano Esposito, Nicolaia Iaffaldano, Mariasilvia D'Andrea
- 17.00–17.15 Effect of rearing systems on meat quality in Nero "14 d'Aspromonte pigs.
   Mengjun Wu, Siria Tavaniello, Sanije Zejnelhoxha, Meng Peng, Marisa Palazzo, Giuseppe Maiorano
- 17.15–17.30 Polycyclic aromatic hydrocarbons in charchoal-grilled beef "15 meat: influence of different concentrations of Vitamin E. Sanije Zejnelhoxha, Olga Viegas Siria Tavaniello, Edgar Pinto, Mengjun Wu, Marisa Palazzo, Isabel M.L.P.V.O Ferreira, Giuseppe Maiorano
- 17:30–17.45 Impact of probiotic dietary supplementation on the colony "16 growth and productivity of Apis mellifera L.
   Sonia Petrarca, Gianluca Albanese, Massimo Iorizzo, Antonio De Cristofaro
- 17:45–18.00 Semen cryopreservation for ex situ management of genetic "17 diversity in Mediterranean trout resulting in the creation of the first European cryobank.
  Emanuele Antenucci, Giusy Rusco, Letizia Lerza, Michele Di Iorio, Nicolaia Iaffaldano

### Wednesday 11<sup>th</sup> May 2022

### *IV Scientific session: Food Science, Technology and Biotechnology* Chairman: Prof. Maria Cristina Messia, Prof. Patrizio Tremonte

- 09.30–09.45 High amylose wheat flours for the development of healthy Page 18 cereal based foods. Martina Angelicola, Maria Cristina Messia, Emanuele Marconi
- 09.45-10.00 From durum wheat to whole-meal pasta: effects of processing "19 and cooking on physico-chemical and nutritional properties. Valentina Di Nardo, Emanuele Marconi, Elisa De Arcangelis

- 10.00–10.15 Combination of empirical and fundamental rheology for Page 20 dough characterization: an integrated approach. Silvio Iacovino, Francesca Cuomo, Emanuele Marconi
- 10.15-10.30Acrylamide in baked products: the case of Neapolitan pizza." 21Michela Quiquero, Emanuele Marconi, Maria Cristina Messia
- 10.30–10.45 Metagenomic characterization of microbial communities in "22 different Pecorino cheese from Campania Region. Diletta Bagnoli, Franca Vergalito, Mariantonietta Succi
- 10.45-11.00 Evaluation of functional properties from Lactiplantibacillus "23 plantarum strains isolated from bee bread. Francesco Letizia, Massimo Iorizzo
- 11.00–11.15 Antimicrobial edible films and coatings in microbiological "24 quality preservation of dried fruits.
   Ida Mercurio, Silvia Jane Lombardi, Patrizio Tremonte, Raffaele Coppola
- 11:15–11.30 Sample preparation, quantification and qualification of "25 microplastics in some food products. Cristina Di Fiore, Pasquale Avino

### **Poster Session**

Screening of Lactic Acid Bacteria for the Biocontrol of American and European foulbrood diseases. Gianluca Albanese

Perception and influence of climate change in the inner area alto e medio SANNIO (AMS). Sara Bispini

Post-domestication diffusion processes and adaptation in livestock and relationship with human populations by using SNPs data in a comparative framework. Marika Di Civita

The role of deuterated compounds in the determination of polycyclic aromatic hydrocarbons (PAHs) in environmental and agro-food matrices. Giuseppe Ianiri, Alessandra Fratianni, Gianfranco Panfili

Novel protective cultures for clean-label food products. Ilenia larusso

Edible coating of fresh-cut fruits by utilizing natural olive-oil by-products extracts as additive. Ayesha Iftikhar

Carbon forms and dynamic in mountain soils. Elettra Longobardi

*Effects of in ovo injection of synbiotics on growth, intestinal microbiota, short-chain fatty acids and meat quality of broiler chickens. Meng Peng* 

### 11:50 - 12.20

# PREFACE

### "EcoSET. Ecology, Science, Education and Technology. Ways to Internationalise the Bydgoszcz University of Science and Technology in Areas of Research and Education"

The project "EcoSET. Ecology, Science, Education and Technology. Ways to Internationalise the Bydgoszcz University of Science and Technology in Areas of Research and Education" (hereinafter referred to as "EcoSET") is carried out between 1 October 2019 and 30 September 2022 by three faculties of Bydgoszcz University of Science and Technology (hereinafter referred to as "PBS"): Faculty of Animal Breeding and Biology (hereinafter referred to as "WBAIIS"), Faculty of Civil and Environmental Engineering and Architecture (hereinafter referred to as "WBAIIS"), and Faculty of Design ("WSP") within the International Academic Partnerships Programme of the Polish National Agency for Academic Exchange ("NAWA"). The aim of the project is to increase the level of internationalisation in research and education at PBS in Bydgoszcz through EcoSET tasks that include *inter alia*: organisation of scientific visits, subsidising common scientific publications and organization of the closing conference that will be held in Bydgoszcz on September 12-14, 2022. The scope of the project includes shared research activities in the following disciplines: animal science and fisheries (at WHiBZ), civil engineering and transport as well as architecture and urban planning (at WBAIIS) and fine arts and art conservation (at WSP). EcoSET is carried out in cooperation with the following Partners:

- University of Molise, Italy
- Tarleton State University, the United States of America
- Aarhus University, Denmark
- Justus-Liebig-Universität Gießen, Germany
- Mendel University in Brno, Czech Republic
- National Centre for Biodiversity and Gene Conservation, Hungary
- Ondokuz Mayıs University, Turkey
- University of Maryland, the United States of America
- Adana Science and Technology University, Turkey
- Polytechnic University of Bari, Italy
- University of Beira Interior, Portugal

The Project has a dedicated website: <u>www.ecoset.utp.edu.pl</u> and e-mail address: <u>ecoset@pbs.edu.pl</u>.

The Project is coordinated by PBS, represented by Professor Marek Bednarczyk authorised by the PBS Rector. For the purposes of the Project a Project Management Team (PMT) is appointed at PBS. The PMT consists of:

- Project Manager / WHiBZ Coordinator: Professor Marek Bednarczyk
- WHiBZ Administrative and Financial Specialist: Magdalena Kolenda, PhD Eng.
- WBAilŚ Coordinator: Magdalena Dobiszewska, PhD Eng.
- Administrative and Financial Specialist: Agata Bukaluk, MSc Eng.
- and Partners' representatives who are PMT honorary members:
  - University of Molise: Prof. Giuseppe Maiorano, Head of Doctoral Studies at Department of Agricultural, Environmental and Food Sciences,
  - Tarleton State University: Dr Barry Lambert, Associate Vice President, Research and Innovation Dean, College of Graduate Studies,
  - Aarhus University: Margrethe Therkildsen, Associate Professor,
  - Justus-Liebig-Universität Gießen: Prof. Gesine Lühken,
  - Mendel University in Brno: Prof. Dr Eng. Petr Sláma,
  - National Centre for Biodiversity and Gene Conservation: Dr Eszter Patakiné Várkonyi,
  - Ondokuz Mayıs University: Prof. Dr Hasan Ónder,
  - University of Maryland: Prof. Dr Mirosław Skibniewski,
  - Adana Science and Technology University: Dr Ahmet Beycioğlu, Vice Dean of Engineering Faculty,
  - Polytechnic University of Bari: Prof. Andrea Petrella, DICATECh dept.,
  - University of Beira Interior: Dr Pedro Serrão, Science and Technology Manager.

### International PhD Programme in Agriculture Technologies and Biotechnologies

In the framework of the cooperation between Italy, Poland and Slovakia and on the basis of the internationalization process of the University education system, a three-year International PhD Program is activated in Agriculture Technologies and Biotechnologies (ATB). The Program will be organised between the following partners:

- Department of Agricultural, Environmental and Food Sciences, University of Molise;
- Faculty of Animal Breeding and Biology, University of Science and Technology in Bydgoszcz, Poland;
- Faculty of Biotechnology and Food Science, Slovak University of Agriculture in Nitra;
- Faculty of Agrobiology and Food Resources, Slovak University of Agriculture in Nitra.

The PhD program is focused on the agri-food and environmental systems as encoded in Horizon 2020. Its objectives are sustainable agriculture, food safety and quality, animal welfare, green economy and environmental law.

The PhD program is organized into 3 curricula: Sustainable Plant Production and Protection; Food Science, Technology and Biotechnology; Welfare, Biotechnology and Quality of Animal Production.

The PhD program aims at providing specific skills and expertise for: the development of sustainable agriculture ensuring quality and safety of food from plants and livestock, food security, soil and environment protection, through innovative biotechnical and biotechnological means; the development, evaluation and monitoring of research and innovation projects of the agri-food sector. The educational goals of the program are designed to address the needs of fundamental and applied research, with particular emphasis on the latter. The Doctoral program is based on courses, seminars and workshops also including soft skills (management of funds, financial and human resources; information management; planning capacity; fund raising; conceiving and preparing research and innovation projects; problem solving).

The Curriculum Sustainable Plant Production and Protection aims at providing skills and expertise for developing new technologies for the sustainable exploitation of agricultural resources, for the sustainable management of forest and agricultural ecosystems, and for the protection of biodiversity by reducing chemical inputs in agricultural ecosystems (Horizon 2020).

The Curriculum Food Science, Technology and Biotechnology aims to train a professional figure able to develop and apply new scientific knowledge in the field of food processing, conservation and evaluation and to promote research and innovation activities in the sector of food production processes. foods.

The Curriculum Welfare, Biotechnology and Quality of Animal Production aims at providing skills and expertise ensuring the capability to compete in the area of animal production, and to develop biotechnological and technological innovation for optimizing animal production, health and welfare and to upgrade the products of animal origin. For these purposes, issues regarding economy, laws and regulations, sustainability, food safety and security are also taken into consideration.

PhD General Coordinator: Prof. Giuseppe Maiorano

Local Coordinator for the University of Science and Technology in Bydgoszcz, Poland: Prof. Marek Bednarczyk Local Coordinator for the Slovak University of Agriculture in Nitra: Prof. Adriana Kolesarova

# AMT-mediated insertional mutagenesis in *Papiliotrema terrestris* PT22AV for assessing the role of extracellular polysaccharide (biofilm) production in biocontrol activity

Farhan Ali\*, Raffaello Castoria, D. Palmieri, Giuseppe Ianiri

Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis snc, 86100 Campobasso, Italy \*Corresponding author: f.ali@studenti.unimol.it@unimol.it

Papiliotrema terrestris PT22AV is an antagonistic yeast, biocontrol agent (BCA) of postharvest pathogens. Studies at a molecular level are essential to provide clues on its genetic mechanisms of biocontrol. This study aims to elucidate the molecular bases of extracellular polysaccharides (biofilm) production through a forward genetics approach to assess their role(s) in the activity of this BCA. To this aim, random insertional mutagenesis through Agrobacterium tumefaciensmediated transformation (AMT) was carried out. About 700 transformants of PT22AV were generated and tested in different nitrogen sources (sodium nitrate, ammonia, ammonium sulfate, ammonium nitrate, urea, apple medium) for their ability to produce different amounts of biofilm in comparison with the parental wild type (WT) of the same strain. Fourteen transformants displayed a different ability, with respect to the WT strain, to produce extracellular polysaccharides in the presence of different nitrogen sources, with apple medium allowing the isolation of the highest number of mutant phenotypes. Moreover, the selected transformants were tested in comparison with PT22AV WT to assess the relationship between nitrogen utilization, biofilm production, and biocontrol activity on apples challenged with Penicillium expansum. While these experiments are in progress, two transformants displayed a different biocontrol activity from WT, suggesting a putative role of biofilm in the biocontrol activity of PT22AV. Future experiments aim to identify the mutated gene(s) in the selected transformants and to carry out their functional and phenotypic complementation. This study confirms the potential of AMT in the forward genetics approach to unveil the genetic bases of biocontrol activity in Papiliotrema terrestris.

# Transcriptomic approach to unveil the interaction between a biocontrol yeast and a postharvest fungal pathogen on the host fruit

Giuseppe Barone\*, Davide Palmieri, Filippo De Curtis, Giuseppe Lima, Giuseppe Ianiri, Raffaello Castoria

Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis snc, 86100 Campobasso, Italy \*Corresponding author: g.barone2@studenti.unimol.it

Biocontrol strategies are a promising alternative to achieve food safety and food security. The aim of this study was to decipher the molecular interactions involving the biocontrol yeast (BCY) Papiliotrema terrestris strain LS28, the postharvest pathogen Penicillium expansum, and Malus domestica. RNAseq analysis was performed during both their dual and tritrophic interactions to identify the differentially expressed genes of BCY, pathogen, and host. Analysis of transcriptome changes in the BCY revealed that, regardless of the presence of the pathogen, overexpression of genes involved in nitrogen catabolite repression and oxidative stress response took place, suggesting that these pathways are crucial for the BCY to rapidly colonize the ecological niche (fruit wounds) and outcompete the pathogen. In the absence of P. expansum, BCY genes involved in metabolism and transport of carbohydrates and branched-chain amino acids were highly represented, suggesting a different nutritional requirement of P. terrestris when it does not compete with the pathogen. To confirm transcriptomic data at phenotypic level, targeted mutants of the BCY were generated for several overexpressed genes. The in vitro phenotypic characterization and biocontrol assay revealed a crucial role of a putative aminoacid transporter in the biocontrol activity of P. terrestris against P. expansum. The transcriptomic analysis of P. expansum revealed that genes involved in transcription, oxido-reductive processes, transmembrane transport, and amine and peptide metabolism were the most represented GO categories, regardless of the presence of the BCY. In the presence of the BCY transcripts involved in metabolic processes of polysaccharides, aminoglycan and glucosamine-containing compounds were strongly enriched, suggesting a substantial nutritional rewiring of the pathogen to directly outcompete the BCY. Analysis of the transcriptomic changes of the host *M. domestica* revealed overexpression of genes involved in host defence signalling pathways in the presence of both the BCY and the pathogen, and a prevalence of pathogen triggered immunity (PTI) and effector trigger immunity (ETI) host genes overexpressed only during interaction with P. expansum. This comprehensive analysis contributed to advance the knowledge on the molecular mechanisms that underlie biocontrol activity and the tritrophic interaction of the BCY with the pathogen and the host.

### Evaluation of essential oils bioactivity on honey bee and Varroa mite

Dalila Di Criscio\*, Sonia Ganassi, Cosimo Tedino, Antonio De Cristofaro

Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis snc, 86100 Campobasso, Italy \*Corresponding author: d.dicriscio@unimol.it

Varroa destructor mite is the main parasite of Apis mellifera and, if not controlled, it leads the colony to collapse in a short time. Over the years, many substances have been used against V. destructor, however none of them are able to control its populations effectively and without contraindications. The aim of the work was the identification of new and more targeted strategies to contain the development of V. destructor populations within the hive. In two years of experimentation, laboratory and field tests have been carried out on V. destructor and on larvae and adults of A. m. ligustica to test the efficacy and toxicity of some essential oils (EOs). The results were compared with standard control tools used in hives (oxalic acid). The tests carried out were: EAG test on adult bees, in vitro tests on A. m. ligustica larvae and on V. destructor adults and field tests. The results revealed that the volatile compounds present in all the EOs tested were able to stimulate the olfactory system of the foragers of A. m. ligustica and that the intensity of the response increases as the concentration of the stimulus itself increases. Furthermore, no difference in the perception of the left and right antenna was detected. Among the tested oils, those of geranium, lavender and cajeput elicited significantly greater responses in both antennas, at almost all concentrations, compared to those induced by the other oils. In vitro tests have shown a lethal effect of the three oils on the mite, thus providing promising results even if compared with those obtained with the use of OA. This study has also showed that the same oil had no lethal effects on the larvae of A. m. ligustica. From the results obtained in the previous tests, the three selected oils were tested directly in the hive, where they confirmed their effectiveness, depending on the concentration, against V. destructor as the number of dead individuals on the anti-varroa background was comparable to that obtained by using sublimated OA. On the contrary, the oil had no lethal effects on adult bees.

### First results of a microclimatic monitoring in a hydroponic greenhouse

Michela Orsino<sup>1\*</sup>, Claudio Perone<sup>2</sup>, Pasquale Catalano<sup>1</sup>, Biagio Bianchi<sup>3</sup>, Ferruccio Giametta<sup>1</sup>, Giovanna La Fianza<sup>1</sup>

<sup>1</sup>Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis snc, 86100 Campobasso, Italy

<sup>2</sup>Department of Agriculture, Food, Natural Resources and Engineering (DAFNE), University of Foggia, via Napoli 25 - 71122 Foggia, Italy

<sup>3</sup>Department of Agricultural and Environmental Science (DISAAT), University of Bari Aldo Moro, via Amendola 165/A- 70126 Bari, Italy

\*Corresponding author: m.orsino2@studenti.unimol.it

The rising demand for food is closely linked to the increasingly exponential demographic growth. To satisfy this request it is essential considering the aspects of sustainability focusing in particular on resource usage. In this context, soilless cultivation represents an interesting solution. In recent years hydroponic systems are spreading in greenhouses and their resources management must be efficiently optimized. The aim of this work is the microclimatic monitoring and analysis in an operating 500 m3 hydroponic greenhouse. The studied greenhouse is in Molise region (Italy) and, actually, is managed like a traditional greenhouse. The lack of the main advanced managing aspects of a greenhouse like mechanical ventilation, artificial lights, sensors etc., involves problems in the handling of this soilless cultivation system. For this reason, a preliminary characterization of the main microclimatic parameter was done. A set of temperature, relative humidity, light, and CO2 sensors were installed to monitor the microclimate in different selected zones in the cultivation environment. These were placed close to the main openings of the greenhouse and close to the selected leafy crops for their growing phase monitoring. In addition, the main agronomic parameters were observed during the microclimatic data acquisition. A supervision system allowed to historicize all measured data. The preliminary results showed the need to operate firstly on ventilation. This depends especially on the variability of the external microclimatic conditions in which the greenhouse is located. Starting from the improved management of the microclimatic parameters, it will be possible to maximize resources efficiency, crops growing and food quality bringing this soilless greenhouse into a basic automatization level.

### The agrarian reform in Albania: history, impact and implications

Arlinda Hoti\*, Angelo Belliggiano

Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis snc, 86100 Campobasso, Italy \*Corresponding author: a.hoti@studenti.uniomol.it

When Albania was restored as an independent state a hundred years ago, one of its political priorities was to implement an agricultural land reform, as the "agrarian issue" was an extremely sensitive socio-economic problem. Immediately after World War II, Albania held extreme choices and undertook radical agrarian reforms, first inspired by communist ideology and then, after 1990, by liberalism. Meanwhile another more limited reform had already been carried out in the early 20th century. This article aims to show that the agrarian reforms that have been carried out in Albania during the last century, instead of achieving their goal, namely improving the situation of the rural population and ensuring social peace, have brought adverse effects such as: collectivization, nationalization of large farms, fragmentation of agricultural land and farms, increasing legal ownership disputes over agricultural land, and reducing land use efficiency. Despite the different goals of the three agrarian reforms in Albania, they contributed to the same result of deepened the socio-economic division and differentiation among farmers and rural population, thus failing at reaching the overarching final aim of improving social-economic wellbeing of the Albanian farmers. The article is a comparative analysis of the three agrarian reforms adopted in Albania during the last century, referring to the ideology that inspired them and evaluating their results, using a methodology based on the combination of literature review, comparative analysis and data analysis. In conclusion, we try to draw lessons from these agrarian reforms and provide recommendations to influence agricultural policies in Albania but also in other countries with similar background.

### Monitoring soil quality of urban ecosystems in Naples (Southern Italy)

Pasquale Napoletano<sup>1\*</sup>, Anna De Marco<sup>2</sup>, Erika Di Iorio<sup>1</sup>, Claudio Colombo<sup>1</sup>

<sup>1</sup>Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis snc, 86100 Campobasso, Italy

<sup>2</sup>Department of Pharmacy, University of Naples Federico II, Naples, Italy \*Corresponding author: p.napoletano@studenti.unimol.it

Soil quality is related to its capacity to provide a wide range of ecosystem services that are crucial for earth system function and stability, e.g. Carbon (C) storage, plant growth promotion, watersheds protection by regulating infiltration and precipitation, water and air pollution prevention by buffering pollutants. Climate change causes alterations to land and resource management, social organization, infrastructures, and urban soils. Therefore, monitoring soil quality in urban ecosystems is pivotal for responding to climate change pressure in large cities. The high sensitivity of soil microbial communities to external disturbances makes it possible to use them as environmental indicators of climate adaptation of plants and animals. The strong relationship between microbial activity and C dynamics is also useful to shed light on soil biological processes occurring in urban ecosystems. The rapid rate of projected climate change, especially in cities, implies that many plant and animal species may not be able to adapt to new conditions. Therefore, the aim of this research was to monitor soil quality over eight years (since 2010) in some urban soils classified as Technosols for the high amount (≥ 20%) of anthropogenic materials mixed to native soils, and located in a highly populated area of Campi Flegrei (Fuorigrotta neighbourhood, Naples, Southern Italy), by using biological indices strictly related to C cycle. In this view, the equilibrium of microbial biomass on organic Carbon (qCmic), metabolic stress (qCO2), mineralization rate (CEM), the ratio of fungal C on total microbial C (FB/MB) were calculated along with soil organic matter (SOM) and C/N ratio. In eight years of monitoring, the investigated Technosols showed no statistically significant changes in qCmic and qCO2. The decrease of FB/MB ratio suggest that microbial structure varied by fostering bacteria more than fungi. The change of biological structure affected the use of C resources: CEM almost doubled as SOM and C/N significantly decreased during the experimentation. This study highlighted an intense microbial activity that affected resources utilization. These findings shed light on the high variability of biological indices that alone are not informative enough in similar studies. They can be used as part of more complex indices, by integrating also chemical and physical features for assessing soil quality variation over time.

# Changes in hepatic gene expression and methylation upon in ovo modulation of the intestinal microbiota with bioactive substances in chickens

Aleksandra Bełdowska\*, Elżbieta Pietrzak, Maria Siwek, Aleksandra Dunisławska

Department of Animal Biotechnology and Genetics, Faculty of Animal Breeding and Biology, PBS Bydgoszcz University of Science and Technology, Mazowiecka 28, 85-084 Bydgoszcz, Poland \*Corresponding author: alebel004@pbs.edu.pl

Literature reports suggest that the gut microbiota plays a key role in signaling at the level of the gut-hepatic axis. Intestinal microbiota produces bioactive peptides, neurotransmitters, shortchain fatty acids, hormones, and amino acids which affect internal communication. Changes in this communication can lead to modifications in gene expression driven by epigenetic mechanisms (induced by environmental signals). This study aimed to determine the impact of the early microbiota modification by bioactive substances delivered in ovo on changes in the relative abundance of bacteria in intestinal content and the level of hepatic gene expression and methylation. On day 12 of incubation, eggs of Ross 308 chicken broilers and Green-legged patridgelike were injected with probiotic- Lactococcus lactis, prebiotic- galactooligosacharide, and synbiotic- the combination of both. Tissues for RNA and DNA isolation and cecal content for bacterial DNA isolation were collected post-mortem on day 42 of rearing. The analysis of the relative abundance of Bifidobacterium spp., Lactobacillus spp., Clostridium difficile, and Escherichia coli was performed using the qPCR reaction. Gene expression analysis was performed by RT-qPCR, while gene methylation was determined by MS-qPCR method for the following genes: KLHL6, SYK, ANGPTL4, NR4A3, CYR61. The number of analyzed bacteria has changed in both genotypes. There is a significant increase in the number of Lactobacillus and Bifidobacterium bacteria, and a decrease in the number of Escherichia coli in Green-legged patridgelike. Clostridium difficile was not detected in both genotypes. A significant increase in DNA methylation of the SYK gene was demonstrated in both genotypes in the experimental group which has received prebiotic in ovo, and of the NR4A3 gene in broiler chickens after administration of the synbiotic. In the Green-legged patridgelike, the probiotic and the synbiotic reduced the methylation of the SYK gene. Changes in gene methylation are correlated with changes in gene expression. Early stimulation of the gut microbiota leads to changes in the gut microbiological profile, thereby responding to the expression and methylation of hepatic genes. This study confirms the epigenetic character of gene expression related to early microbiota programming.

### OVOBIOM: In vitro study on the new probiotics, prebiotic and synbiotic formulations to in ovo modulate the intestine health and organism performance

Niloofar Akhavan<sup>1\*</sup>, Katarzyna Stadnicka<sup>1</sup>, Katarzyna Hrynkiewicz<sup>2</sup>

<sup>1</sup>Department of Animal Biotechnology and Genetics, Faculty of Animal Breeding and Biology, PBS Bydgoszcz University of Science and Technology, 85-084 Bydgoszcz, Poland
<sup>2</sup>Department of Microbiology, Faculty of Biological and Veterinary Sciences, Nicolaus Copernicus University in Toruń
\*Corresponding author: nilakh000@pbs.edu.pl

Healthy gut supports the animal to combat the stresses associated with the intensive production. In poultry, an optimal microbiota and microbiome can be stimulated *in ovo*, with probiotics and prebiotics. We have aimed to select the most potent prebiotics and probiotics that will guarantee an immunomodulatory effect and changes in microbiome post-hatch. Several probiotic bacteria and various prebiotic compounds were tested to optimize candidate synbiotic combinations for *in ovo* application. *Lactobacillus, Bifidobacterium lactis, Carnobacterium divergens, Propionibacterium thoenii* and *Clostridium butyricum* were grown separately in vitro, each in media with addition of the prebiotic (2%), for 24h. The growth rate of probiotics was measured every 2h (optical density at 600nm) using a microtiter plate reader (SpectraMax® ID3 Multi-Mode Microplate Reader by Molecular Devices®). *Lactobacilli* species revealed growth improvements with the selected prebiotic compounds and may be candidates for subsequent *in ovo* trials. The post-hatch results will show, as to how the synbiotic compositions optimized in this study, will affect in ovo development and early colonization of the embryonic intestines. Metabolomic analysis will allow an understanding of the function of the selected synbiotic combinations.

### Acknowledgements

The research is financed under National Science Centre UMO-2019/35/B/NZ9/03186.

### Methods of acquiring a larger number of chicken primordial germ cells

AgataSzczerba<sup>1\*</sup>, Marek Bednarczyk<sup>1</sup>, Takashi Kuwana<sup>2</sup>

<sup>1</sup>Department of Animal Biotechnology and Genetics, Faculty of Animal Breeding and Biology, PBS Bydgoszcz University of Science and Technology, Mazowiecka 28, 85-084 Bydgoszcz, Poland <sup>2</sup>Shiga University of Medical Science, Setatsukinowacho, Shiga 520-2192, Japan \* Corresponding author: agata.szczerba@pbs.edu.pl

In experimental biology, the chicken is one of the model organisms which is used in basic and applied research. Primordial Germ Cells (PGCs), which are precursors of gametes, deserve special attention. They are the only ones capable of transmitting genetic information from generation to generation. After the formation of the vascular system, they circulate in the bloodstream to begin the colonization process in the gonads as the embryo grows. With this in mind, they have become an important trend in the search for new reproductive technologies. These pluripotent cells offer many theoretical possibilities; however, their use is difficult due to their limited number. Therefore, the aim of this study was to attempt to conduct a cell culture and to evaluate the proliferative properties of cells that would significantly resolve this issue. On the basis of the conducted experiments, it was found that the mean blood volume was 8.1, 20.8, 11.4, and 17.5 µl in stages 13, 14, 15, and 16 of Hamburger and Hamilton (HH), respectively. The number of PGCs in 1  $\mu$ l of embryonic blood was determined in stages 13 to 16 of HH. The maximum concentration of circulating PGCs (cPGCs) per 1 µl of embryonic blood was observed at stage 14 HH. The total number of cPGCs in embryonic blood at each embryonic stage was calculated from the number of PGCs present per  $\mu$ l. The highest number of cPGCs in the entire embryonic bloodstream was obtained at stage 14 of HH. Cell culture was carried out in several different variants. 18 HH stage gonadal primordial germ cells and 14-15 HH stage cPGCs were obtained. The gonadal and migrating cultured primordial germ cells were stained with PKH26 and then injected into the bloodstream of recipient embryos at stages 13-16 HH to test the migratory capacity of in vitro cultured PGCs. Then they were cultivated for 7 days. gPGCs multiplied but more slowly as the doubling period was 5 days. After 7 days of in vitro maintenance, three cells were obtained from one cell. cPGCs also proliferated, however, their fold was much higher compared to gPGCs. The doubling period was approx. 2 days. The conducted cell culture and the evaluation of the functional properties of the proliferated cells proved that in vivo and/or in vitro manipulations increase the availability of birds' primary germ cells in birds.

# Mechanisms underlying the function of PGCs in intergenerational and transgenerational epigenetic effects of *in ovo* chicken embryo stimulation

Mariam Ibrahim\*, Marek Bednarczyk, Ewa Grochowska, Katarzyna Stadnicka

Department of Animal Biotechnology and Genetics, Faculty of Animal Breeding and Biology, PBS Bydgoszcz University of Science and Technology, 85-084 Bydgoszcz, Poland \*Corresponding author: miriam.ibrahim@pbs.edu.pl

The *in ovo* injection of bioactive compounds including probiotics, prebiotics, or synbiotics can confer health benefits to the chicken host by boosting the host immune system through gut microbiome modification, and fine-tuning gene expression during early stages of development, that may render the animal more resistant to diseases. It is hypothesized, that the epigenetic factors: synbiotic or choline applied in ovo on 12ED, would change the epigenome and that primordial germ cells (PGCs) are specifically involved in passing down those epigenetic changes through the generations. The main aims of this study are to (i) comprehend the role of primordial germ cells (PGCs) in the mechanisms of epigenetic changes and their inheritance, and (ii) analyse the molecular pathways underlying this sort of transfer. To achieve these aims, the experimental chickens were divided into three groups: synbiotic, choline, and control. For each group, the screening study on PGCs from the embryos in 3 generation lines that were treated in ovo, will be performed. Two types of PGCs are being isolated and purified: the circulating PGCs will be isolated from embryonic blood at 14-15 H&H development stage and the gonadal PGCs will be isolated from the embryos at 29-31 H&H stage. The isolated PGCs will be characterized in vitro and subject to epigenomic study with total cellular RNA-sequencing and DNA methylation. It is expected to provide new evidence for the functions of the precursors of gametes in offspring reprogramming in the course of in ovo epigenetic impact.

#### Acknowledgements

The research is financed under National Science Centre 2020/37/B/NZ9/00497

### Growth and meat features of chickens fed with halloysite

Sebastian Wlaźlak\*, Jakub Biesek, Mirosław Banaszak

Department of Animal Breeding and Nutrition, Faculty of Animal Breeding and Biology, PBS Bydgoszcz University of Science and Technology, Mazowiecka 28, 85-084 Bydgoszcz, Poland \*Corresponding author: sebastian.wlazlak@pbs.edu.pl

The aluminosilicates are natural minerals that have absorptive and stimulating properties. It also has the potential as an alternative to antibiotics in poultry production. Halloysite is obtained in Polish mines. The aim of the research was to evaluate the production results as well as the slaughter yield and the quality of pectoral muscle of broiler chickens fed with the addition of halloysite in the feed. 500 Ross 308 chickens were kept for 42 days. The birds were divided into two groups. The control group (1) was kept without the addition of halloysite in 10 replicates of 10 birds. 400 chickens (40 replicates of 10 birds each) were kept in the experimental group (2). The nutrition in group 2 was based on complete feed with the addition of halloysite at the level of 0.5% (starter and grower), 1.5% (finisher). The birds were weighed (BW) on days 1, 14, 22, and 42. Feed intake (FI) was recorded. Body weight gain (BWG) and feed conversion ratio (FCR) were calculated. After rearing, 10 birds from the control group and 40 from the experimental group were selected and slaughtered. The carcasses were weighed. The pectoral muscles (including m. pectoralis major and minor), leg muscles (thigh and drumstick), skin with subcutaneous fat, and abdominal fat were cut out. The pectoral muscles were analyzed qualitatively. The pH 24 hours after slaughter, the drip loss, and the water holding capacity were analyzed. The color was assessed, taking into account the parameters: L \* - lightness, a \* - redness, and b \* - yellowness. The mean values and the standard error of the mean (SEM) were calculated. Statistically significant differences between the groups were calculated using the Student's t-test, assuming a value of p <0.05. The research showed that the addition of halloysite to the feed at the level of 0.5% resulted in a lower BW of broiler chickens on the 14th (p=0.020) and 22nd (p=0.026) day and BWG during the feeding of chickens with starter feed (p=0.017). In the days from 14 to 21, FI and FCR in group 2 were lower (p<0.001; p=0.001, respectively), and in the entire rearing period (p=0.025, p=0.020, respectively). In group 2, a higher percentage of pectoral muscle was found (p=0.019). No significant differences were found in the rest of the characteristics (p>0.05). The use of halloysite for a feed at the level of 0.5 to 1.5% had a positive effect on production indicators, including FI and FCR, as well as the share of the pectoral muscle, and no negative impact on the quality of meat.

#### Acknowledgements

This research was funded by the project "Safe Farm – innovative products, processes and marketing in the production of broiler chickens", implemented in 2020-2022 and co-financed by the European Agricultural Fund for Rural Development: Europe investing in rural areas". www.bezpiecznaferma.pl

# Genome-wide association study based on EBV ranking in Comisana sheep reveals loci involved in resistance to gastrointestinal strongyles

Christian Persichilli<sup>1</sup>\*, Stefano Biffani<sup>2</sup>, Gabriele Senczuk<sup>1</sup>, Marika Di Civita<sup>1</sup>, Antonio Bosco<sup>3</sup>, Silverio Grande<sup>4</sup>, Fabio Pilla<sup>1</sup>

<sup>1</sup>Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis snc, 86100 Campobasso, Italy

<sup>2</sup>National Council of Research, Institute for Agriculture Biology and Biotechnology, Milan, Italy <sup>3</sup>Department of Veterinary Medicine and Animal Productions, University of Naples Federico II, CREMOPAR, Naples, Italy

> <sup>4</sup>National Sheep and Goat Breeders Association, Rome, Italy \*Corresponding author: c.persichilli@studenti.unimol.it

Gastro Intestinal Nematodes (GIN) represent not only one of the most incisive costs on sheep farming but also a threatening to animal welfare in extensive small ruminant farming. Furthermore, the use of anthelminthic drugs used to treat the infestations has an unfavorable environmental impact. The aim of this study was therefore to identify genetic regions involved in sheep resistance to gastrointestinal strongyles, one of the most common classes of GIN at Mediterranean latitude. Fecal samples from 642 Comisana ewes were collected over a three years period to assess Fecal Egg Counts (FEC) with the Flotac method. Firstly, using pedigree data and logn(FEC+2) as phenotypes, EBVs for gastrointestinal strongyles resistance were estimated by a BLUP animal model. In a second phase, the EBVs were used to rank the animals of the flock. The EBVs in the 99.95th and the 0.05th percentile were used to identify and extract two groups of animals representing the most (31 sheep) and the less (29 sheep) genetically resistant individuals to gastrointestinal strongyles. The animals involved in the study were genotyped with the Illumina OvineSNP50 beadchip. Using the software PLINK a case/control GWAS (Genomewide Association Study) was performed considering the "less resistant" group as case and the "most resistant" as control. The p-values have been adjusted by the False Discovery Rate (FDR) using the Benjamini & Hochberg method and a threshold at the 0.005% most significant p-values was chosen. As a result, 18 significant SNPs (single nucleotide polymorphisms) involving 13 genes were identified on 12 chromosomes, showing strong signals on chromosome 1, 2, 4, 6, 13, 20. These findings confirm previous studies that correlate the same chromosomes to resistance to GIN infection. The identified SNPs fall within genes involved in the physiology or pathology of the intestinal tract. Indeed, the genes of the UGT1A\* family are involved in the metabolism of bilirubin and the metabolism of drugs at the intestinal level, while the genes KIF6, LOXL2 and CALN1 are involved in intestinal cancer and inflammation, immune response and in the regeneration and wounding of tissues, other as TWISTNB are known to have a role in gastrointestinal tract infections. Furthermore, many of them have a role in adaptive processes and production traits (LOXL2, GPC6, MYT1 and SS18L1).

#### Acknowledgements

The research was supported by the Italian National Sheep and Goat Breeders Association (ASSONAPA; Rome, Italy) through the CHEESR project PSRN 2014-2020. Sottomisura 10.2: Biodiversità

# Preliminary results of a machine learning approach to identify informative markers in trout populations

Giovanna Salvatore<sup>1</sup>\*, Valentino Palombo<sup>1</sup>, Stefano Esposito<sup>2</sup>, Nicolaia Iaffaldano<sup>1</sup>, Mariasilvia D'Andrea<sup>1</sup>

<sup>1</sup>Department of Agricultural, Environmental and Food Sciences, University of Molise, via De Sanctis snc, 86100 Campobasso, Italy <sup>2</sup>Mediterranean Trout Research Group, Via Porali 3, 42037 Ventasso, Italy \*Corresponding author: g.salvatore5@studenti.unimol.it

Mediterranean trout (Salmo cettii syn. Salmo macrostigma) is a freshwater fish of particular interest and it is seriously threatened by the introduction of commercial hatchery strains for recreation activities. The selection of hybrids based only on morphomeristic characteristics is insufficient to provide a clear taxonomic classification useful for hybridization control in conservation projects, thus genetic methods are traditionally adopted. Although developed for commercial rainbow trout species, recently high-density SNP arrays have been successfully used also for genetic characterization of local Mediterranean trout populations. In this context, the identification of a subset of most informative SNPs from the original panel may be beneficial for the development of a smaller and cost-effective array for specific conservation and on-field applications purposes. In this context, the use of Machine Learning (ML) approach is gaining attention and Random Forest (RF) classifier has been recently adopted for SNP prioritization. Thus, the aim of this work was to select a reduced panel of most informative markers from the Axiom 57K rainbow trout SNP array, useful to differentiate Atlantic and Mediterranean trout populations within LIFE Nat.Sal.Mo Project. With this general purpose, a total of 576 specimens, collected from the two main rivers of Molise and genotyped with the 57K rainbow trout Axiom array, were enrolled in this study. Axiom Analysis Suite and PLINK v1.09 software were used for sample and SNP quality control filtering, following the manufacturer's best practice recommendations. Respectively, individuals and SNPs with a call rate greater than 0.80 and 0.95 were retained for the analysis. Furthermore, SNPs with a minor allele frequency lower than 0.01 and deviating from Hardy–Weinberg equilibrium (P<10-3) were discarded. ADMIXTURE software v.1.3.0 was used to identify extreme not-introgressed and introgressed specimens. Such subset was used as training dataset to select most informative SNPs. RF classifier's outcomes were compared with those obtained by other statistical tests, notably FST, Delta and Principal Component Analysis and SNPs in common were considered to construct the reduced SNP panel, which was used to classify the entire population as testing dataset. Overall, the results looked promising indeed specimens were correctly assigned to their original population with a relatively low out of bag error (< 5%) also when a moderate-high introgression rate (i.e. 50%) was considered. Although further studies with larger samples size and possibly other Mediterranean populations are required to further corroborate such findings, this approach appeared interesting suggesting the idea to develop a reduced panel of SNP to successfully monitor the level of introgression between native and alien trout populations in Molise rivers.

#### Acknowledgments

The research was funded by LIFE Nat.Sal.Mo. Project



#### Effect of rearing systems on meat quality in Nero d'Aspromonte pigs

Mengjun Wu\*, Siria Tavaniello, Sanije Zejnelhoxha, Meng Peng, Antonella Di Iorio, Marisa Palazzo, Giuseppe Maiorano

Department of Agricultural, Environmental and Food Sciences, University of Molise, via De Sanctis snc, 86100 Campobasso, Italy \*Corresponding author: m.wu@studenti.unimol.it

Influences of pig rearing systems on animal performance, carcass and meat traits result from the interactive effects of: i) housing conditions; ii) feeding level and composition; iii) genotype, particularly in production systems with local pig breeds. The aim of the present study was to determine the effect of rearing systems (outdoor, O; indoor, I) on meat quality of barrows from Nero d'Aspromonte pigs, a local type reared in inner areas of Calabria region, Italy. Pigs reared in the I system were fed with a standard commercial diet. In the O system, pigs were kept on 3 ha land and feed consisted of pumpkin, acorn and pasture. Dietary supplementation with a standard commercial mixture was provided in periods of low pasture availability. Animals had ad libitum access to feed and water. When the pigs attained their target slaughter weight, they were transported to a commercial slaughterhouse. At slaughter, from 35 carcasses (O: n= 21; carcass weight:  $116.0\pm7.8$  kg; I: n = 14; carcass weight:  $115.6\pm9.0$  kg; P > 0.05) longissimus thoracis (LT) muscle samples were removed between the 13th-14th rib for cholesterol, vitamin E and fatty acids analyses. Data were analyzed by non-parametric test (Mann-Whitney). Meat from O pigs had higher (P < 0.05) vitamin E (4.84 vs 3.30 µg/g) and total lipid (3.81 vs 3.12 g/100g) content but lower (P < 0.05) cholesterol amount (56.4 vs 63.8 mg/100g) than that of I pigs. Concerning the fatty acid (FA) profile, meat from O group showed higher (+ 3.8%; P < 0.001) total saturated fatty acid (SFA) and lower (-2.6%; P < 0.01) polyunsaturated fatty acids (PUFA) as compared with I group; while monounsaturated fatty acids were similar between groups (P > 0.05). The nutritional ratios (n6/n3, PUFA/SFA, atherogenic and thrombogenic indices) were affected (P < 0.01) by the rearing system; compared with O group, I group showed higher n6/n3 and PUFA/SFA indices and lower atherogenic and thrombogenic indices. In conclusion, meat from O pigs has a lower cholesterol content and n6/n3 ratio, favorable from a nutritional point of view; moreover, it could result more suitable for technological transformation due to a lower PUFA content, which is prone to oxidation, and a higher vitamin E content, the nature's most effective lipid-soluble antioxidant.

#### Acknowledgments

The authors gratefully acknowledge the valuable contribution of Cooperativa Maiale Nero d'Aspromonte.

# Polycyclic aromatic hydrocarbons (PAHs) in charchoal-grilled beef meat: influence of different concentrations of Vitamin E added prior cooking.

Sanije Zejnelhoxha<sup>1</sup>, Olga Viegas<sup>2,3</sup>, Siria Tavaniello<sup>1</sup>, Edgar Pinto<sup>2,4</sup>, Mengju Wu<sup>1</sup>, Marisa Palazzo<sup>1</sup>, Isabel M.L.P.V.O Ferreira<sup>2</sup>, Giuseppe Maiorano<sup>1</sup>

<sup>1</sup>Department of Agricultural, Environmental and Food Sciences, University of Molise, via De Sanctis snc, 86100 Campobasso, Italy

<sup>2</sup>REQUIMTE, Laboratório de Bromatologia e Hidrologia, Departamento de Ciências Químicas, Faculdade de Farmácia da Universidade do Porto, Portugal

<sup>3</sup>Faculdade de Ciências da Nutrição e Alimentação da Universidade do Porto, Portugal.

<sup>4</sup>Department of Environmental Health, School of Health, Porto, Portugal.

\*Corresponding author: s.zejnelhoxha@studenti.unimol.it

High-temperature cooking methods such as grilling leads to the formation of polycyclic aromatic hydrocarbons (PAHs), potential carcinogenic agents for humans. PAH4 (sum of benz[a]anthracene, chrysene, benzo[b]fluoranthene and benzo[a]pyrene), is established by European Union as the most appropriate indicator for the occurrence and carcinogenic potency of PAHs in food. In the light of this, mitigation strategies consisting on the use of antioxidants to reduce their formation has gain great interest. This study aimed to evaluate the influence of the addition of vitamin E on the formation of PAHs in grilled beef burgers. Vitamin E content of meat was quantified (1.32  $\mu$ g/g,  $\alpha$ -tocopherol) and different concentrations DL- $\alpha$ -tocopheryl acetate (2, 3, 4.5 and  $6 \mu g/g$ ) were added to the meat prior grilling. 14 PAHs were extracted and analysed on well-done burgers using an acetonitrile based-extraction followed by high performance liquid chromatography with fluorescence detection. Data were analysed by one-way ANOVA. Control burgers without addition presented a mean value of 75.88 ng/g and 0.99 ng/g of total PAHs and PAH4, respectively. No effect was observed for total PAHs in samples with 2  $\mu$ g/g vitamin E addition. Significant reduction (P < 0.0001) of total PAHs formation were observed in burgers added with 3 and 4.5  $\mu$ g/g with mean values of 59.23 and 46.14 ng/g, respectively. For 6  $\mu$ g/g, significant reduction (P < 0.01) of total PAHs (65.08 ng/g) was also observed. In relation to PAH4, control samples presented values far the maximum established by the European Union (30 ng/g), nonetheless, the addition of vitamin E reduced significantly PAH4 formation in all concentrations. Although this meat did not generate high PAHs amount, it could be possible to observe that vitamin E influence PAHs formation, depending on the concentration.

### Impact of probiotic dietary supplementation on the colony growth and productivity of *Apis mellifera* L.

Sonia Petrarca<sup>1,2\*</sup>, Gianluca Albanese<sup>1</sup>, Massimo Iorizzo<sup>1</sup>, Antonio De Cristofaro<sup>1</sup>

<sup>1</sup> Department of Agricultural, Environmental and Food Sciences, University of Molise, via De Sanctis snc, 86100 Campobasso, Italy <sup>2</sup>Co.Na.Pro.A, Consorzio Nazionale Produttori Apistici, Campobasso, Italy \*Corresponding author: sonia.petrarca@studenti.unimol.it; petrarca.s@conaproa.it

Honey bee (Apis mellifera L.) is the most important pollinator of numerous crops and consequently it has a great biological and economic value. Therefore, the threats affecting this insect (e.g. pathogens, parasites, agrochemicals, climate change, anthropogenic disturbances) have prompted the scientific community to look for new mitigation tools, as alternatives to therapeutic agents, to preserve its health. Over the past decade, gut microbial symbionts have received a lot of attention for their potential contribution to honey bee welfare. However, to date, only a few studies have attempted to investigate the effect of bacterial supplementation as a strategy to improve the health status of colonies, particularly in terms of productivity and boosting the presence of beneficial microorganisms within the gut of the honey bee. In a preliminary laboratory study, two lactic acid bacteria (LAB) belonging to Lactiplantibacillus plantarum and Apilactobacillus kunkeei species, isolated from the gastrointestinal tract of honey bee, were selected for some bio-functional properties, high osmotic tolerance and for their ability to inhibit in vitro Paenibacillus larvae ATCC 9545 and Melissococccus plutonius ATCC 35311 strains. In a subsequent field study, these two LABs were added  $(10^7 \text{ CFU/mL})$  to sugar syrup (70%) sucrose) used as supplementary feeding in honey bee diet. The experimental apiaries, located in Morrone del Sannio (CB) and Castel del Giudice (IS), were chosen for their different orographic, geographic, climatic and floristic characteristics. The treatments were carried out every other day in the first two months (June-July) and weekly in the following 2 months (August-September). Treated and untreated honey bee colonies were monitored to assess the effects of probiotic syrup on their well-being and productivity. In particular, the administered LABs led to a significant increase of brood extension, bee population (method of the sixths) and harvestable honey. Further studies are underway to verify the in vivo efficacy of these probiotic bacteria in the biocontrol of American Foulbrood (AFB) and European Foulbrood (EFB) diseases.

### Semen cryopreservation for *ex situ* management of genetic diversity in Mediterranean trout resulting in the creation of the first European cryobank

Emanuele Antenucci\*, Giusy Rusco, Letizia Lerza, Michele Di Iorio, Nicolaia Iaffaldano

<sup>1</sup>Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis, 86100 Campobasso, Italy \*Corresponding author: e.antenucci@studenti.unimol.it

Semen cryobanking is a valuable tool to preserve the genetic resources of endangered fish species playing an important role in biodiversity conservation and restocking programmes. This is the case for Mediterranean brown trout (S. macrostigma) which is an endemic species from the Mediterranean area, that is declining as a result of river pollution, poorly regulated fishing and the genetic introgression following the introduction of zootechnical strains for recreational purposes. In the light of these threats, the Mediterranean brown trout is currently listed in Annex II of the Habitat Directive and its overall conservation status has been declared as "bad" and "in decline" at the EU level. The recent project "LIFE" Nat.Sal.Mo, funded by the EU, aims to ensure the recovery and the conservation of native Mediterranean trout (S. macrostigma = S. cettii) in Molise river basins (Molise region-Southern Italy). Among the specific objectives of the project is the restoration of genetic integrity by using frozen semen from pure wild breeders for artificial reproduction in combination with appropriate fertilization schemes increasing the genetic variability of the offspring and maintenance of the fitness within self-sustaining populations. In this regard, a milestone of Nat.Sal.Mo is the first semen European cryobank storing sperm from a large number of native breeders, this is an effective strategy for the protection of the biodiversity of the local Mediterranean brown trout populations. Therefore, the aim of this research consists in the constitution of the semen cryobank from native trout and its use for the restoration of genetic integrity and restocking program of Mediterranean trout population inhabiting the Molise rivers. During three spawning seasons (December/March starting 2019/20), sperm and eggs were obtained from Mediterranean brown trout caught from the Molise rivers and genetically characterized as native. The semen samples were collected by gentle abdominal massage and transported from the river to the laboratory in a portable refrigerator at 4 °C. Only sperm showing a motility higher than 75% were used and cryopreserved. A total of 1934 semen doses were produced from 102 native donators. 68 females were stripped to obtain 264.000 eggs that were split into 298 aliquots and each fertilized with frozen semen dose from high numbers of males (298) from the cryobank. Hatchery water was added to the fertilized eggs, then they were washed and incubated in a longitudinal hatchery tank with running water at a temperature of 9 °C. Unfertilized and dead eggs were continuously counted and removed. 192.000 eyed eggs i.e., close to hatching were obtained (yield rate of 72%). The eyed eggs were restocked and planted in suitable sites in Molise rivers through the innovative technique of building semi-artificial nests. In conclusion the creation of a sperm cryobank of pure Mediterranean trout populations with a high genetic variability represents the first of its kind and it represents a powerful multiplying effect which will affect other neighbouring areas at a national level and in other European river basins. In addition, thanks to the cryobank the percentage of native trout present in the project area increased from 70% to 85% and the introgression grade decreased.

Acknowledgements This study was financed by the LIFE project Nat. Sal. Mo. (LIFE17 NAT/IT/000547)



### High amylose wheat flours for the development of healthy cereal based foods

Martina Angelicola<sup>1\*</sup>, Maria Cristina Messia<sup>1</sup>, Emanuele Marconi<sup>1,2</sup>

<sup>1</sup>Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis, 86100 Campobasso, Italy <sup>2</sup>Campus Bio-Medico University of Rome, Faculty of Science and Technology for Humans and the Environment, Roma, Italy

\*Corresponding author: m.angelicola@studenti.unimol.it

Wheat is a versatile food ingredient to produce finished products of superior eating quality and widespread appeal. Starch is the main component of endosperm in wheat kernel and it is composed of two glucose polymers: amylose, mainly a linear chain, and amylopectin, a large highly branched molecule. Usual starch composition in wheat consists of 25% amylose and 75% amylopectin. To improve the nutritional quality of refined wheat-based food, manipulation of starchy endosperm targeting for changing the amylose/amylopectin ratio in favor of amylose could provide substantial health benefits at a global level through the delivery of resistant starch. In light of these considerations, this research aims to produce innovative cereal products with enhanced nutritional and healthy properties using high amylose wheat flours. To reach this objective, a complete characterization of normal and high amylose wheat samples (Triticum aestivum L.) was carried out. Several milling diagrams with both soft and hard wheat mill were assessed and chemical and rheological characteristics were evaluated on milling products to test technological performances. Flours obtained by conventional grinding of high amylose soft wheat had lower yield (~50%) compared to control line (65%) and higher ash content (>0.80% d.w.). Debranning of high amylose wheat at 6% increased yield and refining level of some fractions and the further application of a hard-wheat milling diagram improved milling performance (yield = 66.3%). The high amylose flours presented enhanced nutritional characteristics, due to the high content of resistant starch (up to 28.9% of total starch). As a consequence, they could be used as raw materials for the production of innovative foods with elevated contents of resistant starch and fiber which might bear nutritional and healthy claims according to EU Reg. 432/2012 and EC Reg. 1924/2006. In addition, during the evaluation of technological characteristics of the flours, it was emerged that official methods of analysis used for conventional soft wheat might be adapted for the modified samples, because the higher content of amylose modifies water absorption and gelatinization properties of the dough.

### From durum wheat to whole-meal pasta: effects of processing and cooking on physicochemical and nutritional properties

Valentina Di Nardo<sup>1</sup>\*, Emanuele Marconi<sup>1,2</sup>, Elisa De Arcangelis<sup>2</sup>

<sup>1</sup>Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis, 86100 Campobasso, Italy <sup>2</sup>Campus Bio-Medico University of Rome, Faculty of Science and Technology for Humans and the Environment, Roma, Italy \*Corresponding author: v.dinardo1@studenti.unimol.it

Folates are essential micronutrients for human health belonging to the group of water-soluble B vitamins. They are involved in numerous one-carbon transfer reactions including the DNA, RNA and protein synthesis and methionine cycle. Folate deficiency is associated with several health disorders, first of all neural tube defects (NTDs) in developing fetus. Cereals and cereal-based products, especially in the whole-meal version, are an important source of folate in the human diet, both for their folate content and for their frequency of consumption. On the other hand, food processing can lead to substantial losses of these compounds since they are highly sensitive to various factors such as oxygen, heat, pH, UV light. Debranning technology, at the same time, can be used to obtain cereal bran fractions with increased folate concentration for the development of foods naturally enriched in folate. In view of these considerations, the aim of this work was i) the evaluation of the effect of industrial debranning and milling processes on total folate content in durum wheat, ii) the selection and use of folate-rich fractions of durum wheat for the production of whole-meal pasta and iii) the study of the impact of cooking process on folate retention in whole-meal pasta. For this purpose, durum wheat samples of different origins were taken into consideration and two pasta making trials were carried out using different formulations. Total folate content was determined using a microbiological assay preceded by an enzymatic extraction in durum wheat and debranning and milling fractions, and in the experimental uncooked and cooked whole-meal pasta. For comparative purposes, whole-meal pasta of different brands available on the market were also used. Experimental whole-meal pasta had folate values (> 30  $\mu$ g/100 g) capable of covering more than 15% of the reference nutritional values (EU Reg. No. 1169/2011), therefore being able to boast related nutrition and health claims on the label (EC Reg. No 1924/2006 and EU Reg. No 432/2012), and showed a high folate retention after cooking. This study led to a better understanding of the effects that food processing can have on total folate content from durum wheat to whole-meal pasta, highlighting how the debranning process prior to milling has led to the obtaining of flour fractions rich in folate which can be used for the production of whole-meal pasta with improved nutritional properties considering the high retention of folate after cooking.

# Combination of empirical and fundamental rheology for dough characterization: an integrated approach

Silvio Iacovino<sup>1</sup>\*, Francesca Cuomo<sup>1</sup>, Emanuele Marconi<sup>1,2</sup>

<sup>1</sup>Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis, 86100 Campobasso, Italy <sup>2</sup>Campus Bio-Medico University of Rome, Faculty of Science and Technology for Humans and the Environment, Roma, Italy \*Corresponding author: s.iacovino@studenti.unimol.it

Fundamental rheology represents a useful tool for studying the viscoelastic and microstructure properties of dough. This approach was combined with empirical rheology to have complementary information. Thus, the relationship between the two approaches was checked. The fundamental rheological characterization of dough made through a rheometer was set up and the outputs were compared to the results of farinograph and alveograph analyses. The rheological properties of flour and semolina, ranging from refined flours to whole grains were studied. Farinograph and alveograph analyses were carried out according to AACC International Methods 54-21 and 54-30, respectively. For fundamental rheological analysis, dough were prepared through the farinograph kneader according to their optimum water content and development time. Rheological tests were carried out through a modular rheometer equipped with a 20 mm parallel plate probe. Samples were checked through creep recovery and oscillatory time, amplitude, frequency and temperature sweep tests. The results showed a good correlation between empirical and fundamental rheology within the linear viscoelastic range. Moreover, farinograph and alveograph analyses indicated that higher protein contents led to higher stability, while higher amounts of fiber led to higher tenacity and lower elasticity values. Amplitude sweep tests evidenced the predominant solid-like character of dough, which showed a linear viscoelastic range within a zone of very small deformation. Frequency sweep test confirmed the main role of protein content on dough stability, while temperature sweep tests showed that starch gelatinization process starting temperature increased with fiber content. Finally, creep and recovery tests evidenced that part of the deformation generated by the loading phase was not recovered because of the viscoelastic character of the dough. Moreover, higher fiber content seemed to decrease the overall recovery ability of the systems. The obtained results demonstrated the utility of this combined approach in order to deeply characterize flours and/or flour blends.

### Acrylamide in baked products: the case of Neapolitan pizza

Michela Quiquero<sup>1\*</sup>, Emanuele Marconi<sup>1,2,</sup> Maria Cristina Messia<sup>1</sup>

<sup>1</sup>Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis, 86100 Campobasso, Italy <sup>2</sup>Campus Bio-Medico University of Rome, Faculty of Science and Technology for Humans and the Environment, Roma, Italy

\*Corresponding author: m.quiquero@studenti.unimol.it

Acrylamide is a compound that originates in some foods starting from asparagine and reducing sugars during the cooking process at temperatures above 120 °C and low moisture, typical of the Maillard reaction. The foods most subjected to acrylamide formation are potato-based products, bakery products and coffee, due to their composition and the high temperatures reached during processing. Acrylamide has been classified as a potential carcinogen for humans (International Agency for Research on Cancer, group 2A) and as genotoxic and neurotoxic for animals. In 2015, the European Food Safety Authority considered the presence of acrylamide in food a public health concern, recommending the need to identify measures to reduce consumer exposure to this substance. The Commission Regulation (EU) 2017/2158 established mitigation measures to reduce the presence of acrylamide in food and reference levels in different foods, in the range 40 - 4000  $\mu$ g / kg. In this context, the aim of the study was to investigate on acrylamide content in Neapolitan pizza, an icon of the Italian gastronomic and cultural panorama, where it can be developed for the natural composition of the ingredients and the high temperatures (430 - 485 °C) reached in the ovens.

Acrylamide has been evaluated in different sections of Neapolitan pizza (external part, internal part, slice) with and without the presence of topping, and in pizza samples obtained with leavened doughs for different times, to evaluate fermentation with microbial starters as a possible mitigation strategy for the formation of acrylamide. Particularly, after the extraction and purification phase, freeze-dried samples were injected in an HPLC-UV instrumentation for acrylamide determination. The research revealed a different distribution of acrylamide both in the various parts of pizza and in the pizza with and without topping, suggesting a likely influence of topping on acrylamide formation as a result of the effect on factors such as pH and moisture, that affect the development and course of the Maillard reaction. A decreasing value of acrylamide has been observed for the samples of Neapolitan pizza subjected to various leavening time, indicating the prolonged fermentation as a possible mitigation strategy.

# Metagenomic characterization of microbial communities in different Pecorino cheese from Campania Region

Diletta Bagnoli\*, Franca Vergalito, Mariantonietta Succi

Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis, 86100 Campobasso, Italy \*Corresponding author: d.bagnoli@studenti.unimol.it

Metagenomic characterization of microbial communities associated with milk and cheese is now considered a key factor in the dairy industry, as it allows the identification of peculiar microbial species that can contribute to the quality and distinctiveness of products. The microbiota of pecorino cheeses from Campania region has never been fully explored. The aim of this study was to investigate the bacterial distribution in these products using the next-generation sequencing (NGS) technique, Illumina MiSeq. Accordingly, samples of sheep's milk and pecorino cheeses matured at 0, 1, 2 and 4 months were taken from three different farms in Campania. The bacterial communities associated with each sample were accessed through sequencing of V3-V4 region of 16S rRNA gene. For both analysed matrices, the most abundant sequences were identified at the genus level (over 78%). The results revealed a rich microbial community, composed on average of 44 and 21 bacterial genera for sheep milk and pecorino cheeses, respectively. More than 20 genera were identified in each analysed matrix, ranging between dominant (relative abundance >1%) and subdominant (<1%) taxa. Specifically, 11 taxa were present in all sheep milk samples (core taxa), including the genera Pseudomonas, Comamonas, and Burkholderia (average relative abundance >5%), while 5 taxa were present in all pecorino cheese samples, including the genera Lactobacillus and Streptococcus. These core taxa reflected the predominance of the phyla Proteobacteria and Firmicutes in the sheep milk and pecorino cheese samples, respectively. Ultimately, the application of the high-throughput DNA sequencing (HTS) technique provided a detailed microbial profile of the sheep cheeses analyzed. This characterization study can contribute to a better understanding of the microbiome of these products, helping interested producers to preserve their identity and optimize their quality, safety and commercial value.

# Evaluation of functional properties from *Lactiplantibacillus plantarum* strains isolated from bee bread

Francesco Letizia\*, Massimo Iorizzo

Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis, 86100 Campobasso, Italy \*Corresponding author: f.letizia@studenti.unimol.it

According to the current definition, formulated in 2002 by FAO, probiotics are "live strains of strictly selected microorganisms which, when administered in adequate amounts, confer a health benefit on the host". In details, a probiotic agent must be non-pathogenic and to possess the following characteristics: capability to survive in the digestive tract, adherence to the intestinal epithelium and colonization of the intestinal tract; production of antimicrobial compounds; good stability in the form of powder, liquid or food. A considerable number of well-characterized probiotic strains are available worldwide, nevertheless the research into novel strains with specific and improved properties is still of major appeal. In fact, recently alongside the conventional screening procedures, the pharmaceutical and food industries have encouraged scientific research towards the selection of new probiotic bacterial strains with particular functional features. In this context, the aim of our study was the evaluation of novel functional properties of five Lactiplantibacillus plantarum strains isolated from bee bread. The main objective was to establish a possible applicability of these Lp. plantarum strains as dietary supplements in the human diet and/or as microbial cultures for the production of functional food. Specifically, antioxidant, antimicrobial and  $\beta$ -glucosidase activities, exopolysaccharides (EPS) production and the ability to synthesize  $\gamma$ -aminobutyric acid (GABA) were evaluated, combining biochemical colorimetric and enzymatic assays, as well as high-performance liquid chromatography (HPLC). The results demonstrated that the investigated Lp. plantarum strains were effective in inhibiting the growth of some human opportunistic pathogens in vitro (Ps. aeruginosa, E. coli, P. mirabilis, E. faecalis and S. aureus). Moreover, the evaluation of antioxidant and β-glucosidase activity and of EPS and GABA production, revealed a different behaviour among the strains, suggesting that these properties are strongly strain-dependent. The results highlighted that the five strains of Lp. plantarum are promising candidates for application as dietary supplements in the human diet and as microbial cultures in specific food productions. Moreover, they indicate that a careful selection within a given species is of utmost importance for the improvement of specific biotechnological applications.

## Food Science, Technology and Biotechnology

# Antimicrobial edible films and coatings in microbiological quality preservation of dried fruits

Ida Mercurio \*, Silvia Jane Lombardi, Patrizio Tremonte, Raffaele Coppola

Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis, 86100 Campobasso, Italy \*Corresponding author: i.mercurio@studenti.unimol.it

The last years have been marked by an increase and diversification of dried fruits which, besides being widely used in the sweet industry, are proposed in single dose sizes as ready-to-eat foods. Ready-to-eat products are offered as raw or minimally processed food, therefore they need a particular attention for microbiological and safety characteristics. The single portion formats offered by the market are mainly based on roasted or dried nuts. The toasting treatment certainly offers relatively higher safety conditions compared to the drying one which is generally conducted at temperatures of about 40° C. However, it should be considered that roasting is capable of eliminating vegetative microbial forms including fungal forms but it is not capable of removing the dangers toxic metabolites produced fungal strains. As well as roasting causes sensorial and qualitative changes in the product. The present study aimed to develop an innovative protective tool based on the activation of an edible film through the use of natural metabolites of microbial origin. The identification of different fungal species and their persistence after the drying process posted the identification of a Lactiplantobacillus plantarum strain with antifungal activity. The primary screening was followed by secondary screening aimed at identifying the metabolite responsible for antifungal action. The identification was done through HPLC analysis, and determined the presence of Phenylactic Acid (PhLA) then added to the edible film for its activation. Based on the properties of the nuts and the characteristics of the antimicrobial compound, carboxymethylcellulose was used. The effect of PhLA added to the edible film on the microbiological quality of dried fruit was evaluated by means of storage tests. The results show excellent efficacy of the activated edible film and represent an important starting point for the development of new biotechnological strategies in dried fruit preservation.

## Food Science, Technology and Biotechnology

# Sample preparation, quantification and qualification of microplastics in some food products

Cristina Di Fiore\*, Pasquale Avino

Department of Agricultural, Environmental and Food Sciences, University of Molise, via F. De Sanctis, 86100 Campobasso, Italy \*Corresponding author: c.difiore@studenti.unimol.it

Microplastic is an environmental problem garnering attention from scientific communities. About 3 900 million tons of plastic were produced only between 2002 and 2015 and 40% of the total is used by the food sector for food packaging. Despite its advantageous characteristics for food storage and transport, plastic material is a serious source of microplastics in food matrices. Currently, the presence of microplastics in food products is well-documented in a few ranges of foods such as fish, salts, honey whereas scares information are provided for cereals, meats, vegetables and fruits. Furthermore, no harmonization of analytical procedures for their determination is available. The present study aims to set up an analytical methodology for quantification and quantification of microplastics in some food products, in particular the two most consumed food by Italian population: pasta and red meat. The first step is the organic matter removal, which affects microplastics quantification and qualification. Several digestion methods were thus studied to evaluate the digestion efficiency, which needs to be higher than 95% to allow an acceptable recovery rate (>95%) of microplastics. Therefore, fluorescent microbeads solution (PS-MPs) (particles diameter 9.9 um) was used to evaluate the effects of different digestion methods on microplastics and their recovery rate. Particularly, Fenton's reagents, potassium hydroxide, hydrogen peroxide and nitric acid were evaluated as agents for the organic matter removal. Solutions obtained after digestion protocol, both with PS-MPs spike and without, were filtered using glass membrane filters and analysed under a stereomicroscope for an optical analysis of the digestion efficiency and a visual quantification of microplastics. Olympus Stream Start software was used for the analysis of microplastics after digestion protocol to evaluate changes in color, size and shape. The experiments showed good results in organic matter digestion. In particular, the percentage digestion efficiencies were of 98.0 % for meat sample using potassium hydroxide 5 molar in concentration and of 95% for pasta sample using nitric acid 65%. The percentage recovery rates were higher than 95%, indicating the goodness of the developed analytical methodology.



#### Screening of Lactic Acid Bacteria for the Biocontrol of American and European foulbrood diseases Gianluca Albanese

#### Introduction

Wild and managed bees have crucial ecological, economic and social importance including and beyond crop pollination [1]. There are several biotic and abiotic threats to which this insect is exposed; biotic factors include diseases caused by pathogens [2]. *Melissococcus plutonius* is the causal agent of European foulbrood (EFB), while *Paenibacillus larvae* is the etiological agent of American foulbrood (AFB) [3,4]. Heavy brood losses, colony collapse, and extreme contagiousness render EFB, and in particular AFB, economically important and notifiable diseases in many world regions [2].

As an alternative to antibiotics, the use of beneficial microbes as dietary supplements in the diet of honey bees could represent a valid eco-friendly strategy [5].

#### Materials and Methods

Four strains of *Lactiplantibacillus plantarum* and four strains of *Apilactobacillus kunkeei*, which were isolated from the gastrointestinal tract of honey bees, were selected for their capacity *in vitro* to inhibit *Paenibacillus larvae* ATCC 9545 and *Melissococccus plutonius* ATCC 35311. The antimicrobial activity of cell-free supernatant (CFS) was evaluated using the agar well diffusion assay. Furthermore, these lactic acid bacteria (LAB) strains were assessed for their carbohydrate fermentation pattern using the API 50CHL system kit and enzymatic patterns using the API ZYM system kit (bioMérieux SA, Marcy l'Etoile, France).

	Antimicrobial activity expressed as zone of inhibition-ZOI (mm)							
Pathogens	LP31	LP42	LP148	LP179	ALK181	ALK222	ALK268	ALK385
P. larvae	14.9±0.2 <sup>b</sup>	15.1±0.5⁵	15.8±0.3ª	13.9±0.4 <sup>c</sup>	15.5±0.5ª	16.3±0.2ª	15.0±0.4 <sup>b</sup>	13.7±0.2°
M. plutonius	15.1±0.3ª	15.1±0.3ª	16.0±0.1ª	12.0±0.2 <sup>d</sup>	15.9±0.4ª	14.0±0.3 <sup>b</sup>	14.2±0.3 <sup>b</sup>	13.0±0.5°

#### Discussions and Conclusions

The eight LAB strains exhibited inhibitory activity against *P. larvae* ATCC 9545 and *M. plutonius* ATCC 35311. The results showed that the antagonist action is not related to organic acids, but to other metabolites present in the CFS. The selected LAB strains, used as probiotic supplements, thanks to their specific enzymatic activities, could contribute to the breakdown of complex polysaccharides, metabolize toxic sugars and consequently improve the dietary tolerance of honey bees. Further investigations are needed to assess *in vivo* the ability of these LABs, used as probiotic supplements in the diet, to improve the well-being of honey bees and be effective in a biocontrol strategy against EFB and AFB diseases.

#### References

Patel V, Pauli N, Biggs E, Barbour L, Boruff B. Why bees are critical for achieving sustainable development. Ambio 2021;50:49–59.
 Genersch E. Honey bee pathology: current threats to honey bees and beekeeping. Appl Microbiol Biotechnol 2010;87:87–97.

[3] Forsgren E. European foulbrood in honey bees. J Invertebr Pathol 2010;103:55–9.

[4] Genersch E. American Foulbrood in honeybees and its causative agent, Paenibacillus larvae. J Invertebr Pathol 2010;103:S10-9.

[5] Iorizzo M, Testa B, Lombardi SJ, Ganassi S, Ianiro M, Letizia F, Succi M, Tremonte P, Vergalito F, Cozzolino A, Sorrentino E, Coppola R, Petrarca S, Mancini M and De Cristofaro A. Antimicrobial activity against Paenibacillus larvae and functional properties of Lactiplantibacillus plantarum strains: Potential benefits for honeybee health. Antibiotics. 2020;9(8):442.

Supervisors: Prof. Massimo Iorizzo and Prof. Antonio De Cristofaro









#### Post-domestication diffusion processes and adaptation in livestock and relationship with human populations by using SNPs data in a comparative framework

#### Marika Di Civita. m.dicivita@studenti.unimol.it

Introduction

Livestock breeds have been shaped over time by a combination of human-mediated pressures and a long history of migrations, selection, and adaptation [1]. Consequently, DNA from domesticated plants and animals contains information about how species were domesticated, exported, and bred by early farmers [2]. In recent years, the use of single nucleotide polymorphism (SNP) marker panels for many livestock species has proven to be particularly useful for analyzing genomic patterns and genome-wide diversity of breeds [3] to provide further insights on the historical past of domestic animals and on the contemporary genetic variability. Moreover, animal domestication and modern animal breeding are closely related, with strong artificial selection, which leads to the genetic improvement of animal production traits. These selection strategies are expected to leave imprints in the genome that are identified as selection signatures [4]. In fact, population genomics selection signatures studies could help to find the genetic resources of livestock [5]. However, such studies have been unevenly applied across domestic breeds worldwide [6]. Hence, genomic and phenotypic resources are still lacking for many non-commercial and traditional breeds [7], which may have unique genetic variants and related to ecological traits, such as adaptation to a particular environment [8].

#### Aim of the research & Methods

The overarching aim of the research will be to study post-domestication diffusion processes employing genome-wide SNP data belonging to different livestock species, using mostly local breeds. The innovative aspect will also be to include genomic SNPs data from several plant species and humans to provide a better overview in a comparative framework and to improve our understanding of anthropogenic pressure on the evolution and diversification of livestock breeds. In addition, at local scales, another necessary goal will involve the use of selection signatures approaches to understand possible genomic regions that influence phenotypic differentiation in livestock breeds.

For this purpose, we will implement descriptive analysis including observed (Ho) and expected (He) heterozygosity, the minor allele frequency (MAF) and effective population size. Concerning the genetic structure and breed relationships we will use Admixture software, Multidimensional scaling and Treemix while approximate Bayesian computation methodologies will be used to test the most reliable scenario of postdomestication diffusion.



SNPs Data Distribution

**Expected results** 

The main expected outcome would be to provide for the first time a general framework aimed at reconstructing post domestication diffusion processes by comparing genomic data from different livestock species including plants and humans. By using selection signature methodologies, we will be able to identify regions of the genome that have been under selection in the history of breeds domestication. The selected regions may harbour functional elements responsible for the development of desired traits that would be useful for improving productive and adaptive traits in European local breeds.

#### References

[1] Wilkinson, S., & Wiener, P. (2018). Population Genomics of Animal Domestication and Breed Development. In Population Genomics (pp. 709-753). Springer,

[2] Decker, J. E., McKay, S. D., Rolf, M. M., Kim, J., Molina Alcalá, A., Sonstegard, T. S., ... & Taylor, J. F. (2014). Worldwide patterns of ancestry, divergence, and admixture in domesticated cattle. PLoS genetics, 10(3), e1004254. [3] Colli, L., Milanesi, M., Vajana, E., Iamartino, D., Bomba, L., Puglisi, F., ... & Ajmone-Marsan, P. (2018). New insights on water buffalo genomic diversity and post-domestication migration routes from medium density SNP chip data. Frontiers in genetics, 9, 53

[4] Saravanan, K. A., Panigrahi, M., Kumar, H., Bhushan, B., Dutt, T., & Mishra, B. P. (2020). Selection signatures in livestock genome: A review of concepts, approaches and applications. Livestock Science, 241, 104257. [5] Cesarani, A., Sorbolini, S., Criscione, A., Bordonaro, S., Pulina, G., Battacone, G., ... & Macciotta, N. P. P. (2018). Genome-wide variability and selection

atures in Italian island cattle breeds. Animal genetics, 49(5), 371-383. [6] Mwai, O., Hanotte, O., Kwon, Y. J., & Cho, S. (2015). African indigenous cattle: unique genetic resources in a rapidly changing world. Asian-Australasian

journal of animal sciences, 28(7), 911.

[7] Bruford, M. W., Ginja, C., Hoffmann, I., Joost, S., Orozco-terWengel, P., Alberto, F. J., ... & Zhan, X. (2015). Prospects and challenges for the conservation of farm animal genomic resources, 2015-2025. Frontiers in genetics, 6, 314. [8] Yurchenko, A., Yudin, N., Aitnazarov, R., Plyusnina, A., Brukhin, V., Soloshenko, V., ... & Larkin, D. M. (2018). Genome-wide genotyping uncovers genetic profiles and history of the Russian cattle breeds. Heredity, 120(2), 125-137.









#### PERCEPTION AND INFLUENCE OF CLIMATE CHANGE IN THE INNER AREA ALTO E MEDIO SANNIO (AMS) Sara Bispini, s.bispini@studenti.unimol.it

#### INTRODUCTION

Mountain ecosystems are particularly vulnerable to climate change, which, together with other factors (resource overexploitation and depopulation), generates strong impacts and consequences for downstream ecosystems as well (Gupta et al. 2021; Schneiderbauer et al., 2021). Analysing the perception of mountain communities to climate d investigating the adaptation mechanisms adopted by them could therefore facilitate the work of policy makers in providing adaptation and resilience strategies change a

change and investigating the adaptation mechanisms adopted by them could therefore hacilitate the work of policy makers in providing adaptation and resilience strategies and outlining precautionary interventions. Recently, national and international policies have focused on strategies for climate change impacts mitigation, but local communities have nevertheless developed adaptation strategies, including through the reintroduction of traditional ecological practices (Ingty, 2017). Mountain territories, although manifesting a significant fragility due to morphological and environmental conditions, present, however, a high propensity to resilience and adaptation (ASVIS, 2022). In fact, as the Agency for Territorial Cohesion (2013) observes, the Inner Areas, despite being those areas that are significantly distant from the centres offering essential services (education, health and mobility), are rich in important environmental and cultural resources that should be enhanced. Among these, in the AMS, certainly the pasture-breeding-cheese spun paste system, on which rural development action should be prioritised, considering the positive externalities underlying this value chain. In recent years, the Inner Areas have been the object of the National Strategy for Inner Areas (SNAI), a place-based action whose main objective, in addition to the atterparthement of functions of strategies to fight demonstration actions for the terminet of these to fight demonstration actions for the terminet of these to fight demonstration actions for the terminet of these to fight demonstrations for the terminet of the strategies to fight demonstrations for the terminet of these to fight demonstration actions for the terminet of these to fight demonstrations of strategies to fight demonstrations for the terminet of these to fight demonstration actions when the terminet and the to fight demonstration actions acti addition to the strengthening of essential services, is to promote a shared vision for the territorial development of these areas in order to fight depopulation and further marginalisation. For this purpose, within the project MOVING (Mountain Valorization through INterconnectedness and Green Growth- H2020- Project ID 862739, whose aim is to build capacities and co-develop - through a bottom-up participatory process - relevant policy frameworks across Europe for the establishment of new or upgraded/upscaled value chains that contribute to the resilience and sustainability of mountain areas to climate change), a field survey was carried out to identify perceptions and awareness of local actors on the reference variable underlying the processes of territorial regeneration based on endogenous rural development (Van der Ploeg et al. 2000)

#### MATERIALS AND METHODS

Methodology was developed from a conceptual framework developed within the MOVING project. The work was carried out in the period 1.11.2021-31.12.2021 in the AMS sub-region, identified as the Mountain Reference Landscape (MRL). This sub-region, composed by the municipalities of Agnone, Capracotta, Carovilli, Pescolanciano and Vastogirardi (fig. 1), represents the major thickening of the dairy value chain of spun paste.

The research materials were collected directly on the field through the follo phases:

1. design of a semi-structured questionnaire aimed at identifying the reference variable and identifying the main drivers attributable to the impact of climate change on it.

2. Survey by semi-structured interviews with a panel of privileged witnesses (naturalists, breeders, cheesemakers, etc.),

3. Reporting of the results through a participatory event (workshop) in which actors of the MRL analysed and discussed the drivers that emerged, hierarchizing them and discussing the possible adaptation mechanisms in place or potential, expression of the potential resilience of the area.







Fig. 2- Alto Molise meadows

RESULTS	RESULTS						
RANKING	DRIVER	PERCEPTION	INFLUENCE	ADAPTIVE CAPACITY MECHANISM			
1	PRECIPITATION	Very relevant	The hay mowing period is changing	-Technological innovations to store hay and herbs in advance to reduce adverse climate events.			
2	TEMPERATURE	Very relevant	Dry grassland; negative effects in hay quality and quantity; livestock healt (and milk production) hit by thermal changes.	-Cultivation experiments: introduction of new vegetable varieties that are arid resistant that reduce the damage of rain variability.			
3	EXTREME EVENTS	Relevant	Late frosts dangerous for grassland; hydro-geological instability.	Breeding experiments: try to introduce ne cow varieties that can produce milk in wor			
4	SOIL PHISICAL DEGRADATION	Relevant	High temperatures pulverize the soil; less persistent snowpack reduces the soil ability to store water.	climate conditions reducing farm costs. -Business diversification to reduce econon damages for climate causes, diversifying far the activity (e.g., rural tourism, goat and she			
5	WILDFIRE	Potentially relevant	Dangeorus for grassland.	breeding, energy production with animal by- products).			

#### CONCLUSIONS

DECLUT

The perception of climate change and its underlying risks proved to be an important factor for the design of adaptation mechanisms. The value chain that runs through the MRL could be at risk, especially with regard to the quality of final products and raw materials (dependent on natural resources such as grassland, hay, etc.), as

climate change and its impacts are the cause. Although depopulation is the most worrying factor in the analysed area, the group of local actors involved outlined a number of possible adaptation mechanisms that will be a starting point for the design and proposal of new policies aimed at ensuring the resilience of mountain areas in the following phases of the project.

REFERENCES Gupta, H., M. Nishi, A. Gasparatos (2021), Community-based responses for trackling environmental and socio-economic change and impacts in mountain socio-ecological systems, *Ambio*. Ingty, T. (2017), High mountain communities and climate change adaptation, traditional ecological knowledge and institutions; Climatic change Schneiderbauer, S., P. Contanelle His, J. L. Delves, J. Pedoh, S. Kartaf, M. Escobaren, T. Thaler, F. carnelle et al. (2021), Risk perception of Climate change and natural hazards in global mou ain regions: a critical review:

Science of the total environment. ASVIS (2022), Le Aree Interne e la m

Supervisor: Prof. A. Belliggiano; Unimol Team coordinator for MOVING project: Prof. C. Ievoli

ence of me some environment. NUS (2022), Le Avera Interne e la montagna per lo svihuppo sostenibile. enzia per la coesione territoriale (2013), Strategia nazionale per le Aree interne: definizione, obiettivi, strumenti e governance <u>https://www.agenziacoesione.gov/it/stategia-nazionale-aree-interne la-selezione-delle-aree/</u> and # Plong, J. H. Renting, G. Brunor, K. Knickel, J. Mamino, T. M. Kest de Roest, E. Sevilla-Guzmán, F. Ventura (2000), Rural Development: From Practices and Policies towards Theory, *Sociologia Ruraliz*, vol 40,

BYDGOSZCZ UNIVERSITY **OF SCIENCE AND TECHNOLOGY** 







#### INTERNATIONAL Phd Student Conference Research and Innovation in

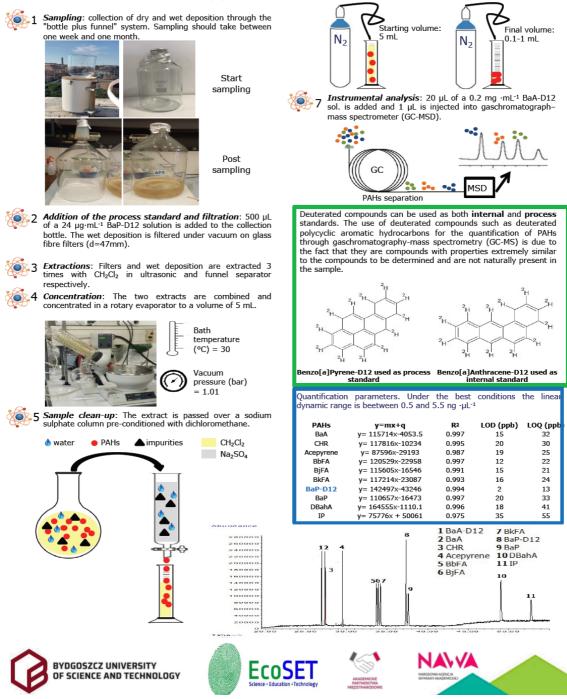
Agricultural, Livestock and Food Sciences

#### THE ROLE OF DEUTERATED COMPOUNDS IN THE DETERMINATION OF POLYCYCLIC AROMATIC HYDROCARBONS (PAHs) IN ENVIRONMENTAL AND ANGRO-FOOD MATRICES

Giuseppe Ianiri, Alessandra Fratianni, Gianfranco Panfili Department of Agricultural, Environmental and Food Sciences, University of Molise via F. De Sanctis, I-86100 – Campobasso, Italy

**Analytical procedure** for the determination of polycyclic aromatic hydrocarbons (PAHs) in environmental matrices using BaP-D12 as process standard and BaA-D12 as internal standard (IS).

6 **Concentration under nitrogen (N<sub>2</sub>) flow:** Nitrogen of the extract.





## **Novel Protective Cultures for Clean-Label Food Products**

PhD student: Ilenia Iarusso Supervisor: Patrizio Tremonte

#### State of Art

The recent focus of scientific investigations has moved from a great attention to the sustainability and to the food products safety (Pires et al. 2021). In addition, the emergency of antibiotic-resistant pathogens and consumers demand for minimally processed foods, highlight the need to adopt "clean label approaches" as screens against foodborne spoilers and pathogens (Heymich et al. 2021). In the control of foodborne pathogens, several authors (Mota-Gutierrez and Cocolin, 2021), reported a great number of protective lactic acid bacteria strains which commonly produce organic acids, peroxides, enzymes and bacteriocins. However, the effectiveness of their application as food additives may be limited for various reasons related to the ecological characteristics of the foodstuff (Acin-Albiac et al. 2022). On this basis, new protective cultures with improved performance under a wide range of ecological conditions should be developed.

#### Materials and methods

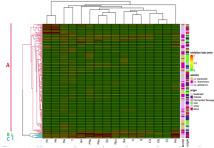
One hundred and seventy lactic acid bacteria strains were isolated from different fermented foods (tested for their antimicrobial activity against 16 undesirable microbial strains (indicators) including fungi and bacteria.



Figure 1. Experimental design. Lactic acid bacteria strains isolated from typical and atypical foods characterized by harsh environmental conditions were preliminarily identified and screened for their antimicrobial activity

#### Results

Based on the inhibitory activity expressed by growing cells, producers were grouped into three clusters (Figure 2).



#### Discussion and conclusion

Figure 2 Heat map showing the similarity in the profiles of antimicrobial activity of growing cells of producer's strains vs indicators strains: Penicillium commune (Pc), Penicillium echinolatum (Pe), Pseudomonas fragi (Pfr), Tricoderma (T), Ryzopus oryzaea (Ro), Clostridium tyrobutyricum (Ct), Pseudomonas fluorescens (Pflu), Escherichia coli (Ec), Pseudomonas aeruginosa (Pau), Clostridium butyricum (Cb), Staphilococcus aureus (Sa), Listeria innocua (Li), Pseudomonas putida (Ppu), Salmonella (S), Clostridium sporogenes (Cs), Aspergillus niger (An) Strains were grouped Lactiplantibacillus plantarum (2) and Lacticaseibacillus rhamnosus (1) showing a strong inhibitory activity; cluster C grouped Lactiplantibacillus plantarum (2) and Lacticasei (2) with moderate or high inhibitory activity against all assayed indicators.

Results indicate that specific food conditions can influence the occurrence of certain strains of lactic acid bacteria able not only to respond to specific adverse conditions, but also to compete with other bacterial populations. Therefore, the choice of the source of isolation could be an important preliminary tool for the selection of antagonistic strains.

#### References

Acin-Albiac, M., Filannino, P., Coda, R., Rizzello, C. G., Gobbetti, M., & Di Cagno, R. (2022). How water-soluble saccharides drive the metabolism of lactic acid bacteria during fermentation of brewers' spent grain. *Microbial biotechnology*, 15(3), 915-930.
Pires, S. M., Desta, B. N., Mughini-Gras, L., Mmbaga, B. T., Fayemi, O. E., Salvador, E. M., ... & Devleesschauwer, B. (2021). Burden of foodborne diseases:

Pires, S. M., Desta, B. N., Mughini-Gras, L., Mmbaga, B. T., Fayemi, O. E., Salvador, E. M., ... & Devleesschauwer, B. (2021). Burden of foodborne diseases: think global, act local.Current Opinion in Food Science. Heymich, M. L., Friedlein, U., Trollmann, M., Schwaiger, K., Böckmann, R. A., & Pischetsrieder, M. (2021). Generation of antimicrobial peptides Leg1 and Leg2

Heymich, M. L., Friedlein, U., Irollmann, M., Schwaiger, K., Böckmann, R. A., & Pischetsreder, M. (2021). Generation of antimicrobial peptides Leg1 and Leg2 from chickpea storage protein, active against food spoilage bacteria and foodborne pathogens. Food Chemistry, 347, 128917 Mota-Gutierrez, J., & Cocolin, L. (2021). Current trends and applications of plant origin lactobacilli in the promotion of sustainable food systems. Trends in Food

Mola-Guterrez, J., & Cocoin, L. (2021). Current trends and appreations of plant origin factobactin in the promotion of sustainable lood systems. Frends in Pood Science & Technology, 114, 198-21









#### Edible coating of fresh-cut fruits by utilizing natural olive-oil by-products extracts as additive

#### **AYESHA IFTIKHAR**

Department of Agricultural, Environmental and Food Sciences, University of Molise, 86100 Campobasso, Italy

#### Introduction [1]

Fresh-cut fruit can be defined as "any fruit that has been physically altered from its original form (trimmed, peeled, washed and/or cut), but remains in fresh



Figure 1\_Change in quality and safety of fresh-cut fruits

#### Role of edible coatings [2]

Edible coating can be use in fresh-cut industry as an emerging technology to reduce the deleterious effects that minimal processing imposes on intact fruit and its tissues. An edible coating is defined as a thin layer of edible material applied to the surface of food products to perform the tasks enlisted in Fig. 2



Figure 2\_Importance of edible coating

#### Edible Coating Materials [3]

The composition of edible coatings is categorized based on the biomolecule used. That include polysaccharides, proteins, lipids, composite materials (i.e., blends of materials), and agro-industrial waste as described in Fig 3. Moreover, additive, and active compounds can be incorporate in the edible coatings with different function, such as antioxidant and antimicrobial as described in Fig 3.



Figure 3 Edible coating formulation materials

#### Supervisor: Prof. Antonella De Leonardis





## The role of HY in human health has been investigated in numerous studies evidencing several bioactivities (antioxidant, cardioprotective, anti-

Edible Coatings enriched with Natural Extracts from Olive-Oil

Oxidative stability in minimal processed food such as fresh-cut fruits is one of

the main parameters which effect the integrity and nutritional properties of dietary constituents. Recent increase in attention towards zero waste concepts

urges the researchers to turn the by-products and industrial wastes in natural

In this context OOBP has been tested as a valuable source of bioactive

compounds, especially hydroxytyrosol (HY) and its derivates (Fig. 4).



By-Products (OOBP) [4,5]

antioxidants

#### Figure 4\_Molecular structure of free hydroxytyrosol

#### Method of Application

The technique or way use for the coating or film adhesion to the food surface is briefly described in Fig 5.

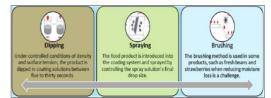


Figure 5 Coating Application Methods

#### Conclusion

Edible coatings worked as a potential vehicle to carry the bioactive compounds and along with improving the physiochemical properties (colour, texture, firmness, total soluble solids), sensory characteristics it also may have health benefits, with potential application in biomedical fields. Release of bioactive compounds in edible layer of coating could also improve the natural antioxidant present in fresh-cut fruits.

Edible coatings enriched with olive by-products extracts are source of higher valuable bioactive compounds especially phenolic compounds (hydroxytyrosol and derivates), due to having highest stretching capacity, resistance to breakage it would also improve structural, functional properties of fresh-cut fruits. Thus, olive extracts avidinated to be use as active ingredients in edible coatings and avoid deterioration during transformation, packaging, and storage

#### **References:**

[1] Yousuf, B., Deshi, V., Ozturk, B., & Siddiqui, M. W. (2020). Academic Press, 1-15 [2] Apicella, A., Adiletta, G., Di Matteo, M., & Incarnato, L. (2019). Chemical Engineering Transactions, 75, 85-90. [3] Díaz-Montes, E., & Castro-Muñoz, R. (2021). Foods 2021, 10, 249.

[4] Bermúdez-Oria, A., Rodríguez-Gutiérrez, G., Vioque, B., Rubio-Senent, F., & Fernández-Bolaños, J. (2017). Carbohydrate polymers, 178, 368-377.

[5] Luzi, F., Pannucci, E., Clemente, M., Grande, E., Urciuoli, S., Romani, A., ... & Santi, L. (2021). Molecules, 26(7), 2104.



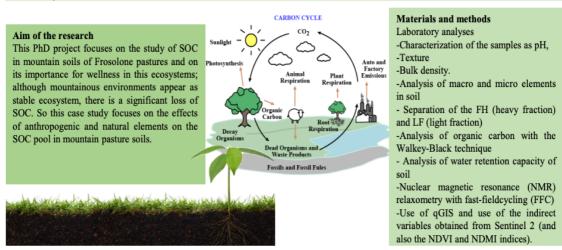


### **CARBON FORMS AND DYNAMIC IN MOUNTAIN SOILS**

INTERNATIONAL

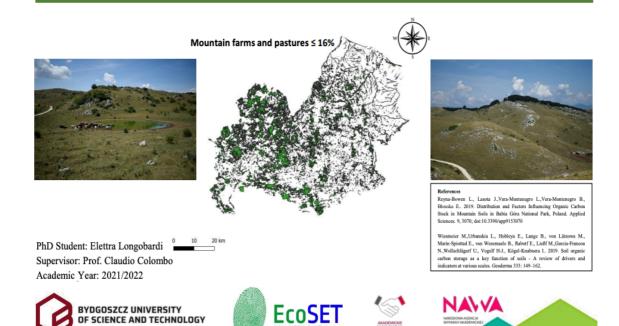
#### Introduction

The main ecological functions of soils, beyond technical and cultural aspects, are: biomass production, storage and filtration of water, storage and recycling of nutrients, habitat for biological activity and carbon storage (Wiesmeier M. et al. 2019). Soil carbon stock is influenced by several environmental factors, such as topography, slope, exposure, elevation, climate, parent material, and vegetation. Site properties (altitude, exposition, slope) influence the soil conditions and soil properties. Mountain areas are characterized by climatic and topographic factors that are highly variable, and this variation results in the diversification of vegetation and, as a consequence, differences in soil properties among locations. Mountain soils are highly dynamic systems that are sensitive to environmental changes (Reyna-Bowen L. et al. 2019)



#### Expected results

We expect the SOC (and therefore the fundamental carbon reserve) evolved as a result of natural and anthropogenic pressures. The increase in temperature has accelerated the process of soil erosion and decreased the amount of water present in soil. Moreover, bad management of pasture, rain distribution and decrease of the heads of cattle, has altered the macro and micro molecular components present in the soil. As a result also the vegetation and therefore the biodiversity of the area has definitely changed and has altered.







## Effects of *In Ovo* Injection of Synbiotics on Growth, Intestinal Microbiota, Short-chain Fatty Acids and Meat Quality of Broiler Chickens

Meng Peng

Department of Agricultural, Environmental and Food Sciences University of Molise, 86100 Campobasso, Italy

#### Introduction

It is imperative that today's poultry industries be proactive in improving animal health and growth, and the safety of poultry products in a sustainable way. Probiotics, prebiotics, and synbiotics are proposed as natural and safe alternatives to antibiotic growth promoters (banned from 2006 in EU) in order to solve the intestinal problems of birds through the modulation of the composition and function of the intestinal microbiota, therefore improving health and performance of birds.

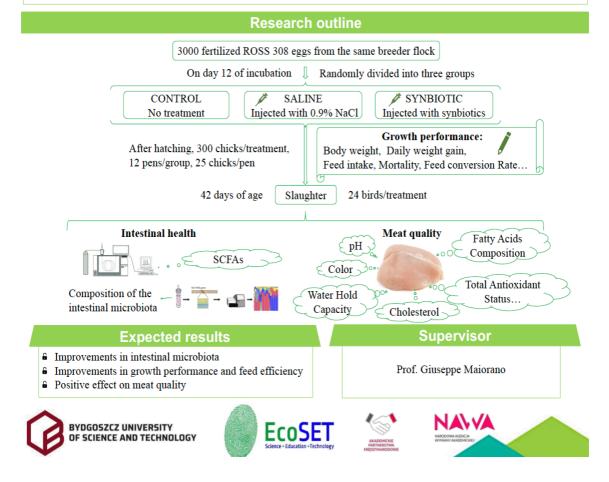




*In ovo* stimulation refers to the delivery of prebiotics or synbiotics in the early-stage embryo (day 12 of egg incubation). The main concept of in ovo technology is to apply substances long before the bird hatches, which helps to program lifelong phenotypes (e.g., immunity, gut microbiome, performance, adaptive) already during the embryonic phase.

### **Objectives**

To evaluate the effects of *in ovo* injection of synbiotics on growth, intestinal microbiota, short-chain fatty acids and meat quality of broiler chickens.



The Scientific and Organizing Committees of INTERNATIONAL PhD STUDENT CONFERENCE: Research and Innovation in Agricultural, Livestock and Food Sciences

gratefully acknowledge the referees for their significant contribution.

This material has been supported by the Polish National Agency for Academic Exchange under Grant No. PPI/APM/2019/1/00003

Publisher: University of Molise Edition: First Number of copies: 50

ISBN 9788896394373