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## **L'Unione Agricoltori e Coltivatori Diretti Sudtirolesi**

L'Unione Agricoltori e Coltivatori Diretti Sudtirolesi è uno dei maggiori organi di rappresentanza del ceto contadino di tutta la provincia. Sono infatti oltre 21.000 i membri che l'Unione rappresenta sostenendoli con un servizio mirato e globale.

In qualità di portavoce del ceto contadino nei rapporti con le autorità, i sindacati e le altre organizzazioni economiche, l'Unione Agricoltori persegue l'obiettivo di rafforzare la posizione del ceto contadino in ambito economico, sociale, culturale e politico.

Per tutelare e rappresentare il comparto agricolo in ambito politico, l'Unione Agricoltori invia i propri portavoce presso le istituzioni a livello comunale, comprensoriale, provinciale, nazionale ed europeo. L'organizzazione interna dell'Unione prevede una presenza capillare su tutto il territorio e si concretizza attraverso le varie associazioni contadine a livello locale, comprensoriale e provinciale, strettamente collegate tra loro da una forte sinergia e obiettivi condivisi.

Le sedi dell'Unione Agricoltori sono dislocate in tutta la provincia con uffici a Bolzano, Silandro, Merano, Egna, Bressanone, Brunico e Vipiteno. Con oltre 270 collaboratori l'Unione Agricoltori rappresenta un importante datore di lavoro.

### **Organizzazione**

Grazie a ben 156 gruppi locali presenti in 116 comuni, vanta sul territorio una diffusione capillare come nessun'altra organizzazione rappresentativa.

Oltre 1.100 funzionari contribuiscono con la loro preziosa attività a livello locale. Circa 90 operano invece a livello comprensoriale. Il Consiglio Provinciale degli Agricoltori (Landesbauernrat) è composto da 20 membri.

### **Consulenza**

L'Unione Agricoltori e Coltivatori Diretti Sudtirolesi offre ai propri membri, in sede e negli uffici comprensoriali e di area, un pacchetto di consulenza e assistenza in diversi ambiti quali fisco, diritto, lavoro e retribuzione, sicurezza sul lavoro, sociale, finanziamenti, imprenditoria, previdenza sociale, marketing e formazione. Alcuni reparti svolgono una funzione speciale:

#### ➤ *Reparto Innovazione ed energia*

Per i contadini con idee innovative, l'Unione Agricoltori e Coltivatori Diretti Sudtirolesi ha creato uno sportello dedicato, i cui collaboratori forniscono consulenza e assistenza a chi è interessato a sviluppare le proprie idee, fino al lancio sul mercato. Allo stesso tempo, il reparto è il primo referente per questioni relative alla produzione di energia da fonti rinnovabili e all'utilizzo efficiente dell'energia.  
Contatto: Tel.: 0471 999363; E-Mail: [innovation-energie@sbb.it](mailto:innovation-energie@sbb.it)

#### ➤ *Ufficio Sostenibilità*

Per raggiungere gli ambiziosi obiettivi nel campo della sostenibilità in agricoltura e in particolare nei settori del vino, della frutta e del latte, l'Unione Agricoltori Sudtirolesi ha fondato l'Ufficio Sostenibilità. I compiti principali sono l'implementazione di progetti modello e il networking tra gli stakeholder e gli agricoltori, la comunicazione e la verifica scientifica dei progetti.

Contatto: Tel.: 0471 999359; E-Mail: [nachhaltigkeit@sbb.it](mailto:nachhaltigkeit@sbb.it)

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## Transizione agroecologica e giovani ricercatori: una grande opportunità da non mancare

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Pandemia, cambiamento climatico, perdita di biodiversità: nel recente passato, mai come adesso l'umanità sta subendo gli effetti negativi di più crisi concomitanti di importanza planetaria. Questa situazione sta facendo emergere la consapevolezza che tutto è collegato e che siamo tutti parte di un grande sistema su cui abbiamo un forte impatto potenziale e reale, nel bene e nel male. Ispirata dai 17 obiettivi di sviluppo sostenibile per il 2030 delle Nazioni Unite, la Commissione Europea ha puntato sulla transizione ecologica come leitmotiv del proprio programma di sviluppo e rilancio economico. Declinata in ambito agro-alimentare, la transizione agroecologica si pone l'obiettivo di trasformare in un'ottica di vera sostenibilità i sistemi di produzione, trasformazione, commercio e consumo delle derrate agricole e degli altri prodotti delle aziende e dei territori. Questa grande sfida, che non può essere più rinviata, presuppone che tutti gli attori del sistema facciano la loro parte, in maniera coordinata. In questo ambito, grandi sono le aspettative e le opportunità per il mondo della ricerca. Tuttavia, la domanda cruciale da porsi è: il sistema ricerca, con particolare riferimento a quello italiano, è pronto ad accogliere queste sfide? A quali temi dovrebbe dare priorità e con quali modalità? Di cosa c'è bisogno – oltre ai fondi, che certamente non mancheranno – per supportare il cambiamento con evidenze scientifiche certe e commisurate alla complessità dei problemi? Che tipo di giovani ricercatori dovremo formare? Nelle more della tradizionale ritrosia del mondo accademico e della ricerca, soprattutto italiano, ad abbandonare lo status quo, la situazione richiede visione, coraggio e motivazione, ma può aprire la strada ad un rinascimento dei ricercatori e del ruolo della ricerca nella società. Verranno presentati alcuni esempi di approcci e temi di ricerca in linea con la transizione agroecologica, con l'obiettivo di stimolare la discussione e far passare il messaggio che non c'è contraddizione tra gli obiettivi di ottenere una produzione scientifica di alto livello e di fare ricerca fortemente orientata al mondo reale.

## New Plant Breeding Techniques (NPBTs) for a sustainable and resilient agriculture

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The ongoing climate changes expand both biotic and abiotic stress conditions forcing plant breeders to select genotypes resistant to water and thermal stresses while coping with an increasing pressure from pests and pathogens. These unfavourable constraints are leading to insufficient yield and a strong decrease in food and feed quality features threatening the food security levels. The development of genetically improved crops has long been taking advantage of crossings and mutagenesis to improve yield and quality features, as well as stress resilience traits. In addition, over the last century, genetic engineering and biotechnologies have broadened the toolbox of geneticists and breeders with new instruments and approaches, leading to the creation of genetically modified organisms (GMOs). Thanks to these techniques, the gene pool available to plant breeders has considerably increased, allowing the transferring of genes to crops from sexually incompatible plant species as well as from other organisms. Although in 2018 GM crops covered 191.7 million hectares with remarkable benefits, their use is still associated with a strong public concern. To overcome the limits associated to GM crops many techniques have during the years been developed up to the latest New Plant Breeding Techniques (NPBTs). These approaches allow a single gene to be transferred, mimicking sexually compatible crosses (cisgenesis) and precise modification of specific DNA sequences (genome editing). In conclusion, a wide range of techniques are becoming mature for substituting GMOs and supporting traditional breeding, with a realistic possibility of being largely accepted by the international community. Several techniques do not leave any traces in the final product, leading to either individual identical to the previous generation from the genetic point of view, but with a single improved phenotype, or even untouched genome, controlled by external molecules, adsorbed and then removed by the plant itself.

# Impiego di antibiotici nell'allevamento suinicolo nazionale: fattori di rischio e prospettive di riduzione

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Gli antibiotici sono uno strumento fondamentale per la medicina veterinaria, mantenerne il potenziale terapeutico è di importanza critica per garantire la salute ed il benessere degli animali. Inoltre, nel contesto delle produzioni zootecniche, queste sostanze sono importate per salvaguardare la salute pubblica e la sicurezza alimentare, contrastando la diffusione di agenti batterici zoonotici e dei *foodborne pathogens*.

In zootecnia, nel corso degli anni si sono consolidate pratiche d'impegno non appropriate degli antibiotici che in condizioni specifiche hanno provocato una pressione selettiva che ha causato la crescente diffusione di patogeni resistenti agli antimicrobici. Con la pubblicazione nel 2011 del "Piano d'azione contro le crescenti minacce della resistenza agli antimicrobici", la Commissione Europea ha iniziato il percorso che a tutt'oggi ha l'obiettivo di ridurre l'impegno degli antibiotici terapeutici in medicina umana e veterinaria. I dati disponibili mostrano che in media il 70% degli antibiotici venduti viene utilizzato negli animali ad uso zootecnico e nel ranking europeo l'Italia insieme a Cipro e Spagna risultano essere i paesi in cui la vendita di antibiotici per animali da reddito è maggiore. Il suino sembra essere il settore a maggiore uso di tali molecole, rendendo evidente la necessità di azioni specifiche per questo settore. Oggi vi è un ampio consenso all'interno del settore zootecnico sull'obiettivo di ridurre l'uso di questi farmaci al fine di migliorare la sostenibilità della produzione sotto diversi aspetti: i. sociale (accettazione dei consumatori e salute umana); ii. ambientale (ridurre la diffusione di antibiotici e AMR); iii. economico (riduzione delle perdite di produzione) sostenibilità. Con la pubblicazione del documento 2015/C299/04, la Commissione ha rafforzato il suo impegno per l'uso responsabile degli antimicrobici in medicina veterinaria evidenziando che l'approccio dell'uso appropriato deve essere sinergico con interventi che prevengono l'insorgenza e la diffusione delle malattie. Inoltre, il rinnovo nel 2017 della strategia UE contro l'AMR, la nuova categorizzazione dei farmaci ad uso veterinario e l'entrata in vigore nel 2022 del nuovo regolamento sull'uso dei farmaci in medicina veterinaria (Regolamento 2019/6) indicano un forte impegno della UE per contrastare l'insorgenza di nuova AMR al fine di preservare sia la salute pubblica sia quella animale.

# Hunted wild game meat consumption in the modern era: a systematic review

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Hunted wild game meat (HWGM) possesses several positive characteristics. In fact, it derives from non-farmed animals, which implies a low environmental impact compared to farmed meat and an optimal nutritional profile. However, HWGM is the product of hunting, which often relates to consumers' ethical concerns and might introduce food safety risks in case HWGM is not slaughtered properly.

The body of literature focused on consumer perception and attitudes towards HWGM growth in the last two decades, but it is fragmented. Consequently, we present a literature review carried out with a systematic approach. The purpose of this study is to understand which variables are related to consumer perception and attitudes toward HWGM.

Based on an initial pool of 2,558 articles, following a rigorous selection approach, this study analyses the content of a final pool of 25 articles. Three categories of variables were identified: socio-demographic variables, supply chain-related variables, and product-related variables. Our findings clearly suggest that among the sociodemographic variables, gender and residence play a crucial role in defining consumers' perceptions and attitudes towards HWGM. Moreover, some positive drivers have been detected. A positive attitude towards hunting and familiarity with hunting are linked to HWGM consumption. Conversely, as expected, food safety concern represents one of the main barriers to HWGM consumption. Finally, results suggest that the seasonality of the product and the relative lack of HWGM market supply may constitute barriers to consumption. Our findings could assist stakeholders in the development of targeted marketing strategies and policies.

# Extending the Theory of Planned Behaviour (TPB) to explore online wine tourism intention among wine consumers

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While the Covid-19 pandemic and the related mobility restrictions brought the wine tourism sector to a standstill, they also push for innovation to improve its resilience. Interestingly, several wineries adopted new strategies like online wine tourism experiences (OWT), gaining increasing attention on the supply and the demand side and the interest of academics.

However, the research efforts on the demand side are still limited and represent the focus of this research. The study applies Azijen (1991)'s Theory of Planned Behaviour (TPB) to analyze the antecedents of the intention to participate in OWT experiences on a large sample of regular wine consumers in Italy. Data were collected through an online survey at the beginning of 2022.

If further tests an extended TPB model including wine involvement, attitude towards technology, risk attitude, and covid phobia, in addition to the common TPB antecedents (i.e., attitude, subjective norms, perceived behavioural control). Notably, wine involvement is acknowledged as a critical antecedent of wine tourism intention. Also, attitude towards technology is essential given that OWTs are delivered through apps and online platforms. Finally, the inclusion of risk attitude and covid phobia is critical to provide a thorough outline of the phenomenon while quantifying the circumstantial effect of the Covid-19 pandemic.

Results provide strategic information to wine tourism operators and policy-makers to further develop OWTs' offer. Moreover, they can be generalized to the Italian population due to the large and representative sample (in terms of age, gender, and geographical place of residence) used for the analysis.



## Organic label and consumer wine choice: the effect of brand equity

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One of the goals of the European Union's "Farm-to-Fork" strategy is to promote organic farming for reaching at least 25% of agricultural land under organic. In Italy, the organic vineyard surface area is increasing, counting in 2020 around 18% of the total vineyard.

About consumers' preferences for organic wine, in literature contrasting results are emerged. This study wants to further investigate the impact of the organic label on the consumers' purchasing behaviour, considering the wine brand equity. In other words, we assume that organic claim has a different impact in terms of quality perception and purchase behaviour if the wine is labelled by a high or low-equity brand.

We have conducted a 2 (brand equity: low, high) × 2 (organic label: present, absent) factorial experiment with a total of four treatments. A high-equity and a low-equity Denominations of Origin (DO)s, which represent collective brand for wine, were chosen after interviews with some wine experts. Then, we administrated a questionnaire on a sample of Italian wine consumers. Each participant to the experiment was randomly assigned to one of the four treatments. A manipulation check was implemented to determine if participants correctly perceived the difference of brand equity among the two DOs chosen. To test our hypothesis, we have measured, for each product, the individual perceived quality, the perceived taste and the behavioural intention, namely the purchase intention and the willingness to pay. The data were analysed by repeated measures of ANOVA and MANOVA.

Data collection and analysis are in progress, and the results will be presented during the conference. How organic label has an impact according to different levels of brand equity may have interesting implications for the stakeholders involved.

## **Valorization of slurry to increase efficiency and sustainability of dairy farm**

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In the last 30 years, mineral fertilizers had advantageous prices leading to an overuse and to less attention to the real uptakes of the crops. In dairy farms, the supply of very high doses of fertilizers, both organic and mineral, has enriched the soil with organic matter and nutritional elements such as phosphorus and potassium. Therefore, to increase the fertilization efficiency is possible to hypothesize the simple return of the removals, which for phosphorus and potassium can be compensated with only organic fertilization.

A four-year trial was planned on two fields of a dairy farm in Northwest Italy to verify the effect of fertilization on the production of high moisture corn (HMC). Each field was divided into four plots and two different fertilization treatments were considered: conventional (CONV, 260 kg/ha of potassium chloride, 210 kg/ha of diammonium phosphate and 240 kg of slow-release urea) and calibrated (CAL, 90 kg/ha of diammonium phosphate and 100 kg/ha of urea), on each treatment 170 kg/ha of N, 51 kg/ha of P<sub>2</sub>O<sub>5</sub> and 198 kg/ha of K<sub>2</sub>O were added as farm slurry. The dry matter (DM) yield, HMC mineral contents and nutrient balances were assessed.

The DM yield showed no differences between the two fertilizations in all the years (15.5 and 15.6 t DM/ha for CONV and CAL). The nutrients uptake of HMC were not different between the two fertilization and was on average 140, 105 and 104 kg/ha of N, P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O, respectively. The fertilization efficiency increased from 0.44 to 0.57 for N, from 0.79 to 0.98 for P<sub>2</sub>O<sub>5</sub> and from 0.30 to 0.51 for K<sub>2</sub>O. The reduction of mineral fertilizers led to an economic saving of over 150 €/ha which became 400 €/ha by projecting the calculations to the current year given the high prices of fertilizers. For dairy farms the valorisation of slurry and the calibration of fertilization can enhance their economical sustainability while contributing to reduce the risk of nutrient losses.

## Effect of the cultivation substrate on production and quality of *Pleurotus ostreatus*

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*Pleurotus ostreatus* is the second most cultivated mushroom in Italy and it's characterized by a less controlled cultivation system which is more influenced by environmental and management conditions.

The experiment (YESP Project – Veneto Rural Development Program 2014-2020; PON Green Topics – action IV.5) evaluated the effect of the most used cultivation substrates in Italy and Spain in order to assess their productive and qualitative performance. Four cultivation substrates (A, B, C and D) inoculated with the same mycelium (P80 – Italspawn) were considered; three of them were representative of the whole national offer (A, B and C) and 1 of Spanish origin (D). The number of primordia and the production of each treatment were recorded together with the morphological characteristics of fruiting bodies and their organoleptic parameters.

The results obtained from this experience showed, especially for the first harvest, a significant effect of the substrate in the timing emergence of the primordia and a high percentage of primordia (78-88%) out of the total available holes per bag was found for substrates A and B. The average diameter of the fruiting body was higher for substrate B, whereas no significant differences were observed for carpophore thickness. Overall, the production per bag was between 4 and 5 kg with some variation among substrates. About the comparisons of substrate use efficiency, the production per kg of substrate was higher in D with over 0.2 kg. Concerning qualitative traits, antioxidant capacity during the first flow was higher in substrates A, B and D with an average result of 2570 mg Fe<sup>2+</sup> eq kg<sup>-1</sup> dw. The total polyphenols content was higher in the mushrooms of the third flow cultivated in D (2004 mg GAE kg<sup>-1</sup> dw).

## The sustainability of agricultural residues valorization for bioenergy production

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Environmental policies have provided significant benefits in Europe over recent decades. However, persistent problems in resource use, climate change impacts and environmental risks need further efforts to be addressed in the near future. Among renewables, biomass for energy continues to be the main source of non-fossil energy in Europe. However, biomass must be produced, processed and used in a sustainable and efficient approach. Although energy production from agricultural residues is perceived as an eco-friendly and non-fossil alternative, it might generate significant environmental loads. Thus, the aim of this study was to assess the energy and environmental burdens of producing bioenergy from agricultural residues.

A life cycle assessment approach was used to analyze the supply chain and the sustainability of using agricultural residues from the Sardinia region (Italy). The boundaries of the system were set from "Field to Wire" which means from on-field collection to biomass-fired power plant. The agricultural residues investigated were cereal straws, grape and olive prunings and artichoke by-products. The results indicated a total potential energy availability of 3,772 TJ per year. The energy and environmental impacts associated with the agricultural residues supply chain showed an average requirement of primary energy of 790 MJ t<sup>-1</sup> and a related emission of 51.7 kg CO<sub>2</sub>e t<sup>-1</sup>. Considering the whole supply chain, the average primary energy required to generate one MWh of electricity accounted for 1,127 MJ, ranging from 884 to 1,472 MJ MWh<sup>-1</sup>. Analyzing the emissions generated from field to wire, the study assessed an average emission rate of 85 kg CO<sub>2</sub>e MWh<sup>-1</sup>. Producing bioenergy from agricultural residues is recognized as a sustainable practice but it might cause noteworthy environmental impacts.

This study identified the main critical points of the agricultural residues supply chain while highlighting the whole energy and environmental sustainability of the process.

## Sea urchin waste valorization in egg production in a circular economy perspective

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In the circular economy framework, the project "BRITEs - Byproduct Recycling: Innovative TEchnology from the Sea" aims at completely recycling and valorise a food by-product, namely sea urchin wastes originating from food industry (restaurants and seafood enterprises) and transforming them into innovative products, including alternative calcium and antioxidant rich supplement feed for animals.

In this context, this study investigates the environmental impact of egg production under two different production scenarios: in the baseline laying hens are fed with the widespread calcium carbonate to support eggshell production; in the alternative one this was replaced with sea urchin waste. To carry out this comparison the Life Cycle Assessment was applied, with 1 kg of eggs considered as Functional Unit and a from "cradle to farm gate" approach. Therefore, all the processes at the poultry farm were considered (hatchery, feed production, energy and other materials consumption, emissions related to feed cultivation and animal rearing) while packaging and distribution of eggs were excluded, because not directly affected by the use of sea urchin wastes. Inventory data were collected in poultry farms of different size mainly located in Northern Italy. A full set of environmental indicators was evaluated using the Recipe Midpoint characterization methods. The results highlight how the substitution of calcium carbonate involves a slightly impact reduction for all the evaluated impact categories. Environmental benefits arise from the replacement of the sea urchin waste common management in the alternative scenario. This benefit is small on the egg life cycle because the impact of calcium carbonate itself is limited in the baseline scenario. However, the alternative management presents interesting prospects, especially if conceived on a large scale. However, a trade-off between environmental impacts can occur if the transport distance of sea urchin waste is not taken carefully into account.

## **Importance of fish traceability and familiarity with fish sustainability labels: evidence from Italy and Spain**

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Fishing provides important income and commercial opportunities in Italy and other Mediterranean countries. However, there are still several issues related to traceability, environmental protection, and consumers' awareness of fish sustainability.

This study aimed to investigate fish traceability perception and familiarity with the MSC fish sustainability label. An online survey was filled in by 2000 consumers, involved in both Italy and Spain. The survey was structured into different sections: 1) fish consumption habits; 2) traceability and sustainability perception (including the importance of the fish traceability and the familiarity with the MSC label); 3) psycho-attitudinal traits (Food Neophobia scale and Food Choice Questionnaire); 4) social-demographic characteristics (age, gender, living area, etc.). Subjects were grouped for provenance (two different provenances, Italy and Spain), gender and age (four age groups), for a total of 16 observations. A Partial Least Square (PLS) regression model was used assuming the fish traceability and MSC familiarity for each group of subjects as response variables and 38 explanatory variables. First, consumers were well separated in terms of nationality and age. Secondly, the interest in fish traceability increased with the increasing age. On the contrary, young consumers resulted more familiar with the MSC label than adult/old ones. Consumers who were interested in fish traceability were also concerned about food ethical aspects and food natural content. Moreover, MSC familiarity was mainly affected by the living place (consumers who lived near the sea resulted more familiar with the MSC label than those living in the internal areas) and the purchase place (consumers who declared to mainly buy at the fish wholesale and fish market resulted more familiar with the MSC label than others).

In conclusion, the importance of sustainability is not fully clear for all consumers. Therefore, dissemination strategies are still needed to make aware the consumers of the fish sustainability aspects.

## Nanotechnology-based agrochemicals for the control of *Fusarium* spp. disease and boost bread wheat growth and immunity

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*Fusarium* are cosmopolitan fungal pathogens for wheat, causing Fusarium head blight (FHB) and crown rot (CR). *Fusarium* diseases are managed by the application of synthetic fungicides showing negative or unknown side effects on the environment. Thus, there is an urgent need to find novel, efficient and bio-based solutions to mitigate plant diseases. Nanotechnology-based agrochemicals are promising, since cellulose nano-crystals (CNCs) can be obtained by plant wastes and employed as green nano-carriers to deliver active molecules. High-amylose starch (HAS) consisting of a high content of amylose, can act as excipient by increasing the solubility of active compounds. We extracted CNCs and HAS from wastes of the bread wheat cultivar Cadenza high-amylose obtained by Targeting Induced Local Lesions IN Genomes (TILLING) and characterized by mutations in the *starch synthase II (Sgp-1)* and *waxy proteins (Wx)* genes, thus accumulating high-amylose starch. Chitosan and gallic acid were assayed as active molecules. *In vitro* experiments individuated the optimal active concentration in order to formulate a bio-based composite of CNCs, HAS, chitosan (25% w/w), and gallic acid (2.5% w/w). The bio-based composite (tested *in vivo* at 2%) displayed biostimulant properties by enhancing the % of seed germination, the Nitrogen Balance Index (NBI) values and the dry biomass of wheat plantlets. Artificial inoculation of *Fusarium culmorum* (causing CR) and *Fusarium graminearum* (causing FHB) after a pre-treatment of the bio-composite drastically reduced the symptomatic progression of *Fusarium* diseases, compared to conventional fungicide (tebuconazole). Furthermore, the accumulated fungal biomass and mycotoxins into the infected tissues as much as the grain yield will be evaluated to further validate the effect of such bio-based composite on disease management. Further research will be focused on analysing the gene expression of several disease resistance responsive genes in relationship to the application of the bio-based composite to validate its elicitor-like properties.

## Low intensity magnetic fields enhance growth and disease protection in tomato plants

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In plant defense, priming leads to a physiological state in which plant responds faster and/or more accurately to a future (a)biotic attack. This physiological state can be induced by beneficial microbes, by treatment with natural or synthetic compounds and by exposure to abiotic agents. The aim of our work was to test the low-intensity magnetic field as priming agent in tomato (*Solanum lycopersicum* L.) young plants challenged with the necrotrophic fungus *B. cinerea*. To this end, we designed and manufactured a Helmholtz-Coils device able to generate a rather constant magnetic field in a region large enough to cover the whole plant. Experiments were carried out by exposing plants to a flux density field of 1 mT for 10 minutes. Treated plants showed increased resistance against the pathogen than control plants, highlighting a potential role of magnetic fields as a priming agent. In particular, we observed reduced necrotic lesions, decreased reactive oxygen species (ROS) generation and increase activity of ROS scavenging enzymes in leaves of plants exposed to magnetic fields. Additionally, we observed that magnetic fields have beneficial effect on plant growth and health. To obtain a general overview about changes induced by magnetic fields, -omics analyses are underway. The outcome will be crucial for understanding the molecular mechanisms significantly altered by magnetic fields towards disease protection and fitness enhancement.



## LED light supplementation in strawberry (*Fragaria × ananassa*) plants: leaf physiological responses and fruit resistance to pathogens

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Supplementation of narrowband light can promote plant development and stimulate targeted secondary metabolism branches. Secondary metabolites can play a key role in plant defence mechanisms by enabling plants to deal with subsequent pathogen infections.

This work offers new insights on the effects of greenhouse light supplementation applied with light emitting diodes (LEDs) on physiological traits of *Fragaria × ananassa* leaves, measured after 1 (T1) and 17 (T17) days of treatment. A possible light priming effect on fruit against pathogens was also investigated after one month of light treatment. Monochromatic red (R), green (G), blue (B), and polychromatic white (W—R:G:B; 1:1:1) LED lights ( $250 \mu\text{mol photon m}^{-2} \text{s}^{-1}$ ), were applied as supplementation to ambient light. At T1, W, B and G lights supplementation induced an enhancement of non-photochemical quenching (qN) and de-epoxidation state of xanthophylls. However, no changes in maximal photosynthetic rate ( $P_{N_{\max}}$ ) were observed in any treatment. At T17, xanthophyll content increased only under R and G lights (+38 and +53% respectively), where qN resulted lower than control, and flavonoid levels raised under B light (+20%).  $P_{N_{\max}}$  was promoted under W light and depressed under B and R lights. However, W, B and R lights stimulated superoxide radical formation. G light did not affect photosynthetic traits nor oxidative burst.

Overall, none of the light treatments induced visible injury. Then, ripe strawberry fruits were harvested and inoculated with *Botrytis cinerea*, and the disease severity was monitored every 12 h. Between 36 and 48 h post inoculation, fruits grown under R light showed a significant slackening of infection development (up to -27%), as confirmed by the reduction of the area under the disease progress curve. Thus, it is conceivable that R light promoted the biosynthesis of specific secondary metabolites in strawberry fruits which translated to a higher resistance to the pathogen.

# **Assessing the effectiveness of sustainability and quality attributes in consumer selection of a wood waste heating product**

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The greater attention of consumers to environmental issues finds in certified wood products a feedback able to convey valuable elements, overcoming in part the problem of information asymmetry. The most widespread forest certification schemes in the European market are PEFC and FSC, representing a vehicle for meeting consumer demands for sustainability. Energy products have also efficiency and quality aspects, such as caloric values and ash residues, which are identified by quality certifications such as ENplus. Our study aims to assess the importance that consumers attach to sustainability and quality certifications when purchasing wood energy products in particular "pellets": a by-product generated from wood waste, therefore a virtuous example of circular economy. Another objective is to identify homogeneous groups of consumers based on their individual utility pattern. A multi-section online survey was administered to collect data. A ranking conjoint experiment was developed to assess the importance of pellet attributes: price, sustainability and quality in terms of mean relative importance and utility. Following the conjoint, a cluster analysis was performed and three groups were identified. Mean relative importance results indicate that price is the most important aspect, followed by sustainability and quality certification. Regarding the utility estimates, low-price products provided a higher utility to consumers. FSC and PEFC showed similar utility suggesting that they have a similar role in product differentiation. ENplus certification provided high utility indicating that consumers are interested in quality attribute of heating products. Cluster analysis provided a deeper understanding of consumer preferences identifying three segments: price-sensitive, sustainability-oriented and quality-oriented consumers.

The study indicated that quality and sustainability certifications can be considered useful tools to better market pellet products. However, the role of sustainability certifications does not seem to be fully expressed, suggesting that through consumer awareness actions, these certifications could ensure higher remuneration for wood products.

## Detection of winterkill events of white mustard (*Sinapis alba* L.) by satellite-based remote sensing

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Cover crop management mainly consists of sowing and termination, whose timing affects the agro-ecological services provided by cover crops. Thus, identifying the correct time windows is crucial for field management. Autumn-winter cover crops can be terminated by frost damage (winterkill termination) if they are exposed to sub-zero temperatures exceeding their frost tolerance. Assessing winterkill occurrence frequencies allows comparing scenarios of crop management options for farms or regions, thus supporting the informed choice of cover crop species and sowing dates. A few studies regarding cover crop species (*Sinapis alba* L. specifically) frost tolerance are reported in the literature and those present consisted of local indications. Satellite-based remote sensing is suited for the estimation of the effects of biotic and abiotic crop stressors from field to regional scales. However, a few studies addressed the monitoring of frost damage and none of them on cover crops. The study aimed to develop a relationship between seven different vegetation indices values and winterkill occurrence. A remote sensing monitoring campaign was carried out, using satellite images retrieved from Sentinel-2, during the 2021-2022 autumn-winter season in six commercial farm fields of the Lombardy region. Field measurements of aboveground biomass and visual assessment of frost damage symptoms were performed to validate satellite data. Significant ( $p < 0.05$ ) relationships were detected between the ground-measured frost damage index and the vegetation indices (following She et al., 2015): EVI ( $R^2 = 0.65$ ), MSR ( $R^2 = 0.67$ ), NDRE ( $R^2 = 0.57$ ). Furthermore, the procedure developed by Zhao et al. (2020) was adapted to identify the timeframes during which a decrease of the vegetation index value can be accounted for a frost damage event. The timeframes obtained through this procedure were consistent with field observation opening the possibility of using satellite images to support the field management of autumn-winter cover crops on a regional scale.

## Between light and selective shading: morpho-biochemical and metabolomics insight into the effect of blue photosensitive shading on nursery seedlings

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High nursery densities reduce seedling quality due to the competition for light. High light intensity, shading, and blue light depletion activate morpho-physiological and metabolomic responses in plants, resulting in size modification to gain an advantage over neighboring plants. Our research aimed to unravel the effects of light intensity and quality on nursery seedlings at the morphological and biochemical levels. The effect of black shading and blue photosensitive shading nets were investigated in terms of morphometric, ionic, and untargeted metabolomics signatures in *Cucurbita pepo* L., *Citrullus lanatus* L., *Solanum lycopersicum* L., and *Solanum melongena* L. seedlings. Plant height, diameter, sturdiness index, leaf area, specific leaf area, shoot/root ratio, and mineral content were evaluated. In *Cucurbita pepo* L. and *Citrullus lanatus* L. the blue net reduced the shoot/root and chlorophyll a/b ratios and increased stem diameter and total chlorophyll. The black net increased plant height, stem diameter, and sturdiness index in *Solanum lycopersicum* L. and *Solanum melongena* L. Unshading conditions reduced leaf area, specific leaf area, shoot/root ratio, and total chlorophyll. The blue net improved the sturdiness index and quality of *Cucurbita pepo* L. and *Citrullus lanatus* L. Such impact on morphological parameters induced by the different shading conditions was corroborated by a significant modulation at the metabolomics level. Untargeted metabolomics phytochemical signatures of the selected plants and the subsequent multivariate analysis coupled to pathway analysis, allowed highlighting a broad and diverse biochemical modulation. Metabolomics revealed that primary and secondary metabolism were affected by the different shading conditions, regardless of the species considered. A common pattern arose to point at the activation of plant energy metabolism and lipid biosynthesis, together with a generalized down accumulation of several secondary metabolites, particularly phenylpropanoids. Our findings indicate an intriguing scientific interest in the effects of selective shading and its application on other species and different phenological stages.

## **A google earth engine set of tools for forest disturbance mapping and area estimation**

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Forest disturbances monitoring is needed for assessing greenhouse gas balance and for properly guiding sustainable forest management in a view of climate change. Remote sensing data and the Sentinel-2 satellite missions together with Google Earth Engine (GEE) cloud computing capabilities represent a game-changer in this topic.

Here we present a GEE set of tools that allows selecting an area of interest everywhere in the world, exporting Sentinel-2 cloud-free composites and - using a specifically convinced algorithm (i.e., 3I3D) - exporting spatially explicit datasets (10-meters resolution) of forest disturbances and magnitude from 2017 to 2021. Forest disturbance maps are crucial for guiding the sample selection and increasing the precision of forest disturbance area estimates. Indeed, our GEE tool provides data that, if integrated with a photointerpreted phase, permits the calculation of statistically rigorous forest disturbance area estimates and relative confidence intervals.

The data our tool provides support greenhouse gas balance, forest sustainability assessment, and decision-makers forest managing, they help forest companies to monitor forest harvestings activity over space and time, and, supported by reference data, can be used to obtain the national estimates of forest harvestings and disturbances that many countries across the world are called upon to provide.

## Progress for the timber assortment evaluation through LiDAR. A review

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Forests provide many benefits to humans (i.e., economic, social, and environmental) and are essential for life. This is particularly evident under the current trend of climate change, for which forest practitioners are called to implement a climate-smart forestry. Accurate estimates of timber assortments are required to enhance the appropriate allocation of timber and thus supporting forest chain. Light Detection and Ranging (LiDAR) is a powerful technology to derive tree measurements (i.e., tree diameter) rapidly and accurately from forest stands, however standardised LiDAR methods for timber assortment assessments are still needed.

This study analyses scientific papers that deal with the use of LiDAR technology for timber assortment assessment in the period 2000–2021. A total of 304 papers, stored in Elsevier's Scopus® database, were collected and analysed, according 5 topics (accuracy, climate change, biodiversity, inventory, and productivity). We detected a continuous increment of the number of papers from 2000 to 2021, within which forest inventory and timber productivity were the most investigated topics. Unmanned Aerial Vehicles (UAVs) represented the most promising Airborne platforms to host LiDAR sensors, while terrestrial laser scanning (TLS) and portable laser scanning (PLS) technologies were the most used Terrestrial platforms. Spaceborne LiDAR attracted interest from researchers because it allows forest monitoring at very large scale and for different periods. TLS resulted to be the best candidate for describing the trunk and branches architecture through cylinder/circle-fitting and allometric scale methods. Machine and deep learning algorithms became crucial to handle huge LiDAR data, especially to predict, detect, classify, upscale, model LiDAR data.

Further efforts are necessary to promote the use of LiDAR data for the evaluation of ecological-related aspects such as biodiversity, tree health, and functionality to support the implementation and monitoring of sustainable forest management.

## **Effects of vegetal-derived protein hydrolysate and its fractions on response of greenhouse tomato under optimal and sub-optimal nitrogen conditions**

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Protein hydrolysates (PHs) are biostimulant products containing a mixture of amino acids and small peptides applied to the plants or the rhizosphere to enhance nutrient uptake and use efficiency, abiotic stress tolerance, and product quality, independently of their nutrient content. Their potential is best expressed under limiting environmental conditions, thus representing an opportunity to face climate changes, undesirable soil properties, and agronomic management inefficiencies. The aim of the current study was to understand the biostimulant action of a Solanaceous derived-PH (S) and its fractions [S1, S2, and S3 that are high (>10 kDa), middle (1-10 kDa), and low (<1 kDa) molecular weight] on greenhouse tomato crop under optimal and sub-optimal nitrogen level (14 and 2 mM N-NO<sub>3</sub>). Their effect was evaluated on morpho-physiological traits, yield quality, and biochemical stress markers. All parameters were influenced by nitrogen level in the nutrient solution with highest plant biomass, total and marketable yield (+142% and +116%, respectively), fruit quality under 14mM N-NO<sub>3</sub>. Nitrate reductase activity and oxidative stress (MDA contents and antioxidant enzymes) were reduced for 14 mM N-NO<sub>3</sub>. According to the fractions' molecular size, Solanaceous derived-PH affected fruit quality and plant response to nutritional stress. S1 induced an increase of total soluble solids (TSS) (+60%) and firmness (+18%), independently from nitrogen level; S2 and S3 also enhanced TSS. The highest titratable acidity (TA) was observed with 2mM N-NO<sub>3</sub> without PH application; S2 and S3 brought TA values to those obtained for the untreated control fertilized with 14 mM N-NO<sub>3</sub>. All PH treatments significantly reduced oxalic acid concentration; S, S1, S3 reduced citric acid content. Biochemical stress markers (MDA, antioxidant enzymes as CAT, APX) were reduced by PH treatments, and NR activity was increased mainly by lower molecular size fractions, confirming the positive effect of PH for enhancing stress tolerance under sub-optimal nitrogen availability. S3 gave the best results and further studies could confirm its positive effect on fruit quality.

## Development of a new protein hydrolysate from fish by-product. Potential application in the cultivation of *Ranunculus asiaticus*

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The EU regulation 2019/1009 governs the suitability of by-products for use as biostimulants. In this study, a new protein hydrolysate from fish by-product (FISH n.7) was developed through the FISH project (<https://test.progettofish.cf/>). FISH n.7 was applied in the soilless cultivation of *Ranunculus asiaticus*. Plants were weekly treated by sprayer with deionized water as control, FISH n.7 at the concentration of 0.3, 0.5 and 1 ml L<sup>-1</sup> and the commercial product FISHMIX (Biobizz) at 2 ml L<sup>-1</sup>. SPAD evaluation and biometric surveys were conducted. Both in the pre- and full-flowering phase transpiration rate, stomatal conductance and net photosynthesis rate were evaluated. The daily flower production, biometric traits and post-harvest longevity of cut flowers were also assessed. Leaves were then analyzed for the quantification of chlorophylls and carotenoids. At the end of the cultivation, the rhizomes were finally taken and their number, size and average weight were evaluated. Few significant differences in terms of vegetative development and SPAD index were obtained. As regards the physiological state, plants treated with both protein hydrolysates showed a significantly higher photosynthetic rate than the control plants. No differences emerged in the biometric traits of the flowers, while the treatment with FISH n.7 at 0.5 ml L<sup>-1</sup> induced a significant increase in the vase life. Treatment at 1 ml L<sup>-1</sup> induced a significant higher daily water absorption by the cut stems and higher content of total chlorophylls and carotenoids in the leaves. Total polyphenol content and the antioxidant activity (FRAP assay) were evaluated in fresh petals. Data showed that FISH n.7 at 0.5 ml L<sup>-1</sup> induced a significant increase in both parameters compared to plants treated with FISHMIX and, only for FRAP, also compared to the control. Based on the promising results obtained during the first trial, new evaluations are underway regarding four new protein hydrolysates.



## Towards the common bean pangenome

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Our agricultural system and hence food security is threatened by a combination of events, such as increasing population, the impacts of climate change, and the need for more sustainable development. Because of their nutritional quality, biological nitrogen fixation capacity, and broad adaptation to several agro-ecological conditions, legumes are crucial for these agriculture-related societal challenges. Among legumes, *Phaseolus vulgaris* (common bean) is the most important grain legume for direct human consumption in the world. Moreover, the well-documented history of its multiple domestications and further adaptation to different environments make it a model system to study crop evolution. To fully explore the genetic diversity of *P. vulgaris*, we present the first common bean pangenome constructed by using the reference genome and 340 low coverage WGS accessions, comprising wild and domesticated forms. PAVs (presence/absence variations) in the pangenome were categorized according to their gene presence frequencies as “core” or “variable”. The variable genes were then used for performing population genomics analyses with the aim to better understand the genetic basis and phenotypic consequences of the parallel common bean domestications and diversification. PAV-based analyses confirmed the population structure of the common bean characterized by five main genetic groups. Moreover, the discovery of novel variants, absent from the reference genome, confirmed that the pangenomic approach is a robust and comprehensive method to capture with a greater resolution the variation present in a certain species. These preliminary results provide a good starting point for evolutionary genomics studies as well as to identify novel functional variants related to agriculturally and economically important traits for use in future legume breeding programs.

## Polyhydroxyalkanoates-protein hydrolisates coupled production: from waste to valuable resources

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Plastic has notoriously been an environmental hazard for the planet and among the compounds which could substitute it there are the polyhydroxyalkanoates (PHAs), a class of compounds microbially synthesized in N-poor/C-rich cultures which can be exploited to produce biodegradable bio-plastics. Unfortunately, their production cost, mostly related to the extraction procedure from the bacterial cell in a pure form, are still high, therefore limiting their much needed commercial use. To favour the transition towards this sustainable source of bio-plastics we analysed the feasibility to integrate the production of commercial valuable products, i.e. three different plant biostimulants, together with the PHAs obtained from agricultural and animal residues in a mixed microbial cultures process. We therefore carried out three different protocols of hydrolysis on the bacterial biomass to concentrate the PHAs from the solid mass, and to collect the liquid fraction – consisting of disrupted proteins – which was then neutralized and resulted in three different protein hydrolysate (PH) plant biostimulants. The PHs were further tested in growth trials with cucumber and tomato plants (*Cucumis sativus* and *Solanum lycopersicum*) to assess any positive effect on their growth and productivity. Preliminary results show that all the PHs were effective as biostimulants, therefore suggesting that it is possible to biologically convert agricultural residues, a cost for the farmers, into products with high commercial value, such as biostimulants for agriculture and bioplastics. However, further analyses are needed in order to ascertain the best protocol for the simultaneous extractions of PHAs and PHs from the bacterial biomass, in order to obtain higher yields of the end products. Moreover, ongoing experiments, which focus on plant gene expression and ROS metabolism, will try to cast a light on the PHs mechanism(s) of action.

## A new breeding tool: the haplotype-phased genome assembly for *Ficus carica*

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The availability of genome sequences is a key prerequisite to apply modern breeding procedures to crops, and it is increasingly important to decipher the haplotypes that make up the genome, representing a pivotal resource to study allele-specific expression and regulation patterns. This is even more important for fruit trees, whose condition of heterozygosity is maintained by clonal propagation. Although fig tree (*Ficus carica* L.) has a great potential for commercial expansion thanks to its esteemed nutritional and nutraceutical characteristics, combined with its ability to adapt well to marginal soils and difficult environmental conditions, high-quality genomic resources have been released only recently. Here, we report a haplotype-phased genome assembly, achieved combining single-molecule, real-time sequencing technology with a methodology of chromosome conformation capture. Two pseudo-haplotypes of 538 sequences with mean size of 0.65 Mb and N50 of 1.99 Mb and 1.93 Mb, respectively, were obtained, representing ~98% of the estimated 356 Mb fig genome. A total of 400 out of 538 sequences were associated to the 13 fig chromosomes generating the fig genome v2.0. *Ad hoc* approaches including RNA-seq data, protein alignment and *de novo* prediction allowed us to predict 34,288 and 33,698 protein-coding genes per pseudo-haplotype, respectively, and ~82% of the total protein-coding genes were functionally annotated. The sequence of fig genome is being used to evaluate genetic variability of fig varieties on available Spanish, Tunisian and Turkish fig collections using a genotyping by sequencing approach in the frame of a PRIMA (Partnership for Research and Innovation in the Mediterranean Area) project, FIGGEN. This is the prerequisite for carrying out genome wide association studies (GWAS) for the identification of genes or molecular markers linked to several traits linked to fruit quality and to adaptation of difficult environmental conditions, favored by climate change, leading to the genetic improvement of this species.

## A new natural biostimulant for controlling green mould of citrusfruit

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Post-harvest fungal diseases are the main causes of economic losses in the citrus industry around the world. Green mold caused by the pathogen *Penicillium digitatum* is one of the most economically important disease of post-harvest citrus fruit in arid and semi-arid climates. Typically, this disease has been controlled by the application of synthetic chemical fungicides. However, the attention of public opinion and the scientific community are increasingly focused on reducing the potential safety risks to the human health and environment associated with the use of chemical fungicides in agriculture and the selection of resistant strains of pathogens. In this scenario, the use of biostimulants, alone or in combination with pesticides, can represent a valid alternative to reduce the chemicals. This study was aimed at testing the use of a natural biostimulant (NatBios) in combination with reduced dosages of the conventional synthetic fungicide Imazalil (IMZ) to manage post-harvest orange fruit rot. This combination resulted to be effective in reducing the incidence and severity of green mold as well as an inducer of fruit resistance against the post-harvest rot caused by *P. digitatum*. Additionally, NatBios used with reduced doses of fungicide induced a significant increase in the level of expression of the  $\beta$ -1,3- glucanase, peroxidase and phenylalanine ammonia-lyase encoding genes in fruit peel, suggesting that the natural biostimulant causes a long-term resistance of citrus fruit to *P. digitatum* infections. Quite interestingly, the residual concentration of IMZ in the fruit treated with the NatBios/IMZ mixture was significantly lower than the residues of IMZ in the fruit treated only with the fungicide at the same dose and far below the threshold values set by the European Union.

## Root exudates reuptake and alteration of carbon isotope fractionation by tomato plants under phosphorus deficiency

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Root exudation is one of the most important plants responses to biotic and abiotic stresses. Indeed, plant exposition to phosphorus (P) deficiency stimulates the release of exudates by roots increasing P solubility and thus P uptake. However, plants root exudation represents high energy and carbon (C) consumption. Therefore, we hypothesized that plant roots are able to uptake some of the exudates released saving energy. This study exploited  $^{13}\text{C}$ -labelled molecules to study the re-uptake of specific metabolites generally present in root exudates by hydroponically grown tomato plants suffering P deficiency. Therefore, tomato plants have been grown for 21 days in full and P deficient nutrient solution. Re-uptake of exudates has been evaluated by immersion of roots in a solution containing  $^{13}\text{C}$ -labeled glycine, glucose, fructose, citrate, and malate.  $^{13}\text{C}$  analysis was performed using an Elemental Analyzer coupled to a Continuous Flow Isotope Ratio Mass Spectrometer (CFIRMS). As compared to controls, P deficient tomato plants were able to reacquire significantly more citrate (+37%), malate (+37%), glycine (+42%) and fructose (+49%). On the contrary, plant nutritional status did not affect glucose re-acquisition. Unexpectedly, results highlighted that P deficiency led to a  $^{13}\text{C}$  enrichment in both tomato roots and shoots over time (shoots +2.66 ‰, roots +2.64 ‰, compared to control plants). This could be explained by stomata closure triggered by P deficiency resulting in a higher  $^{13}\text{CO}_2$  fixation as compared to  $^{12}\text{CO}_2$ , usually preferred by RuBisCO in standard conditions. In this study, we therefore observed for the first time that tomato plants are able to take up a wide range of metabolites generally found in root exudates, thus optimizing C trade off. This trait is particularly evident when plants grew in P deficiency.

## Iron and Nitrogen nutrition in tomato plants: a physiological and molecular study

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Nitrogen (N) and iron (Fe) deficiencies are two widespread limiting factors for plant growth in agricultural fields since they are involved in innumerable physiological processes in living organisms and their poor bioavailability in soils frequently exerts a strong constraint on plant growth and yield. Aim of the present work is to investigate the interplay between N and Fe nutritional pathways. Tomato plants (5 weeks old *Solanum lycopersicum* L. cv Marmande, grown under Fe-deficient (-Fe) or Fe-sufficient (+Fe) conditions) were supplied up to 24 hours with three different N sources (2 mM total N: nitrate, Ni; ammonium, A; or urea, U). Ionic, metabolomic and transcriptional analyses were carried out on young leaves (YL), old leaves (OL), and roots (R). As a control, some plants were maintained in N-free nutrient solution (-N+Fe) or in N- and Fe-free nutrient solution (-N-Fe). The elemental analyses pointed out significant changes among treatments. In comparison to the -N+Fe control plants, the U-treatment to +Fe plants determined an increase of Mn, Mg and P concentration in YL and an overall increase of Zn in plants. The supply of Ni to -Fe plants led to overconcentrate Mn and Zn in YL in comparison to the other -Fe treatments. Metabolomic analyses were carried out and revealed different metabolic profiles among treatments both in +Fe and -Fe conditions. Amino acids and, in particular those related to GS-GOGAT pathway, seemed to be strongly affected by N supply. Root metabolic profile was further integrated by the expression analyses of those genes involved in N and Fe acquisition and by activity of Fe<sup>III</sup>-chelate reductase. The obtained results and the ongoing analyses will help to define the interactions between the plant nutritional pathways of these two nutrients that could give clues on the use efficiency of N and Fe sources in tomato plants.

## Understanding of grapevine mechanisms mediated by volatile organic compounds against downy mildew using a metabolomics approach

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Plants can produce a wide variety of volatile organic compounds (VOCs), which can play a crucial role in the regulation of plant responses against stress. Different modes of action against phytopathogens have been attributed to VOCs, such as induction of plant resistance and direct inhibition of pathogen growth. In particular, the amount of some VOCs was higher in resistant than in susceptible grapevine genotypes upon *Plasmopara viticola* inoculation, indicating their possible involvement in resistance mechanisms against this pathogen. This work aims at identifying the metabolic response of VOC-treated grapevine leaves and the potential activation of VOC-mediated resistance mechanisms using a metabolomics approach. Functional analysis confirmed that a sesquiterpene reduced downy mildew severity on susceptible grapevine leaf disks. An untargeted metabolomics approach was applied using ultra-high pressure liquid chromatography-high resolution-quadrupole-time of flight-mass spectrometry (UHPLC-Q-TOF-MS) analysis of leaf disks at one and six days post inoculation. Principal component analysis applied on the features (specified by retention time and mass to charge ratio), discriminated samples according to VOC treatment and time point, indicating global metabolite changes after VOC treatment. Features with significant changes in abundance were identified according to the Kruskal-Wallis test ( $P \leq 0.05$ ) and a fold-change higher than two in at least one comparison. The selected features will be annotated comparing retention times and mass spectra accuracy with different databases. These results will help to improve the knowledge on plant defence mechanisms activated by VOCs, in order to identify active compounds for plant protection against pathogens.

## Mitigation of the infection induced by *F. oxysporum* f.sp. *lactucae* by Volatile Organic Compounds (VOCs) produced by a strain of *Streptomyces* sp.

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Volatile organic compounds (VOCs) produced by microorganisms are a mixture of metabolites belonging to several chemical classes and characterised by a low molecular weight. Bacterial VOCs have been shown to be involved in a wide variety of mechanisms, included the ability to promote plant growth, the control of plant pathogens and the induction of resistance in plants. For such reasons, they are currently attracting more and more interest as a sustainable replacement of common chemical pesticides in agriculture. *F. oxysporum* f.sp. *lactucae* (FOL) is a soil-borne fungus that infects all types of lettuce, therefore causing remarkable crop losses. Resistant cultivars are extensively used as a strategy against this pathogen but, in response, they can stimulate the development of new FOL races. In this study we demonstrated that VOCs produced by a strain of *Streptomyces* sp. can negatively influence the growth of FOL race 1, both *in vitro* and *in planta*. *In vitro*, an inhibition of fungal growth of 8% was observed whereas, *in planta*, the McKinney index (MKI) used for assessing the plant health decreased by 25%, when infected plants grew in the presence of bacterial VOCs. Furthermore, we demonstrated that bacterial VOCs produced by our *Streptomyces* sp. strain might also promote plant growth. Indeed, uninfected plants grown in the presence of bacterial VOCs, showed a 48% decrease in the McKinney index. To identify the VOCs produced by our *Streptomyces* strain, a headspace solid phase micro extraction (HS-SPME), coupled to gas chromatography with mass-spectrometry detection (GC-MS) was employed. Our research revealed a complex VOCs profile that included some interesting compounds with already known antagonistic properties, such as Germacene D and Phenylethyl Alcohol.



# The interaction between strigolactones and mycorrhiza: a morpho-physiological and metabolomics investigation in tomato

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Strigolactones are emerging phytohormones involved in the control of root hyphal branching processes and plant communication with several rhizosphere organisms, including arbuscular mycorrhizal fungi. During the symbiosis, arbuscular mycorrhizal fungi obtain carbohydrates from their host plant and, in return, the plants obtain water and mineral nutrients (mainly phosphorous and nitrogen) from their fungal partners. Considering the dual role of strigolactones in the rhizosphere as signals for arbuscular mycorrhizal fungi colonization and root branching, we investigated the potential implications of exogenous application of strigolactones on tomato plant both in absence and in presence of arbuscular mycorrhizal fungi interaction. In this work, morpho-physiological measurements, including root architecture and photosynthetic activity, were carried out to investigate the effect of the treatments in tomato plant. Thereafter, untargeted metabolomics of root extracts was carried out to focus on the main biosynthetic pathways modulated by exogenous application of strigolactones in combination or without mycorrhizal colonization. Finally, root exudation profile and root architecture were investigated. Overall, the combination strigolactones and arbuscular mycorrhiza reported a synergistic effect on morphological and photosynthetic measurement. Metabolomics reported a distinctive metabolic profile of root treated in combination with mycorrhiza, compared to single treatments. Interestingly, phytohormones such as abscisic acid, brassinosteroids, cytokinin and jasmonates were up modulated by the combined treatment of strigolactones and arbuscular mycorrhizal fungi, corroborating their involvement in the mutualistic interaction between plants and mycorrhiza. This finding was also confirmed by the exudation profiles, where an enrichment of flavonoids and amino acids was also highlighted.

## Development, formulation and pre- efficacy evaluation of an innovative microbial-based biostimulant

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An increasing need for sustainable agricultural systems is required to preserve soil fertility and reduce losses of soil biodiversity. An eco-friendly alternative to the use of agrochemicals fertilizers is the microbial biostimulants containing selected microorganisms (Plant Growth-Promoting Rhizobacteria, PGPR). PGPR can benefit plant fitness through hormones, siderophores, and phosphatase synthesis, as well as N<sub>2</sub>-fixing activity. Moreover, PGPR can protect plants against biotic or abiotic stresses. The present study aimed to isolate, characterize, new effective PGPR strains and develop an innovative low-cost microbial-based biostimulant. Thirteen potential PGP bacteria were isolated from the rhizosphere of wheat plants cultivated under drought stress and nitrogen deficiency. Among these, the two isolates TL8 and TL13 showed multiple plant growth promotion activities. The two isolates, identified as *Kosakonia* (*K.*) *pseudosacchari*, were able to produce IAA up to 12.69±1.79 mg L<sup>-1</sup> and 21.41±2.64 mg L<sup>-1</sup>, respectively, without L-tryptophan. The formation of haloes around *Kosakonia* colonies (25.0±1.1 mm and 30.0±0.0 mm) grown on CAS agar highlighted their ability to produce siderophores. Following the growth of TL8 and TL13 on DF salt media with ACC as sole nitrogen source, determine their ability to synthesize ACC deaminase. They resulted also tolerant to abiotic stress and were able to efficiently colonize plant roots as observed in vitro assay under fluorescence microscope. Strain TL13 was tested to develop biostimulant prototypes using low-cost alternative carbon sources such as whey, protein hydrolysate, exhausted yeasts, molasse, or vinasse. This approach allowed us to obtain suitable formulations both, solid (dusty powder and vermiculite-based) and liquid (raw castor oil/alginate-based emulsion). Moreover, the application of *K. pseudosacchari* TL13 based formulations in pot experiments improved growth performance of maize plants. Finally, the ability of *K. pseudosacchari* TL13 to use agro-industrial organic by-products as carbon sources for its metabolism makes it a promising candidate for the development of innovative biofertilizers.

## Fermented foods: a reservoir of health-promoting lactic acid bacteria

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Fermentation processes have been used for centuries for food production and preservation. Nowadays, there is a growing attention in fermented foods, which have regained popularity in Western diets, for their health-promoting potential, mainly related to the role of Lactic Acid Bacteria during the fermentation process. Among them, *Lactiplantibacillus (Lpb.) plantarum* strains, with a long history as starter cultures in the production of a wide variety of fermented foods, are being investigated for their beneficial properties which are similar to those of probiotic strains, in several *in vitro* and *in vivo* studies. Our study was aimed to investigate selected *Lpb. plantarum* strains, isolated from fermented foods, for their adaptation and adhesion ability in the GI tract and the potential to affect host health through various beneficial activities. For these purposes, food- and gut-associated *Lpb. plantarum* isolates were genetically characterized by MultiLocus Sequence Typing and phenotypically investigated for properties that could influence their probiotic potential. We also examined, using a targeted metabolomics approach (UPLC-QTOF/MSE), the ability of these strains to metabolize bile acids and salts (BAs) discovering a selective ability of food-associated strains to modulate BAs in favour of receptor activation that would be beneficial for the host, demonstrating also an evolutionary-conserved property, previously reported only in gut microbes. In addition, the specific ability of each strain to modulate ROS levels in response to either oxidative or inflammatory stress and to restore inflammation via IL17/IL23 axis in an inflamed intestinal cell model was investigated as a promising strategy to treat intestinal inflammatory diseases. Overall, this work highlights the beneficial contribution of fermented foods through their microbial components (as *Lpb. plantarum* species) that prompt us to develop multifunctional starter cultures with desirable functional and technological features for possible application in dairy and non-dairy products fermentation to produce health-promoting and quality improved functional foods.

## Legumes affect bacterial communities of intercropped wheat due to phosphorus availability

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The increase of phosphorus (P) uptake in a less P-efficient crop (wheat) when intercropped with legumes is generally ascribed to their root exudation (carboxylates, H<sup>+</sup> and phosphatases) but P yield is not always consistent to root exudation rates. Since root exudates also influence soil microorganisms, this study investigated rhizoplane bacterial communities and their role in soil P dynamics. In a pot experiment, durum wheat was intercropped (IC) with pea and lupin and grown in P-limited soil supplied with different chemical forms of P, in order to obtain four P-availability levels: KH<sub>2</sub>PO<sub>4</sub> as available-P (AP), Ca<sub>3</sub>(PO<sub>4</sub>)<sub>2</sub> as unavailable-P (UP), no added P (NoP) and the same soil under several years of ordinary amendment (NPK). The bacterial community composition and phosphomonoesterase activity (PME-activity) of the rhizoplane were studied and the results compared to that from the respective sole crops (SC) and bulk soils (BS) managed under the same conditions. Bacterial communities were primarily affected by crop, followed by P treatment. Legumes reduced bacterial diversity compared to wheat and when P became a limiting nutrient (UP, NoP), it favored certain taxa considered phosphate-solubilizing bacteria (PSB) and plant growth-promoting rhizobacteria (PGPR) in SC and IC, generating a more interconnected community and complex network in IC than in SC (at UP). The results suggest that this "selective ability" was extended to the intercropped wheat due to the intermingling of the roots. At the same time, alkaline PME-activity was significantly higher in UP and positively correlated with the relative abundance of *Pseudomonadales*. Acid PME-activity was higher when lupin was included in the intercrop and positively correlated with orders containing the phoD phosphatase gene (*Streptomycetales*) and polyphosphate bacteria (*Betaproteobacteriales*). The results suggest that the taxa selected by the legumes can affect P uptake by plants and contribute to explaining the increasing of P yield observed in legumes intercropping.

## The fundamental role of Tree-related microhabitats for saproxylic beetles conservation

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Forests are the principal sinks of terrestrial biodiversity and provide many ecosystem services. The Mediterranean forests, due to environmental heterogeneity are considered a very important biodiversity hotspot. Over the years, the abandonment of rural and mountain areas, in addition to human actions and climate change, strongly impact on forest ecosystem's functions and in particular on biodiversity conservation. To improve the forests' multifunctionality different innovative managed systems were developed. Integrated forest management is an innovative and complex managed approach that aims to reduce the trade-off between biodiversity conservation and timber production. The study, realized in 60 plots belonging a regularly managed Apennine beech forest, aims to analyze the relationships between structural, environmental, and Tree-related microhabitats (TreMs) features on beetles' community abundance and diversity. The insect sampling was accomplished by placing a window trap in the center of each plot, while TreMs were collected from each standing tree, according to the hierarchical framework of the TreMs field catalogue. The results demonstrate that Terms occurrence, altitude and slope, influence significantly the beetles' abundance and diversity. In addition, TreMs impact on both beetles' community species and the saproxylic IUCN red list.

## Pheromone-mediated mating disruption to control chestnut tortrix moths

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In the last decades, the development of effective and environmentally sustainable pest management strategies has been strongly encouraged, aiming to reduce pesticide use and to obtain residue free foods, as well as fewer negative impacts of chemicals on human health and the environment. Pheromone based devices are successfully used to control insect pests in agriculture. Mating disruption (MD), in particular, has recognized as one of the most successfully strategies for controlling various pests. It involves the use of synthetic sexual pheromones in large amounts to confuse males and limit their ability to locate females, reducing pest mating and preventing crop damage. Non target effects are reported to be rare, and neither the target organism nor other species are killed, decreasing the risk of resistance to pesticides as well. Investigations were conducted to evaluate the effectiveness of MD to control the chestnut tortrix moths, *Cydia fagiglandana* (Zeller) and *Cydia splendana* (Hübner) (Lepidoptera: Tortricidae), in the two-year period 2019–2020 in chestnut-growing areas of Northern Italy. The effectiveness was assessed by recording male adult catches in pheromone baited sticky traps and larval infestation in chestnut fruits, comparing MD and control plots. The total number of trapped males was significantly lower in MD plots than in control ones, for all sites and years. Trap catch suppression in MD plots averaged 89.5% and 93.8% for *C. fagiglandana* and 57.4% and 81% for *C. splendana* in 2019 and 2020, respectively. The larval infestation rate in fruits was not significantly reduced in MD plots except for one site where a reduction of about 71% was recorded in 2019. Specific investigations about background population density, dispersal and mating/oviposition behaviour are essential for a viable management strategy.

## Mapping melliferous potential and productive honey areas through GIS: a more sustainable approach to beekeeping

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In the last years, climatic conditions and adverse natural events such as drought and frost, severely affected the honey sector. Honey production was seriously damaged. Meantime beehives have significantly increased determining overcrowding and beekeepers competition. The identification, through mapping, of potential new honey production areas can certainly mitigate these effects, leading beekeeping to a more sustainable one.

In this work, with reference to the 2021 year, a GIS-based approach is proposed in order to map melliferous potential (MP) and expected honey production (EHP) areas. The study area is located within the Torino province (NW Italy), being the main chestnut and acacia honey production zone in the Piemonte region.

MP index was obtained from literature and associated to the local (main) crops and forest types as mapped on the regional vector land cover map. Unfavourable and restricted zones for beekeeping were masked out and an estimation of EHP given. Computations were operated on raster basis (GSD = 10 m). Assuming that a bee can fly for a maximum of 3 km during the foraging phase, a squared moving sliding window (sizing 3 x 3 km) was considered to locally compute by spatial sum the correspondent EHP map (EHPM). MP map (MPM) can be used to give a local estimate (on pixel basis) of MP. Differently, EHPM can give the local estimate of EHP (possibly associated to a hive positioned at that location) taking care about the surrounding useful vegetation as mapped by MPM. It is expected that MPM and EHPM could support beekeepers' associations for addressing management and monitoring of beekeeping sites towards a more sustainable direction. Specifically, new sites for hive hosting could be planned in unexplored areas having high EHP values minimizing unfavourable effects related to beehive overcrowding and beekeepers competition.

## Carbon dynamics in constructed soils of New York City (USA)

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The construction of artificial soils for urban greening is very common in cities across the world. These soils are classified as Technosols by the World Reference Base for Soil Resources (WRB) and they provide ecosystem services related to water purification and soil contaminant reduction, climate regulation, nutrient cycling and production of biomass for urban farming in many urban contexts. Moreover, they function as a site for carbon (C) sequestration, by stabilizing several forms of organic compounds as byproducts of microbial metabolic processes. In this study, carbon dynamics were studied in twenty-four plots made up of 1/3rd compost and 2/3rd fine sand glacial sediments established in Corona, Queens (NY, USA) over two years (four samplings total). Plant cover treatments included: bare soil, sunflower, cover crops, edible, sunflower and cover crops, sunflower and edible, sunflower and cover crops and edible. The investigated parameters were total C, water soluble C, particulate and mineral absorbed organic C. Measurements of enzymes involved in the carbon cycle were also made, including dehydrogenase,  $\alpha$ -glucosidase, protease and catalase. The plots showed an active and dynamic carbon cycle with marked changes in variables over time and significant differences between treatments. Changes in total C and stable fractions, mainly mineral absorbed organic C, and increases in enzymatic activity, mainly  $\alpha$ -glucosidase and protease, suggest that Technosols have a high potential for C sequestration and nutrient cycling to support plant growth.



## Spatiotemporal analysis of marginalization drivers in Gennargentu-Mandrolisai inland areas

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The Gennargentu-Mandrolisai district, located in central Sardinia, is part of the national strategy for inland regions intended to foster the economic growth of marginal areas. After the Second World War, the massive abandonment of cereals and orchards, combined to overgrazing or abandonment, has caused the degradation of grasslands or their ecological succession to woodlands. The present research aims to understand the marginalization process integrating a direct approach with the main stakeholders (farmers, public bodies) combined with quantitative spatial analysis. Qualitative data from local actors were collected through questionnaires about the main drivers that led to the abandonment of rural areas. A diachronic analysis of land use from Corine Land Cover maps on four different dates over thirty years (1990, 2003, 2008 and 2018) was used to understand the main changes. In total, thirty farmers from six municipalities were interviewed. Their farms cover 1602 ha in total between two municipalities in the mountain area (670 ha in Desulo and Tonara) and four municipalities on the hill area (932 ha in Atzara, Samugheo, Sorgono, Ortueri). The spatial analysis allowed to highlight relevant differences among municipalities caused by different geo-morphological characteristics, social attitudes, and historical decisions of the former and current local government. The interviews revealed some recurrent marginalization factors such as: unprofitability (50% of answers), property atomization (37%), the distance of the fields (27%), stringent legislation (10%). Some 93% of the interviewed have undertaken operation of recovery, regarding viticulture, pasture, forage production and orchards. In particular, the analysis of the fragmentation pointed out that the average farm has 10 fields, ranging from 1 to 63. The analysis of these factors of marginalization will lead us to hypothesize new models and innovative systems to promote agricultural activities in the territory.

## Carbohydrate reserves limit *Quercus ilex* ability to recover after severe water stress more than embolism formation: multiple mechanisms underlying the maintenance of xylem hydraulic functionality

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Holm oak (*Quercus ilex* L.) dominates the landscape of several Mediterranean coastal forests. Although, it is considered a well-adapted species to water stress, worrisome declining symptoms, such as crown defoliation and leaf desiccation, leading to holm oak dieback have been recently observed in Southern Tuscany. We investigated the physiological and molecular mechanisms leading to embolism formation and depletion of carbon reserves in holm oak seedlings exposed to severe water stress and rewatering. We measured gas exchange, stem water relations and hydraulic conductivity. The content of non-structural carbohydrates (NSC), the expression of drought-induced genes in wood parenchyma and wood anatomical changes were also investigated. Under water stress, a stem midday water potential of - 4.6 MPa corresponded to a 50% loss of hydraulic conductivity. The low plant hydraulic conductance was accompanied with a prolonged stomatal closure with a consequent depletion of carbon reserves. NSC consumption was the consequence of the upregulation of  $\alpha$ -amylase and the downregulation of glucose and sucrose transport genes, suggesting glucose utilization to sustain cellular metabolism. Wood anatomical adjustments showed an increase in cell wall thickness along with changes in lignin composition, helping the plants to regulate water storage inside the tissues and to maintain the water transport in xylem vessels. In conclusion, our results showed that, under severe water stress, *Q. ilex* activates several physiological and molecular mechanisms to preserve xylem functionality at the expense of wood carbon reserves. Therefore, recurrent drought events may expose this species to the risk of carbon starvation, compromising its resilience and survival in the Mediterranean coastal area.

## Effect of different regulated deficit irrigation strategies on berry secondary metabolites in *Vitis vinifera* L. cv Sangiovese

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Irrigation management in the vineyard represents a key factor for achieving high-quality production, especially in the face of the new challenges imposed by climate change. An experiment was carried out on 6-year-old potted vines (*Vitis vinifera* L. cv Sangiovese) subjected to six different irrigation regimes: full irrigation during the entire irrigation period (FI); severe water deficit applied during the lag-phase (RDI-L); moderate (RDI-1M) or severe (RDI-1S) water deficit applied between fruit set and veraison and moderate (RDI-2M) or severe (RDI-2S) water deficit applied from veraison to harvest. The vines were fully irrigated when not subjected to deficit irrigation. The vine water status was monitored weekly by stem water potential (SWP) and leaf gas exchange measurements. The berry soluble solids content, titratable acidity, pH, anthocyanins and flavonols content and composition were monitored from veraison to harvest. The berry aroma profile was analyzed at harvest, identifying over 90 glycosylated volatile organic compounds (VOCs). Moderate (RDI-M) and severe (RDI-1S) water stress imposed before veraison strongly improved the accumulation of anthocyanins (+ 55%) and flavonols (+ 85%) compared to FI vines. The preveraison water deficit, particularly when severe, also increased the berry total VOCs at harvest. The highest concentrations of total VOCs were observed in berries of RDI-1S vines (12.5  $\mu\text{g g}^{-1}$  berry dry weight), while FI berries showed the lowest ones (6.3  $\mu\text{g g}^{-1}$  berry dry weight). Even the shortterm RDI imposed during the lag phase significantly increased the total VOCs compared to FI treatment (+ 51%), while both post-veraison RDI had minimal effect on berry aroma. Similar effects were also observed on many classes of detected VOCs, particularly monoterpenes, C13- norisoprenoids, vanillins, benzene derivatives, aliphatic alcohols and phenols. Our results show that timing and volumes of irrigation differently affect berry secondary metabolites, allowing to manage fruit and wine quality.

## Evaluation of different ornamental perennial species combinations for weeds' containment in the urban environment

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Sustainable cities planning, that is one of the 17 Sustainable Development Goals defined by Agenda 2030, have to consider the design of urban green areas. To them an important role in ecosystem services provision and human wellbeing is recognized. Ornamental perennials can play a strategic role in keeping the soil always covered, sparing water and avoiding weeds, for the reduction of maintenance costs. The aim of this research was to evaluate the growing performances of different ornamental perennial herbaceous species combinations against the onset of weeds. A randomized block design experiment was performed for three growing seasons, starting from April 2019 to November 2021 in a nursery (Vivaio Purpurea) located in the municipality of Piobesi (Turin, Piedmont, Northwest Italy). Six species were selected: *Hemerocallis* 'Stella de oro' (A), *Sedum spurium* 'John Creech' (B), *Tulbaghia violacea* (C), *Phlox subulata* 'Trot Pink'® (D), *Potentilla newmanniana* (E) and *Gaillardia aristata* 'Kobold' (F). In 32 plots, 4 m<sup>2</sup> each, four replicates for each combination were evaluated: AB, CD, EF, AB+CD, AB+EF, CD+EF; AB+CD+EF; T (no plants as control). Each year, three manual weeding were performed and dry weights of the aerial part of the weeds was calculated. Deep observations, biomass production, and image analysis for each plot were done. CANOPEO software was used to determine the coverage aptitude of the tested combination during the entire experiment. Data were statistically analyzed. As results, all plots with perennial plants showed a better effect in weeds' containment than T plots. *Gaillardia aristata* 'Kobold' resulted in a quick covering effect in the first year, with a strong regression in the second; CD combination showed the best role in weeds' containment. The research allowed to identify the best solutions to keep the soil always covered and improve the ornamental and environmental value of urban green spaces. More in-depth studies on the citizens perception of perennial herbaceous species in urban green areas will be conducted in a new trial starting in 2022.

## **The role of sustainable agroforestry practices and food consumption to prevent tropical deforestation and increase ecosystem services**

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Tropical forests harbor most of global terrestrial biodiversity and provide essential ecosystem services, although they have been largely replaced by agricultural activities. Amazon is the largest tropical forest remaining, and cattle ranching and soybean cultivation are by far the greater drivers of deforestation, that together with other few commodities constitutes the main products responsible for the imported deforestation caused by European countries. Agroforestry systems and sustainable intensification are pointed out as solutions to conciliate food production with environmental conservation, thus reviewing the results of studies comparing these practices against conventional ones are needed. We performed a systematic review of papers assessing the effects of cattle ranching and soybean cultivation on Amazon deforestation as well as performed a meta-analysis on the effects of sustainable practices on different ecosystem services. Our results highlight the negative impacts of cattle ranching and soybean crops on the ecosystem dynamics and functionality of the Amazon biome, also due to speculative economical mechanisms. Our meta-analysis indicated a huge gap of knowledge of empirical studies comparing sustainable and conventional practices, with just 13 studies meeting the requirements for inclusion. The overall model comparing the ecosystem services provided in conventional land-uses and those adopting sustainable practices indicated a non-significant effect. Such result was due to the negative effect found on the few studies assessing crop yield and herbage biomass, a pattern also found in a European meta-analysis. However, livestock productivity, soil organic carbon, soil fertility and woody biomass were positively affected, indicating a wider beneficial effect. Our review also shed light on the responsibilities in the consumption of many food products that are the cause of tropical deforestation. In conclusion, we discuss challenges and opportunities to advance research into the real effects of sustainable practices to conciliate agriculture and tropical forests conservation.

## Does tree position along a hillslope affect sap flow rates? A case study in a beech forest stand in central Italy

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Forest ecosystems play an important role in water regulation through evapotranspiration (ET) fluxes. Variation in forest ET is a complex process due to the influence of numerous variables, such as climatic forcing (e.g., air temperature, air humidity, solar radiation and wind speed), forest structure (e.g., LAI), and soil water availability. The present study aims to understand and quantify sap flow spatio-temporal variability in response to air temperature and soil moisture along a steep hillslope. Sap flow rates were measured in a European beech stand (*Fagus sylvatica* L.) during the growing season (April-September) in 2021 at three different positions along a hillslope (bottom, mid and upper hillslope) in the Re della Pietra catchment, Tuscany, central Italy. The monitoring was performed by using 12 TreeTalker devices installed on four trees per hillslope position. The devices measure environmental and tree variables simultaneously with hourly resolution. Soil moisture was measured at three locations at 15 and 35 cm depth through 6 frequency-domain reflectometry probes. The results showed statistically significant differences in the sap flow rates in the three different positions in May, June and July. Soil moisture content, measured at 15 and 35 cm depths, showed variability along the hillslope in the same period, while there were not significant differences in air temperature among the three locations. Sap flux density was maximum in the upslope position in June, when the soil moisture content at the two different depths showed intermediate values between May (higher values) and July (lower values). This study highlighted that sap flow rate variability is affected by hillslope topography and explained by differences in soil moisture content.

## Investing in sustainable intensification for smallholders: quantifying large-scale costs and benefits in Uganda

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In Uganda, upgrading smallholder agriculture is a necessary step to achieve the inter-linked sustainable development goals of hunger eradication, poverty reduction and land degradation neutrality. However, targeting the right restoration practices and estimate their cost-benefit at the national scale is difficult given the highly contextual nature of restoration practices and the diversity of small-scale interventions to be adopted. By analysing the context-specific outcomes of 82 successful case studies on different Sustainable Land and Water Management (SLWM) in Uganda, we estimated that out-scaling of existing successful practices to 75% of agricultural land would require a one-time investment of US\$ 4.4 billion from smallholders. Our results show that, besides the many social and environmental benefit commonly associated to SLWM, a wide outscale of SLWM could generate US\$ 4.7 billion every year, once the practices are fully operational. Our context-specific estimates highlight the profitability of investing in smallholder farming to achieve the SDGs in Uganda, with geographical differences coming from specific social-ecological conditions. This study can guide sustainable intensification development by targeting the most suitable SLWM practices and plan for adequate financial support from government, investors and international development aids to smallholder farming.

## Role of the bacterial community in the outbreak dynamics of the European spruce bark beetle *Ips typographus* in Dolomites

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Heavy storm and snow falls have severely affected the Dolomite region's conifer forests in recent years. Consequently, Norway spruce-dominated forests got more susceptible to infestations by the European spruce bark beetle *Ips typographus*. Like most insects, *I. typographus* is associated with microorganisms that play an essential role in beetle's biology, like nutrition, development, and fitness. However, their role in outbreak dynamics of this important pest species is currently not known. Here, we present our project on the community structure and diversity of microorganisms of *I. typographus* using a high-throughput sequencing approach. Beetles from three regions of the Dolomites – Eastern Tyrol, South Tyrol and Belluno were sampled and compared to populations from the Apennines, Russia, Romania, and Croatia, for potential Pleistocene refugia of *I. typographus*. Sequencing was performed using an Illumina MiSeq platform for the amplification of a partial fragment of the 16S rRNA gene. Pairwise comparison of the alpha-diversity showed a significant difference among locations. Similarly, the bacterial community composition varied among the sampled regions. In general genus *Erwinia*, *Spiroplasma*, *Wolbachia*, *Tyzzarella* and *Pseudomonas* were abundant in all the studied localities. More detailed information about the role of each genus will be discussed. Our results increase our understanding of symbiotic interactions in *I. typographus* and can be utilized to develop new pest management strategies.



# Long-term influence of a municipal solid waste compost on the mobility of potentially toxic elements (PTEs) and bio-chemical properties of a contaminated soil: a field study

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In-situ immobilization is a technique that involves the use of amendments to reduce the mobility of potential toxic elements (PTEs) and improve the functionality of contaminated soils. To this end, this study evaluated the long-term effect (4 years) of a municipal solid waste compost (MSWC), added at 1.5-3.0-4.5% w/w on the mobility of PTEs present in an historical mining site (i.e., Sb 416 mg kg<sup>-1</sup>, Cd 47.6 mg kg<sup>-1</sup>, Pb 2653 mg kg<sup>-1</sup> and Zn 7666 mg kg<sup>-1</sup>). The MSWC effect on the chemical and biochemical properties of the soil was evaluated at the field scale. The MSWC addition increased soil pH (from 6.36 to 7.23), OM (from 2.1 to 3.66%), dissolved organic carbon content (from 0.01 to 0.05 mg DOC g<sup>-1</sup>) as well as cation-exchange capacity (from 4.64 to 13.9 cmol<sub>(+)</sub> kg<sup>-1</sup>). The addition of MSWC at the highest concentration (i.e., 4.5%) reduced the fraction of Cd (27.1%) and Zn (27.8%) extracted with Ca(NO<sub>3</sub>)<sub>2</sub> and that of Sb (125%) extracted with (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, while increased the residual ones (e.g., 7.6% for Sb and 4.0% for Pb). This can be explained by adsorption and precipitation processes triggered by the addition of compost. MSWC at the highest concentration significantly increased microbial respiration (up to 5.1-fold higher than control) and soil enzymatic activities. For instance, dehydrogenase, -glucosidase, and urease activities increased in 4.5% amended soils up to ~10.4-, 7.2-, and 3.4-fold compared to control soil respectively. Taken together, the results obtained in this field experiment highlight the ability of the MSWC to immobilise Sb and co-occurring PTEs, and to improve the health of a contaminated soil. Importantly, such positive effects are visible after 4 years since MSWC addition.

## The dark side of the light: influences of modern nocturnal illumination on urban trees

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The light/dark cycle of the sun is the principal environmental factor that has influenced the biological rhythms of life since its genesis. However, from the middle of the nineteenth century, the use of nighttime artificial lighting (e.g., streetlamps) has undoubtedly interfered with the natural light/dark cycle, thereby altering the physiological behaviors of living organisms, including trees, the so-called light pollution. Light pollution is one of the most impacting factors that interfere with plant photodynamics in the urban environment. An experiment was conducted in *Tilia plathyphyllos* (T) and *Platanus x acerifolia* (P) to test whether different 4000 K light-emitting diode (LED) streetlamp intensities (~300 and ~700  $\mu\text{mol m}^{-2} \text{s}^{-1}$  PAR during the night, respectively) alter tree physiology. Gas exchange parameters, (determined at dawn, midday, dusk and night) and leaf chlorophyll content were measured monthly from leaf expansion to senescence. In both species, the night-illumination induced leaf  $\text{CO}_2$  assimilation rate ( $P_n$ ) during the night, irrespectively of the light intensity, indicating photosynthetic activity compared to relative controls (Cnt). However, during the night, the activation of  $P_n$  processes in night-illuminated species led to lower  $P_n$  values at dawn than their relative Cnt plants. Also, leaf chlorophyll content was higher in night-illuminated than Cnt individuals. A delay of leaf senescence was observed only in nightilluminated P plants, in which the leaf fall stage occurred two months later with respect to Cnt. We suggest that the streetlamp illumination, characterized by a similar red:blue ratio to midday sunlight, altered plant photoperiods, "cheating" trees that day, was longer than it is. We offer clear evidence that streetlamp lighting should be carefully considered to reduce its impact on urban trees, and new less-impacting solutions on tree performances should be found in future.

## **Social Life Cycle Assessment of pig rearing facilities equipped with smart solutions for pollution control**

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In 2020, in Europe the value of the animal-based products achieved 172 bn €, the 40% of the total agricultural output. Consistently, European pig production has a relevant socio-economic weight. In the EU-27, in the last years averagely have been reared 150 million pigs, representing nearly half of total EU meat production and the 35 % of total meat output.

In parallel with this growth, significant changes are required to the sector to meet new concerns and requirements. Consumers are increasingly calling for ethical production processes for food products. The Social Life Cycle Assessment (S-LCA) defined by the United Nations Environmental Program guidelines is one of the most widespread methodologies in evaluating social sustainability of production. S-LCA evaluates how production processes impact or relate to social issues, called subcategories in the methodology, such as Child Labor, Health and Safety, Cultural Heritage, Poverty Alleviation. Subcategories can be allocated to six stakeholders' categories: Worker, Local Community, Value Chain Actors, Consumer, Society, Children and Animals.

This study proposes a methodological framework for a life cycle evaluation, through S-LCA methodology application, of pig livestock system paying particular attention to the adoption of solutions for emissions reduction. For the analysis, workers, farmers, local communities, consumers, society and animals were considered as stakeholders. A set of indicators was selected through a literature review and farmers surveys. For each of these indicators, 4 levels reference scales were defined for data evaluation.

This study is carried out as part of the APPROACh project aiming at abating piggeries indoor emission by adopting air filtering systems.

This is one of the first studies that aims to define a shared methodology to apply S-LCA to livestock farms. Considering the future requirements of livestock activities, S-LCA can play a key role in improving the social performances and sustainability of the sector.

## Cytofluorimetric analyses revealed a pathogen-specific immune modulation in response to intramammary infection in Holstein cows

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Intramammary infection is still one of the main challenges for dairy industry, due to its significant effect on both milk production and quality, and animals' welfare. For a more accurate definition of the udder health status, the somatic cell count (SCC) has been recently associated with differential somatic cell count (DSCC), which represents the combined proportion of polymorphonuclear neutrophils and lymphocytes (PMNLYM). However, obtaining more information about the role of individual leukocyte populations within the mammary gland could lead to a deeper understanding of the dynamics of udder health in dairy cattle. Therefore, the aim of this study was to investigate the association between subclinical intramammary infection from *Streptococcus agalactiae* and *Prototheca* spp. and individual leukocyte populations assessed by cytofluorimetric analysis in Holstein cows. After an initial bacteriological screening on 188 animals, cytofluorimetric analyses were performed on milk samples of 47 animals (parity>2 and DIM>120). The statistical analysis was performed with a linear model including the fixed effect of the bacteriological status (negative, positive for *Strep. agalactiae*, and positive for *Prototheca* spp.) as fixed effect. Results showed that *Prototheca* spp. infection increased the proportion of total T lymphocytes (TL) and T-helper lymphocytes (THL) ( $P<0.05$ ,  $P<0.01$ ), whereas *Strep. agalactiae* infection increased the proportion of PMN ( $P<0.01$ ). Regarding MAC, results showed different immune response patterns: *Strep. agalactiae* increased MAC content ( $P<0.001$ ), whereas *Prototheca* spp. decreased it ( $P<0.001$ ). Findings suggest that these two microorganisms promoted different host immune responses: adaptive immune response and chronic inflammation for *Prototheca* spp., innate immune response for *Strep. agalactiae*. Differentiating the individual leukocyte populations allowed to better understand host immune response following the intramammary infection and to reveal pathogen-specific patterns, which could help in the development of more specific diagnostic and therapeutic tools.

## **Effect of organic bedding materials on poultry's growth performances and welfare**

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The trial aimed to compare organic poplar (PP) and vineyard pellet (VP) as alternative to conventional wood-shavings (WS) bedding on poultry's growth performances and welfare. A total of 252 1d-old Ross 308 broilers, were randomly allocated to the 3 bedding groups (BG) with 3 replicates (28 birds, according to organic livestock regulation). Feed and water consumption (FC, WC), bedding temperature (T), humidity (H) and pH were recorded daily. Health status, body weight (BW), hock (HS), pad (FP) and cleanliness scores (CS) were evaluated 42d and 84d of experiment. The broilers were slaughtered both at 42d (n=108), according to conventional farming, and at 84d (n=97) in-line with organic guidelines. At slaughterhouse, coelomic content and single organs were weighted, while plasma was individually sampled for metabolic profile and antioxidant capacity determination. The data were statistically analysed using IBM SPSS software v28. The FC, BW, mortality, feed to gain and feed efficiency were unaffected by BG. At 42d no differences were observed for FS and PC, whereas the worst FS was observed on WS. At 84d, WS animals were the cleanest and showed the best FS and HS. No differences were observed at 42d on litter T and pH, while WS was the moistest. Bedding T was unaffected by BG at 84d, whereas WS was the driest and had the highest pH. Antioxidant capacity was similar among BG at 42d while VP had the highest value at 84d. The WS animals showed the highest metabolites in plasma at 42d and the highest coelomic content, gizzard and hot weight. On the contrary, WS at 84d had increased markers of protein catabolism and muscle damage, associated with a lowest organ development and carcass weight. Results suggest that PP and VP are less advantageous as bedding materials until 42d but are more effective in longest periods.

## **Influence of biscuit flour administration on buffalo milk quality**

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Nowadays consumers are more interested in animal derived products with nutraceutical properties, more sustainable, with less environmental impact and respectful of animal welfare. The adoption of feed plans useful for maintaining high levels of functional molecules in buffalo milk represents a strategy to be pursued and enhanced. It has already been seen that the administration of green forage (GF) improves the amount of functional molecules in buffalo milk, however it is impossible to administer GF to the animals all the year. In this regard, it was necessary to evaluate whether the introduction of biscuit flour can substitute the use of GF. The experimental trial was carried out in a buffalo farm and lasted 90 days. Fifty Italian Mediterranean buffaloes were involved, kept in free housing and milked twice daily. The animals were homogeneously divided into two groups: Group F (diet with GF) and Group B (standard 'dry' diet with biscuit flour). Bulk milk samples were taken monthly during the morning milking session. The qualitative components of milk (fat, protein, casein, lactose), the content of functional molecules (betaines and carnitines) and the total antioxidant capacity of milk were determined. An analysis of variance for repeated measures was done. No statistically significant differences emerged regarding the quantitative and qualitative composition of the milk. Group F showed higher levels ( $P < 0.01$ ) of  $\gamma$ -butyrobetaine, Glycine betaine, Carnitine and Propionyl-L-Carnitine, while no differences emerged for  $\gamma$ -valerobetaine. The antioxidant capacity of milk was higher ( $P < 0.05$ ) in group F animals than in group B. The administration of diets characterized by high levels of simple sugars, obtained through the use of biscuit flour, favored the synthesis of the aforementioned molecules, reducing the differences with the administration of GF. In conclusion, the use of biscuit flour can be a valid alternative to GF in winter periods.

## Polyculture of catfish, rainbow trout and Swiss chard in halaponics

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The study evaluated the effects of three water salinity levels (0.5‰, 3.0‰, and 6.0‰) on adaptability, growth performance, and carcass traits of black bullhead catfish (*Ameiurus melas*) and rainbow trout (*Oncorhynchus mykiss*) in polyculture, as well as on the production of Swiss chard (*Beta vulgaris*, ssp. *vulgaris*) in a low-tech and brackish-water aquaponic (haloponic) system composed by 9 independent units. A total of 261 catfish (initial weight 147 ± 22 g) were randomly distributed among the three experimental treatments (three units per treatment; initial stocking density 8.50 kg m<sup>-3</sup>) and reared for 268 days from September to June. In December, 150 trout (initial weight 153 ± 22 g) were added to the system (initial stocking density 5.06 kg m<sup>-3</sup>) and reared for 103 days in polyculture with catfish from December to March. During the trial, two cultivation cycles of Swiss chard of “Nostrana” (1<sup>st</sup> cycle) and “Pugliese” (2<sup>nd</sup> cycle) varieties were carried out (12 plants m<sup>-2</sup>, 24 plants per unit). Water salinity did not affect trout final weight (348 g, on average), survival (89.7%), as well as eviscerated carcass (87.5%) and fillet (52.0%) yields. Similarly, no differences were found in catfish final weight (193 g), survival (75.8%), and fillet yield (48.3%), whereas a higher carcass yield was found at 0.5‰ salinity than at 3.0‰ (87.3% vs. 85.2%; P<0.05) with intermediate values for 6.0‰ salinity. The marketable biomass of Swiss chard was 337 g for “Nostrana” and 823 g for “Pugliese” varieties, without differences due to water salinity.

In conclusion, polyculture of catfish, trout and Swiss chard is achievable until 6‰ water salinity, therefore increasing the diversification of cultivable species and the resilience of haloponic systems.

## Investment, subsidies, and environmental performance: conflicting effects on farms using dose-response function

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This research investigates the effect of private investment and environmental subsidies on Italian farms' environmental performance in the five main Italian regions from 2004 to 2013. The data used in this study was extracted from the Farm Accountancy Data Network (FADN), the EU main source of farm accounting data. The first stage of the empirical analysis estimates two environmental performance indicators (fertilisers per hectare of utilised agricultural area and plant protection products (PPP) per hectare of utilised agricultural area), which reflect farms' environmental pressure. After the two indicators estimation, we apply the Dose-response function (DRF) model, described in Cerulli (2015), to measure the impact of investment and subsidies on the holdings' environmental performance. We calibrated this model using investments and subsidies as treatment, in the same year of treatment and with lagged values. This paper's novelty is that we introduce the DRF (a model commonly used in diverse socioeconomic and epidemiological contexts, mainly for policy analysis and evaluation) to measure the relationship between investment or subsidy (treatment) and fertilisers/hectare or PPP/hectare (as treatment effect). The results reveal a conflicting effect of investment and subsidies on farms' environmental performance. From an economic point of view, our results show that higher levels of investment – and subsidies increase the use of fertilisers and plant protection products in the short run, reflecting healthy farms operating activity but low environmental performance. Nevertheless, our results show a positive effect of investment and subsidy on farms' ecological performance in the medium term at the lower-mid doses of treatment. These findings suggest the importance of fostering and enhancing private investment to improve farms economic and ecological performance.



## Data fusion of soil and vegetation maps for site-specific nitrogen recommendations in cereal crops

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In the context of nitrogen (N) fertilization, the implementation of precision agriculture's approaches allows a better use efficiency of the element and reduces economic and environmental losses. To achieve this goal is crucial to study the intra-field spatial and temporal variability. Then, data integration within a Decision Support System (DSS) is needed to prepare a prescription map containing the recommended site-specific fertilizer doses. Current DSSs available to farmers are based on empirical models with low integration of the different data sources.

For this reason, this study aimed to develop a DSS able to integrate soil and crop variability maps to define the site-specific N dose during the 2021 maize season of the ConSensi project (funded by rural development 2014-2020 for Operational Groups). The proposed DSS modifies the most widespread empirical DSS (GreenSeeker) used to calculate the optimal N doses proposed by Oklahoma State University. It relies on the hypothesis that a vegetation index can predict both in-season crop response to nitrogen fertilization and yield potential. Soil variability contribution is not considered. Therefore, the modification proposed identifies different zones of the field by integrating soil variability maps of electrical conductivity, considered stable over time, and vegetation index maps, which take into account seasonal variability. Then values of the vegetation index are used to define site-specific N doses by estimating crop yields (with a calibration curve,  $R^2= 0.75$ , specific for maize in Lombardy, Italy) and maize response to nitrogen. The DSS was tested on two maize fields of 2.5 ha in Lombardy. Potential economic benefits were estimated compared to uniform fertilization management: the average rate applied was 190 kg N/ha compared to 250 kg N/ha applied by farmer's common practice (an average of 24% less of applied N) also leading to possible environmental benefits to be tested in future field experiments.

## Impact of defoliation treatments on the nutrient balance of club variety apple trees

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Club apple varieties are high value sellers on the Central European market behind top vendors like Golden Delicious and Gala. Despite their popularity with consumers and producers, several bicolored Club varieties, for instance Cripps Pink / Pink Lady® and Nicoter / Kanzi®, require elevated sun exposure of the fruits during the last weeks before harvest to generate the desired coloration for a successful commercialization. Defoliation of the plants during the vegetation period is an efficient method to enhance sun exposure of fruits.

This work investigates the effect of three different agronomical approaches (summer pruning, pneumatic defoliation, summer pruning in combination with pneumatic defoliation) on the nutrient balance of the plant and discusses the impact of the treatments on fruit coloration compared with untreated plants. Particular attention is drawn to the influence of defoliation treatments on the carbohydrate cycle of the plant, thereby comparing available carbohydrate reserves in shoots and roots of treated and untreated trees. By confronting various sugar derivatives and their respective concentrations in plant organs during different growth stages, the impact of defoliation treatments on the carbohydrate reserves of the plant is elucidated. This knowledge allows conclusions on the availability of the nutrients during and after dormancy of the plant and permits to predict the influence of defoliation treatments on fruit growth. Additionally, the coloration of apple fruits on defoliated and untreated trees is assessed in terms of anthocyanin content in the fruit skins. Besides the determined colour indices, the quantity of anthocyanins in fruits is a measure for the elevated amount of sunlight in treated plants since these components accumulate with sun exposure. Therefore, sugar and anthocyanin content provide useful information on the efficiency of the three investigated treatments of the Club apple varieties Cripps Pink / Pink Lady® and Nicoter / Kanzi®.

## Low-phytate grains to enhance phosphorus sustainability in agriculture: advances on the *lpa1-1* maize mutant

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In the last decades, phosphorus has become one of the main sustainability issues as a non-renewable resource. In plant seeds, the main reserve form of phosphorus is phytic acid (PA), a strong anti-nutritional factor. PA is poorly digested by monogastrics and is expelled with manure, becoming a pollutant, and contributing to eutrophication. Moreover, the consumption of PA-rich diet leads to mineral deficiencies (mainly iron and zinc) in human population. Therefore, the reduction of PA in cereal seeds has become an important challenge in breeding programs: many *low phytic acid (lpa)* mutants have been isolated in major crops and *lpa1-1* is the most promising in maize. Unfortunately, PA decrease is followed by negative pleiotropic effects on the seed and on plant performance. Aim of the present work is to study the two main pleiotropic effects in *lpa1-1*, i.e. the seed weight reduction and the susceptibility to drought stress. First, we report the results of a two-year field experiment conducted in two different genetic backgrounds: in high-input conditions, *lpa1-1* exhibits a comparable seed weight/ear than the wild-type, limiting the problem of the mutant to the reduced field emergence. Additionally, we have investigated drought stress using three approaches, including hydroponics, mesocosms and field. Epigeal and hypogeal measurements show that drought stress in mutant plants seems to be caused by a reduced photosynthetic efficiency and not by a shallower root system: in controlled conditions, *lpa1-1* exhibits a reduced efficiency of photosystem II (Fv/Fm, 0.810 vs 0.800) and a reduced leaf temperature (-0.5°C) compared to wild-type, probably due to increased water loss; in the field, a drop in the net photosynthetic CO<sub>2</sub> rate (-50%) and in stomatal conductance (-62%) was observed. Overcoming these pleiotropic effects would determine several potential benefits for the nutritional quality of food/feed and for the environmental P sustainability in agriculture.

## Can the interaction between pollen and toxic compounds affect honey bee survival?

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Honey bees (*Apis mellifera* L.) play a vital role in ecosystems and are essential for both plant biodiversity and agricultural production; indeed, one third of world crop production relies on animal pollination. Pollen is the only source of proteins for honey bees and contains substances that are necessary for growth and development. However, pollen may also contain toxic compounds resulting from plants metabolism or the contamination with pesticides used in agriculture. In presence of potentially harmful substances, the metabolic detoxification (based on Cytochrome P450 monooxygenases) is activated. Therefore, both pollen and toxic compounds could engage the same physiological response system resulting in a potential interaction between the two factors.

To gain insight into the possible interactions between pollen and xenobiotics potentially associated to it, we carried out dedicated lab experiments using nicotine as toxic compound, both because it is common in some nectars and pollens and for its affinity with some insecticides. To this purpose honey bees were fed with nicotine, pollen or both and treated or not with the insecticide synergist piperonyl butoxide (PBO), a P450 inhibitor, normally used to enhance the toxicity of pyrethroid and neonicotinoid insecticides. Replicated experiments were carried out both early in the season, when the prevalence of a common viral pathogen (DWV) is low, and later in the season when viral infection is widespread.

Preliminary results suggest that nicotine, at the tested dose, negatively affects bee survival only early in the season. Under these conditions, pollen appeared to counteract the negative effect exerted by nicotine. Instead, late in the season, when viral infection is higher, nicotine alone doesn't seem to be similarly harmful, whereas the concurrent presence of pollen and nicotine significantly reduces honey bee survival. Finally, PBO shows a different trend according to the season.

## An overview of the Marie Curie Project: “CONSUMEHealth. Using consumer science to improve healthy eating habits”

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This presentation aims to present the overview of a Marie Skłodowska-Curie Action (MSCA) project titled “CONSUMEHealth” Using consumer science to improve healthy eating habits. This MSCA (2017-2020) has focused with healthy eating habits, a key issue for the EU in contributing to safe, healthy and nutritious food for preserving life and making positive impact on health and society.

In fact, although today’s consumers can make informed decisions about which foods, and in what quantities, are best for a healthy life style, in recent years in the European Union there has been an increase of diet-related health problems caused by unhealthy and over-consumption of food (e.g. overweight, obesity, and other chronic diet-related diseases).

The objective of this MSCA was twofold: (1) understand what drives consumers to make healthier food choices and (2) provide evidence-based recommendations for stakeholders and policy makers to develop and communicate innovative win-win solutions to improve eating habits. Three main projects were carried out:

- Health messages & pasta choices study Aim: to test the impact of labelling whole grain pasta with a health message descriptor displayed at the Point-Of-Purchase. Methods: controlled-field experiment in a college dining venue, over a nine-week period.
- Healthy eating & students habits study Aim: to explore barriers and enablers that US and Italian college students perceived as influencing healthy eating behaviors. Methods: In person-interviews; Focus Groups and Nominal Group Techniques
- Blended burgers study Aim: to explore the factors influencing acceptance and intention to consume a mushroom-beef burger. Methods: survey among US college students, with a follow-up survey to evaluate behavior after one month.

This presentation will show the acquired expertise and results of the projects as well as explain the career benefits of carrying out a MSCA project in Agricultural Economics.

## The implementation of nudges to foster healthy and sustainable eating at the university canteen

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Today's dietary choices impact consumers' health and the planet. Therefore, exploring strategies to foster healthy and sustainable eating choices is critical. University students are the target group of this research as they are in a critical phase that could contribute to altered dietary habits. Furthermore, eating behaviors of young adults would play an important role to shape the population's future diet. Considering the current increased use of online platforms for food purchasing – especially after the COVID-19 pandemic – investigating approaches that could be effective in the online environment is crucial. The purpose of this study is to determine the effects of *nudging* on students' choices in university canteens using pre-ordering apps. The research includes an explorative stage and an experimental study. First, focus groups and a short questionnaire were implemented to investigate factors that could influence students' food choices in the university canteen and virtual settings. Students from the University of Parma (n= 24) participated in four focus groups. The information gathered from the notes, audio recordings, and answers collected during the projective techniques was analyzed using content and thematic analysis. Then, an online questionnaire was developed to further investigate the outcomes of the focus groups. Respondents (=184) were university students enrolled in several Italian universities. Basic descriptive statistic was used to describe the samples in terms of socio-demographics, attitudes towards logos, and healthy and sustainable dishes. Finally, using the results of the explorative stage, an online questionnaire was developed, and it will be administered to 1400 Italian students in April 2022. The study will investigate the effects of different nudges (a logo and the dishes placement) on students' food choices in the context of preordering online apps of the university canteen. The findings will also contribute to providing professionals (e.g., catering managers, nutritionists, etc.), researchers, and policymakers with evidence-based recommendations.

## Chocolate consumer preferences and profiles considering lifestyle and socio-demographic features

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The aim of this study was to investigate the consumer purchasing and consumption preferences towards chocolate. A total of 657 consumers were interviewed online using a questionnaire structured in three main sections: the first about individuals' socio-demographic, lifestyle and diet information, the second on chocolate purchasing and consumption habits and the last one dedicated to the implementation of the Best-Worst scaling (BWS) methodology. The BWS allowed to define the relative importance assigned by consumers to 12 attributes describing chocolate. Additionally, the Latent Class analysis was employed to identify different preference-based chocolate segments that were characterized considering individual's lifestyle and sociodemographic features. The main BWS findings showed that "typology", "brand" and "label information" were the most important attributes for chocolate choices. On the contrary, "certifications", "ethical attributes" and "packaging" resulted not important in the choice process. From the Latent Class Analysis, 5 consumer groups were identified. The first two clusters *Fidelity consumers* cluster (21%) and *Price Sensitive consumers* (20.0%) included, in both cases, respondents who were attentive to the brand and price of chocolate. However, they differed in terms of purchasing habits: in fact, the former chose chocolate quickly and quickly, while the latter used a more rational purchasing process. The third cluster, *Sustainable consumers* (17%) was careful and informed about sustainability, considering important the cocoa origin and preferring eco-friendly brands. The *Healthy consumers* cluster (20.0%) paid attention on chocolate health claims, origin, and certifications. Finally, *Traditionalist's consumers* (22%) based its choices on product aroma and taste and was loyal to a knowledge brand. This research has shown that the choice and purchase decisions of chocolate consumers are multifaceted and complex, as they are influenced not only by the product characteristics, but also by the lifestyle of individuals, their eating habits and socio-demographic characteristics.

## Strategic packaging: the use of bundling for the valorization of local Extra-Virgin Olive Oils

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Extra-Virgin Olive Oil (EVOO) production represents an important share of Mediterranean economies and a valuable source of fat for healthy diets; therefore, the valorization of higher quality and healthier olive oils will improve the welfare of both producers and consumers.

Currently, local PDO products suffer from three main problems: 1. the lack of success of PDO productions in terms of market share and price; 2. the lack of appreciation of superior quality oils characterized by bitter and spicy notes from consumers; 3. the lack of education of consumers to a differentiated EVOO use in the culinary preparations.

The project BOXNARRANTE<sup>s</sup> aims to provide a practical solution to these problems by providing an innovative packaging for EVOOs. This box contains three types of EVOO with different origins: 1. PDO; 2. PGI; and 3. 100% Italian. They have different taste characteristics (with respectively decreasing bitter and sensory notes) and different prices (PDO is the most expensive and the 100% Italian the most affordable). By purchasing this box the consumer can buy an assortment of EVOOs covering all the needs of the different culinary preparations done throughout the week with an affordable price achieved by combining different prices products. The label reports storytelling elements that valorize the quality and taste characteristics of the products and inform about the best pairings between the contained EVOOs and the family meals.

In the end, the presented product concept achieves the following aims: (1) Informing how the sensory profile of the products can influence their healthiness; (2) suggest in detail how to use different EVOOs, suggesting the most suitable pairings; (3) provide a new package capable of having an acceptable medium price among the three bottles, which provides detailed information and contributes to forming new use behaviors such as having "Different oils for different uses".



## ***Lemna minor* and digestate: opportunities for a circular economy**

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The core idea of the experiment was to grow duckweed (*Lemna minor*) on digestate to increase the nitrogen (N) content and to obtain an organic fertilizer and a biomass to produce biogas, in a circular economy view. Duckweed is an aquatic invasive weed representing an issue for farms using greenhouse floating production system. Due to the high multiplication rate, is a strong competitor with cultivated crops. However, in the last few years it received a growing attention for nutrients (N) removal from wastewaters. Preferred N source of duckweed is  $\text{NH}_4^+$  which, although above a certain concentration it becomes inhibitory, represents the most available N form in digestate. In this experiment we compared different digestate dilution (0 and 50 mgN/l, 0N and 50N respectively) for duckweed cultivation. Experiments were carried out for 4 weeks in two periods (spring/autumn) to understand the duckweed growth dynamics. Based on N-removal potential and elemental composition, we estimated duckweed fertilization potential and biogas production (Buswell equation). Higher yields were obtained at 50N in spring (2.8 kg/m<sup>2</sup> fresh weight) suggesting the photoperiod/temperature sensitivity of duckweed. Observations reports a relevant N-removal potential with a maximum production of approximately 26 kgN/ha at 50N in spring. Elemental composition of duckweed doesn't change much between treatments and differences in biogas production have depended to yields. Similar biogas potential production with maize was observed (0.406 and 0.484 m<sup>3</sup> CH<sub>4</sub>/kg biomass, respectively) suggesting a new scenario for duckweed use as biomass for renewable energy production. This is emphasized by the fact that maize produce approximately 50 tonnes/ha of biomass in 4-5 months compared to the monthly yields of duckweed of about 30 tonnes/ha. Large scale-duckweed cultivation could conflict with food-crops reducing the sustainability of agricultural systems. Nevertheless, our results suggest new approaches for duckweed management shifting from an issue to a resource.

## Sporeforming bacteria during one year storage of UHT milk at different temperatures

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Aerobic sporeforming bacteria, such as *Bacillus* spp. and *Paenibacillus* spp., are a significant concern for dairy industry. Spores can survive to the pasteurisation process and become dominant in heat-treated milk, in absence of post-pasteurisation contamination of the product. Some sporeformers can cause off-flavours and structural defects in milk due to the production of proteases and lipases. Considering the increasing demand of UHT milk worldwide, especially in Asian countries, producers are oriented in expanding the shelf-life of this product up to 12 months. The aim of this research was to evaluate microbiological and chemical quality of UHT milk samples stored at two different temperatures for one-year. Samples were stored at 20°C and 35°C to simulate commercial storage (in a controlled environment) and transport conditions, respectively. Specimens were analysed after 1, 2, 3, 4, 5, 6, 8, 10, 12, and 14 months of storage. Changes in aerobic spore count, pH, colour, non-casein nitrogen (NCN) and non-protein nitrogen (NPN) were evaluated. For the count of aerobic sporeformers, each UHT milk sample was centrifuged and the pellet was resuspended in a peptone physiological salt solution. The resuspension was heat-treated at 80°C for 12 minutes and pour plated in Nutrient Agar. After incubation at 30°C for 2 days, the grown were isolated and molecular identification was performed using analysis of 16s rRNA. Results showed that sporeformer bacteria survived ultra-high temperature process in milk: the mean content value during one-year storage was less than 10 spores/L and there were no significant differences between storage conditions. *Bacillus licheniformis* and *B. pumilus* were dominant in the early stages of the shelf life, whereas *Brevibacillus* spp. become dominant at the late stages. On the other hand, samples stored at 35°C showed a greater change in colour and decrease in pH than those stored at 20°C.

## **The influence of greenhouse climatic conditions on 13 strawberries varieties in quantitative and qualitative traits**

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The microclimate inside the greenhouse is affected by the transfer of heat and mass among the plants, the air and the greenhouse plastic cover. Specific climatic gradients can cause significant differences in crops yield and qualitative traits, as well as the development of diseases. Air temperature and humidity are the two most important parameters that directly influence the uniformity of crops growth. A precise and accurate indoor climate condition monitoring system is needed to evaluate the crops growth and to control the effects of climate on them. This study reports the results of fall and spring monitoring campaigns carried out to assess climate conditions (air temperature and humidity) inside a low-tech greenhouse in Veneto region where 13 new strawberry varieties were grown. For each variety, the specific response to climatic conditions was also monitored. Regarding phenological results, the flowering phase was earlier in the variety 9 than the others. Nutritional status, indirectly evaluated using the SPAD index, ranged from 22.1 (variety 2) to 51 (variety 6). High variability was observed among varieties considering both production and qualitative traits. The air temperature and humidity were not highly variable inside the greenhouse and apparently, they did not influence the strawberry response. However, this contribute reports the preliminary results of a pluriannual study which is still ongoing. In the next years the experiment will be replicated, and the more complete dataset will be used to build a three-dimensional thermal model of the growing facility. This tool, in combination with the agronomic variables recorded for the crop, will allow to identify a predictive map of strawberry production performance.

## Nutrient's uptake and quality of lettuce and curly endive in aquaponics systems is affected by supplemental LED lighting

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Aquaponic is a high resource and cost-efficient integrated production system that relies on fish feed as the only input for growing both fishes and vegetables. However, in coupled Recirculating Aquaponics systems, plant ability to absorb nutrients may be reduced due to suboptimal pH conditions or in the winter months due to the lack of solar radiation, which in turn could cause accumulation fish toxic by-product containing of nitrate, nitrite, and ammonia in the water, forcing growers to renew the water more often. The aim of the study was to evaluate the growth, physiological response, nutrient uptake, and quality of lettuce (*Lactuca sativa* L.) and curly endive (*Cichorium endivia* var. *crispum*) grown in floating raft in combination with tilapia (*Oreochromis niloticus* L.), under natural light (NL) or natural light integrated with 16 hours of supplemental white LED lighting (IL, PPFD:  $173 \mu\text{mol m}^{-2} \text{s}^{-1}$ , DLI,  $10 \text{ mol m}^{-2} \text{d}^{-1}$ ) in a Recirculating Aquaponic System (RAS). Results show a species-specific response to the lighting regimes. Compared to NL, despite IL promoted plant growth only in curly endive, plant nutrient use efficiency and quality increased in both species. In both species, leaf net photosynthesis was promoted by supplemental lighting. However, it decreased the maximal photochemical efficiency ( $F_v/F_m$ ) in lettuce. In addition, the different lighting regimes affected nutrient accumulation and translocation in both leaves and roots. To conclude, based on our findings, curly endive performs better than lettuce in aquaponics. Supplemental lighting can guarantee a stable filtration capacity improving overall system performances and plant qualitative attributes (i.e. ascorbic acid content).

## Application of LCA methodology to a Recirculating Aquaponics System (RAS) prototype

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In the increasing urbanization and food consumption current scenario, the aquaponics system is generally regarded as an environmentally friendly food production system as well as a sustainable agriculture practice. However, its environmental burdens (energy consumption, materials, etc.) were not yet deeply investigated. In order to systematically assess aquaponics environmental performance, it is important to take the whole life cycle into account. The aim of this study was to identify and to evaluate the environmental impact of a Recirculating aquaponics system (RAS) prototype compared to hydroponics, using a life cycle assessment (LCA) methodology. Leafy vegetables (i.e. lettuce (*Lactuca sativa* L.) and curly endive (*Cichorium endivia* var. *crispum*) and basil (*Ocimum basilicum* L.) were grown on floating rafts in combination or not with tilapia (*Oreochromis niloticus* L.) and two different lighting regimes. In agreement with the ISO 14040, our LCA analysis included four different steps: 1) Definition of the goal and scope of the study; 2) Life cycle inventory (data collection); 3) Life cycle impact assessment (data translation into environmental indicators); 4) Interpretation and analysis of the results. The OpenLCA 1.6 software was used for calculation. Our preliminary results suggest that electricity and fish food were the main contributing factors to environmental impact. This LCA study can be useful for providing the groundwork to reduce the potential environmental impact of aquaponics systems.

## Betacyanin accumulation and physiological responses in *Beta vulgaris* L. leaves under N starvation and salt stress

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Salinity has become an important constrain in the Mediterranean area even in reason of the Climatic Change. *Beta vulgaris* has been identified as salt-tolerant species likely thanks to its adaptive betacyanin accumulation. For the red-purple colour and their characteristic glycosylated form, betacyanins were commonly compared with anthocyanins in terms of their ecological roles. Moreover, the presence of nitrogen (N) in betacyanin rings resulted contradictory with their accumulation as plant defence against poor soil conditions. In the present work, *B. vulgaris* L. var. Bull's blood (constitutively red-purple leaves; RB) and *B. vulgaris* L. var. Robuschka (constitutively green leaves producing betacyanins under stress; GB) plants were hydroponically cultivated, subjected to salt stress (SS; 150 mM NaCl) and to different N supplies in the growing medium (0, 10, 40 mM NH<sub>4</sub>NO<sub>3</sub>). Gas exchange parameters, chlorophyll fluorescence and betacyanin accumulation were measured after two (T1), three (T2) and four (T3) weeks of treatment. At T1, no-SS RB leaves showed a dose-dependent response in which the lowest CO<sub>2</sub> assimilation rate (P<sub>n</sub>) was found in plants subjected to N starvation (0 mM NH<sub>4</sub>NO<sub>3</sub>). Conversely, no-SS GB leaves showed a reverse pattern in which P<sub>n</sub> values resulted higher in N-starved plants. At T3, N starvation effect was reverted; in RB leaves photosynthetic values were higher than those found at T1 while the opposite occurred in GB leaves. Moreover, in GB, higher betacyanin content was found in non-SS than in SS leaves. Differently, at T1, RB leaves showed a coherent increase in betacyanin level following a nitrogen dose-dependent response but irrespectively to SS. These preliminary results indicate that *B. vulgaris* leaf reddening due to betacyanin accumulation might be a key adaptive strategy for coping with N starvation and not-SS. However, further investigation is needed to understand the differences between green and red-purple leaf species and to deepen the betacyanin ecological roles.

## The effect of microplastic foliar exposure on rhizosphere ecology in tomato plant (*Solanum lycopersicum* L.)

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In agroecosystems, a considerable amount of microplastics (MPs) reach the crop plants through aerial depositions. Here, we investigated the impact of MPs aerial depositions on the growth, root metabolome, and bacterial and fungal communities associated with the rhizosphere of tomato (*Solanum lycopersicum* L.). Tomato plants were treated with three concentrations (10, 100, and 1000 mg L<sup>-1</sup>) of a solution composed of polyethylene microspheres (PE-MS) and distilled water. Control plants were supplemented only with distilled water. The leaves were sprayed with the solutions after 15 and 21 days of growth. After 31 days of growth, shoots, roots, and rhizosphere soils were collected. The plant material was used to measure shoot biomass, shoot water content, and root metabolomic profiling through high resolution gas-chromatography/mass-spectrometry. Rhizosphere microbial diversity was investigated via DNA metabarcoding of the bacterial 16S rRNA gene and fungal ITS2 region. Tomato shoots did not show differences in dry and fresh weight, but a significant reduction in water content was observed at 100 and 1000 mg L<sup>-1</sup>. PE-MS aerial depositions increased the roots relative abundance of amino acids but also of carbohydrates and their conjugates. PE-MS significantly decreased the relative content of a pyrimidine derivative (5,6-Dihydrouracil), organic acids (lactic acid and tartaric acid), and fatty acids (palmitic acid and stearic acid). Further, microbial analysis revealed that PE-MS affected bacterial but not fungal beta-diversity. In conclusion, exposure of tomato leaves to PE-MS showed no effect on plant growth. Oppositely, PE-MS significantly altered the root metabolome and the bacterial diversity in the rhizosphere.

## **Blockchain technologies: the potential for application in forest nursery traceability systems**

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Abstract (max 300 words): Traceability is the possibility to trace a process, from raw material supply to consumption and disposal, in order to follow its steps in a transparent way and to improve the perception of quality in an area in which consumers are increasingly interested. Most forest nursery traceability systems are centralised, as each piece of information has to pass through a central point. These systems, however, are more susceptible to error, tampering and loss of data, resulting in less trust and dissatisfaction on the part of consumers. Blockchain technologies are based on a decentralised and distributed system and can facilitate the process of tracking and digitising data. Our aim is to investigate the potential and economic feasibility of a blockchain-based forest nursery traceability system. The methodological framework adopted covered: i) the development and deployment of a traceability web application with blockchain architecture; ii) the analysis of implementation cost; iii) the demand analysis on knowledge of traceability and blockchain technologies and willingness to pay a premium for products tracked. The first phase involved ab.100 forest plants tagged with NFC tags and tracked with the web app in three different nurseries distributed along the national territory. The second and third phases are in progress and include a scenario costs analysis of a BAU situation and a blockchain situation. In detail, a customer questionnaire has been designed and more than 70 questionnaires were collected so far. The research represents one of the first concrete cases of blockchain in the forestry nursery market and the preliminary results show the advantages of traceability, in guaranteeing security, product quality, transparency and liability. In addition, anticipating the maturity phase when this technology will be used on a mass scale, it will be important to verify the level of consumer knowledge and their willingness to pay for the technology.



## Effects of vegetation parameters on LAI as a proxy for mountain grassland yield estimation

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Leaf Area Index (LAI) is a well-known parameter used for yield estimation of grasslands. The great availability of LAI measurements retrieved from satellite imagery allows for yield estimation at great spatial and temporal resolutions. In order to test the reliability of the yield predictions for a specific system, it is, although, important to assess the relationship between yield and LAI through ground measurements and to test for potential variables influencing it. A validation campaign of ground-measured LAI has been carried out between April and October 2021 at eight differently managed grassland sites in the Italian provinces of Bolzano and Trento. In addition to yield and LAI, visual assessments of following parameters were taken: the estimate of the yield proportion of grasses, forbs and legumes, the phenological stage of the vegetation on an 8-level-scale (vegetative stage, stem elongation, begin heading, mid heading, full heading, begin flowering, mid flowering, end flowering, and senescence), forage moisture on a 3-level-scale (dry/moist/wet) and lodging on a 4-level-scale (no lodging, light lodging, medium lodging, heavy lodging). The influence of these parameters on the relationship between yield and LAI was explored. The results indicate that sward type (based on the yield proportion of grasses, legumes and forbs) and the growth cycle affect the relationship between yield and LAI. Swards rich in grasses displayed a stronger increase of yield as a function of LAI compared to swards rich in forbs and especially to those rich in legumes. Similarly, also measurements taken during the first growth cycle resulted in a steeper relationship compared to subsequent growth cycles.

## **SOIL BANK project: invest on soil**

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SOIL BANK project's goal consists in the recognition of the soil resource as a fundamental asset for agriculture and environment, but also as an exhaustible resource which, if not ameliorated or maintained over time, can't bring eternally its positive action. For this reason, it is necessary to ensure that farmers make investments on the soil, meaning it like a capital on which to invest. The Soil Organic Matter (SOM) content is one of the most contributing factors in conserving or enhancing soil quality and the related ecosystem services. To increase SOM, SOIL BANK project aims to follow two different agronomic strategies: organic fertilizer application (compost vs. digestate) and cover crop use (triticale vs. cover crop rotation vs. no cover crop). An open-field experiment is running since 2020 with this purpose at San Donà di Piave (VE, Italy), with maize as cash crop. Not significantly different cover crop dry biomass production was observed among treatments at the termination time, with an average weight of 0.71 Mg ha<sup>-1</sup>. Not even comparing corn plants' growth at fourth leaf stage, measuring heights and leaf greening with SPAD index, differences were observed. Similarly, plants grown with compost did not significantly diverge from those treated with digestate. Since the biomass obtained in the no cover crop treatment was due to weeds, an equal weight reached sowing cover crops resulted in a much less weed occurrence, hence using cover crop can limit the related issues and management costs. This, coupled with the fact that cover crops did not bring to corn growth reduction and that their final biomass was not inferior than that of the no cover crop treatment, leads to conclude that cover crop use can represent a competitive choice for open-field corn culture.

## **Potential of cultivation of oats (*Avena sativa* L.) for human consumption in South Tyrol**

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Cereals were important crops of South Tyrol in the past centuries, as farmers used to produce their own staple food. From 1900 to 2000 their cultivation was almost abandoned, but ten years ago with the project "Regiokorn" a value chain for rye and spelt was established with success. Due to a growing appreciation for regional products and the necessity of field rotation, an agronomic field experiment was conducted to investigate the agronomic performance of 16 different oat varieties, two of whom were free threshing varieties. The study was conducted in a mountain environment in Dietenheim (South Tyrol, NE Italy) in the years 2018, 2019 and 2021. The experimental design was a randomized complete block design with three replications and a plot size of 4.8 m<sup>2</sup>. We investigated if the standard quality requirements for food use can be achieved in this environment. The examined agronomic traits were grain yield, hectolitre weight, thousand seed weight and kernel sorting. Statistical analyses showed that all varieties had the same yield over three years; only free threshing varieties had significant lower yield than the other oat varieties. For hectolitre weight only six varieties complied with the required quality standards; therefore this parameter could be seen as the limiting factor. Concerning thousand seed weight and kernel sorting, all varieties reached the minimum quality standards.

## Validation of a novel sensor for non-destructive online monitoring of plant water status

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The improvement of agricultural water management is of paramount importance considering the expected increase in water demand for the near future. Leaf-Water-Meter (LWM) is a novel leaf sensor allowing non-destructive online monitoring of plant water status, based on the photon attenuation of the passage of radiation through the leaf. LWM signal (dehydration level, DH) was validated by comparing DH with physiological measurements performed on multiple plants species in three different experiments. In the first experiment, plants of *Acer platanoides* L., *Citrus limon* L., *Olea europaea* L., *Arbutus unedo* L. were subjected to multiple cycles of dehydration (by withholding water) and re-hydration to compare DH with leaf relative water content measurements ( $RWC_{leaf}$ ). In the second experiment, the relationship between DH and leaf water potential ( $\Psi_L$ ) was investigated in well-watered and water-stressed *Quercus ilex* L. seedlings. Water stress was imposed by providing 70% of transpirable soil water fraction for two weeks. In the third experiment, stomatal conductance ( $g_s$ ) was monitored on *Viburnum tinus* L. and *Photinia x fraseri* Dress. potted plants subjected to a progressive water stress similar to the one applied for *Q. ilex*. The good agreement obtained between leaf water relation parameters ( $RWC_{leaf}$  and  $\Psi_L$ ) and DH values of the first two experiments, highlighted the great potential of the LWM sensors for monitoring water status in multiple species characterized by different leaf traits. Furthermore, a significant relationship was also observed for DH and  $g_s$ . Our results demonstrate that LWM sensor is a non-destructive and reliable device to monitor leaf water status continuously during water stress progression. The future development of automatic LWM sensor technology may contribute to the optimization of irrigation scheduling and to enhance precision irrigation management both for food and ornamental plant production.

## **The Tuscany Living Lab for increasing the digitalization of agricultural advisory service for olive growing**

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Digital tools including Decision Support Systems (DSS) may contribute to increasing the sustainability of agriculture by supporting farmers and advisors in optimizing the application of agricultural inputs. However the diffusion of digital tools in agriculture is still limited, especially among farmers. Encouraging their adoption by agricultural advisors could be an efficient way of increasing the digitalization of agriculture, indirectly involving also smallholder farmers that are usually unwilling to invest in new technologies. In this context, the H2020 FAIRshare (Farm Advisory Digital Innovation Tools Realised and Shared) project is aimed at supporting farm advisors to use Digital Advisory Tools and Services (DATS) to increase the productivity and the sustainability of agriculture. Within the FAIRshare project, AEDIT is leading the Italian User Case "Tuscany Living Lab to support a wider adoption of sustainable crop management DATS", focused on olive growing. The aim of the Tuscany Living Lab is to develop a unique DATS for olive growing, with advisors as cocreators, using a user-centric approach for sensing, prototyping, validating and refining solutions in a real-life context. The DATS will be a web-app for olive growing including DSS for fertilization, irrigation and olive fly control, using both traditional models and machine learning techniques. Moreover, the web-app will include a tool for the monitoring of the infestation of olive fly and a tool for the self-evaluation of biodiversity preservation at the farm level to be used for marketing purposes. The advisors accessing the web-app will also access a chat for the communication with farmers, that will allow them to send alerts and exchange data. The development of the DATS will be carried out using a living lab approach in which interactive workshops are organized with the User Case stakeholders to define the vision of the User Case and the business plan.

## **Smart Land South Tyrol: optimized irrigation scheduling through soil moisture tension measurements with sensor-based, wireless technology**

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Irrigation in agriculture is essential for the maintenance of agricultural production, supporting crop growth and fruit development especially during periods of prolonged drought. Considering future climatic scenarios and increased pressure for sustainability, the need of support systems for irrigation scheduling is growing. The lack of precise data about the water status of soils is generally the main factor limiting the improvement of irrigation efficiency. The Smart Land project aimed to close the gap by developing tensiometers able to transfer real time data about soil moisture tension through LoRaWAN and provide their visualization on a Smartphone Application. Today, a network of low power, wireless communication technology has been established throughout the main growing areas of the region of South Tyrol. During the test phase, 120 tensiometers were placed in apple orchards, which allowed the measurement of soil moisture tension during the growing seasons of 2019-2020. Datasets transferred over adapted low power transmitter modules placed on the sensing devices showed the potential for a reduction in irrigation water supply with no negative effect on crop quality and quantity in several apple orchards. This confirms that the utilization of the developed technology can support precision irrigation practices to enhance water use efficiency in wide areas. We predict that a continuous flow of information about soil moisture tension can provide objective datasets for optimal irrigation scheduling for farmers. The development of tensiometers equipped with wireless information transfer through LoRaWAN is a first step towards an easy, ready-to-use technology which allows to have insights into a crucial parameter for plant water uptake. This technology enables the end user to rely on the punctual measurements of the sensing devices, leading to a more objective decision path for orchard irrigation.

# The effect of Blue:Red light proportion on germination parameters, growth attributes, and quality of borage (*Borago officinalis* L.) sprouts

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Sprouts (i.e., young seedlings) are widely recognized as a source of secondary metabolites, i.e., phytochemicals, to which many health benefits are attributed. Borage (*Borago officinalis* L.) is native to the Mediterranean region, with widely recognized beneficial properties due to the phytochemicals, mainly phenolic compounds, extracted from different organs of this plant. Borage represents an underutilized species in the ready-to-eat market, but recently borage sprouts were found rich in phenolic acids and antioxidants. It appeared worthwhile to investigate in depth borage sprouts, as a novel functional food with high sensory and bioactive characteristics. Blue and red light affect the phytochemical content in sprouts of many plant species. This work aimed to study the effect of blue:red light proportion on the germination (germination %), growth (i.e., biomass, sprouts lengths, etc.), and quality traits of borage sprouts. Seeds were incubated over sterile cotton contained in plastic trays, wetted with distilled water and placed for 8 days under six LED (light emitting diodes) light treatments having the same total incident photon flux density (PFD) of  $200 \mu\text{mol m}^{-2} \text{s}^{-1}$ : only blue; only red; three blue:red combinations at different proportions of total PFD (75:25%, 50:50%, and 25:75%); a wider spectrum and a dark treatment selected as controls. Preliminary results indicated that light treatments significantly affected some growth attributes of sprouts. Further analyses were planned to evaluate the effect of light treatments on the pigments, the nitrate concentration, the phenolic content, and the antioxidant activity.

## Assessing the carbon stock in a Mediterranean forest plantation through UAV-LiDAR data

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Forest ecosystems play a key role to counteract the negative impacts of climate change because they can capture carbon dioxide from the atmosphere and store it in tree biomass. Moreover, they play significant role to protect soil and water. Accurate measurement of forest carbon stock became a crucial requisite for implementing sustainable forest management with a climate-smart vision. Over the years the use of Light detection and ranging (LiDAR), particularly Airborne Laser Scanning mounted on Unmanned Aerial Vehicle (UAV) is increased for forest inventory and planning activities at local and regional scale because it is considered a suitable technology for describing the forest covers remotely and accurately. The aim of this study is mapping the carbon stock stored in a Mediterranean forest plantation created to protect the water quality of Occhito Lake located along the edge between Molise and Apulia (South Italy). The most frequent tree species planted are coniferous (i.e., Aleppo pine, and coniferous spp.). To reach this aim, the area-based approach (ABA) was implemented using LiDAR data collected by a sensor mounted on UAV device. Field measurements (i.e., tree height, tree diameter, and tree position) were collected through Field-map tool. These measurements were used as proxy to calculate stem volume and carbon stock of standing trees through Italian National Forest Inventory equations. Two maps displaying the zones with the stem volume and carbon stock of forest plantation was provided. Results revealed that the stem volume estimation was slightly more accurate (Root Means Squared Error 'RMSE' = 1.28; R-squared = 0.789) in comparison with carbon stock (RMSE = 0.36; R-squared = 0.729). The study was realised within the activities of P.A.B.L.O project, a regional project sponsored by the rural development program of Apulia Region.



## Chitosan nanocarriers-mediated delivery of double-stranded RNA “in planta”

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Agriculture is currently facing numerous challenges: the rapid rise of the world population, the consequent growth in food demand, the global decrease in crop yield. Particularly regarding the last issue, climate change is worsening the environmental stresses that commonly affect crops, and the use of resources – such as fertilizers and pesticides – is highly inefficient and pollutant. In this context, research is looking for new approaches to improve crop productivity by more efficient and environmentally friendly practices. It has been shown that nanomaterials are suitable for the development of cutting-edge technologies with the aim of improving the delivery of bioactive substances on plants and to promote their resistance to biotic and abiotic stresses. Among organic polymers, chitosan, if used in the nanoscale form, shows both properties; it can induce biological responses concerning plant defense against diseases and pathogen attack, and it is particularly suitable as a carrier for several molecules. Another innovative method for the defense of crops is the exploitation of the spray-induced gene silencing (SIGS) based on the activation of the so-called RNA-interference (RNAi). It involves exogenous double stranded RNAs (dsRNAs) targeting an essential pathogen gene, which trigger the RNAi pathway leading to the translational repression by degradation of target homologous mRNAs. In our case, the research aimed to verify the feasibility of dsRNA distribution on plant surface by means of functionalized chitosan nanoparticles (CH-NPs), thus allowing the protection of the doping agent and its efficient delivery. Here we show the preliminary results regarding the characterization of CH-NPs, their loading with dsRNAs and their interaction with the leaf surface of *Nicotiana benthamiana* plants. The effects of the dose-dependent distribution were analyzed by confocal microscopy upon incorporation of a fluorescent probe.

## An RNAi-based approach for non-chemical management of the troublesome weed *Amaranthus hybridus* L.

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The European legislation on the sustainable use of agrochemicals intends to reduce reliance on herbicides and encourage integrated weed management strategies by developing innovative solutions that are more environmentally sustainable and less dangerous for animal and human health. Herbicides are currently the most effective tool for weed control, however continuous use on the same active ingredients has resulted in the evolution of resistant biotypes. Due to lack of new herbicide sites of action (SoA) in the market, new weed management technologies should be developed. In this regard, the development of RNA interference (RNAi)-based technologies could: i) represent a potential improvement in non-chemical weed control; ii) provide an emerging GMO-free strategy for managing invasive and resistant weeds; and iii) provide a valid opportunity to look inside the molecular mechanisms of weed biology. *Amaranthus hybridus* L. is a monoecious and self-pollinated weed that has evolved multiple resistance to herbicides with different SoA including acetolactate synthase (ALS) inhibitors, which are the most used herbicides in soybean. ALS represents an optimum target for gene silencing mediated by dsRNAs because it is an intronless, nucleotide-stable, and single-copy gene. Given these characteristics, *A. hybridus* ALS gene was chosen as the target of silencing to investigate the effectiveness and applicability of a non-chemical RNAi-based approach for endogenous gene silencing and weed control. In this study, we first identified three distinct dsRNA molecules of various lengths targeting different regions of ALS gene, which were then synthesized in vitro and externally applied to the abaxial leaf surface of *A. hybridus* plants. The transcript level of ALS gene was found downregulated when synthetic ALS-dsRNAs were applied to the leaf. Our ongoing research focuses on developing application strategies and dsRNA delivery systems to maximize the efficiency of gene silencing by exogenous dsRNAs, and on *A. hybridus* RNAi pathway genes expression analysis.

## *Arachis hypogaea* L. cultivation in Campania: technical and operational characteristics of mechanised

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This study is part of a larger project to reintroduce peanut (*Arachis hypogaea* L.) cultivation in the Campania region. The aim is to place production in the inland areas of the region, which are marginal agricultural areas and almost always used for cereal and fodder crops. The experimental activities were aimed at identifying the most suitable cultivation and mechanisation techniques for the area. Sowing took place in the Avellino and Caserta areas using a cultivar of Bulgarian origin. Mechanized harvesting represents one of the main critical points in peanut cultivation because it is carried out with specific machines for peanuts in two parts: the first phase, generally done with a digger-inverter, consists of digging the plant and turning it over in the field to ensure initial drying of the pods. The second step, done with a pod separator operating at a fixed point, is to separate the pods from the biomass. In this experiment, a machine already present in the area for other crops was adapted and used for the first stage, and a fixed-point pod separator from China was used for the second stage. The objective was therefore to evaluate the harvesting efficiency and performance of the pod separator machine by assessing its working capacity, product losses and the level of product cleanliness (impurities in the separated product). These evaluations were carried out under two different conditions: whole plants or plants with reduced biomass from previous mowing. The results showed the reliability of the separating machine in terms of working capacity and the pre-harvest mowing has a significant influence on the separation efficiency. In conclusion, the objective of improving the peanut harvesting phase can be achieved, which would lead to an increase in the profitability of the crop in Campania as well as throughout the country.

## **Smart Glasses for Augmented Reality: a valid support during maintenance activities**

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Nowadays, digital transformation can be seen in many of the productive sectors. The spread of digital technologies has been occurring also in agriculture, where digital devices and services are introduced to support farmers in their daily activities. Smart glasses (SG) for augmented reality (AR), are digital tools currently studied and tested for business applications, including agricultural purposes. Scientific studies have highlighted that the SG are a valid support tool for farmers' activities and for the decision-making processes. At this moment, one of the most applied augmented reality solutions concerns the operator's assistance for maintenance activities of devices or machineries. Thus, the objectives of this work were to evaluate the performance of the SG Vuzix M400 (USA), with a video display system, testing its available functions in a perspective of assistance scenarios for farmers or technicians. Moreover, a specific workflow for periodic checking of milking machines was built in AR. The experiments have been performed assessing the AR information detection capabilities, the video call quality, the battery life, and the voice control system. The results showed the capabilities of SG to reach the AR information from a long distance, in a short time interval and to transmit audio-video with a high level of detail allowing discriminate small objects during remote assistance with reduced delay. Furthermore, the M400 SG can be managed completely hands-free with the voice control system when the noise level is less than 70-75 dB. The developed maintenance scenario represents an example of AR remote assistance application in the agricultural context, highlighting the great potentiality of SG devices to support both farmers and technicians to solve problems efficiently. Complex applications or assistance scenarios for the agricultural sector should be developed considering the farmers' needs as well as the cloud computing services.

## **MULTIFRU project: Multifunctional Valorisation of Pigmented Wheat Varieties for production of Flavonoid Reach Foods and Nano-Agrochemicals for Sustainable Wheat Protection**

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Anthocyanins are abundant secondary metabolites responsible for most blue to blue-black, and red to purple colours of various plant organs. They have many interesting properties, first of all antioxidant activity. Several researches show as polyphenols, like anthocyanins, can contribute to maintain human health, showing considerable antioxidant activity, with antitumor and antimutagenic properties, beneficial effects on diabetes, and heart diseases. Polyphenols are also beneficial for the plant itself as physiological active compounds, as stress protecting agents and by their significant role in plant resistance against plant pathogens. Pigmented wheat cultivars represent an interesting category of wheats with flavonoids-rich kernels, due to the presence of anthocyanins in pericarp layer (purple pericarp varieties) or in aleurone layer (blue aleurone varieties). MULTIFRU is a regional and multidisciplinary project focused on 3 purple and 2 blue aleurone bread wheat cultivars. The main topics are: 1) production of flavonoid-rich functional foods with protective activities for promoting human health using cultivar rich in polyphenolic compounds; 2) studies about plant – pathogen interaction, in particular between pigmented wheat plants and fungal pathogen *Fusarium graminearum*, causal agent of Fusarium Head Blight; 3) use of wheat production chain wastes (like bran and straw) to develop new and green pesticides for Fusarium Head Blight management and control. The main purpose of the project is to exploit the beneficial properties of the polyphenols present in the kernels for the production of functional foods (bread and pasta) and to valorise and reuse the wastes from the wheat supply chain using a circular economy approach. This work shows some preliminary results obtained in the first year of the project.

## **Evaluation of the variability of C mineralization in different management zones defined by soil electrical conductivity mapping of a rice field**

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Soil, thanks to its intrinsic characteristics, has the ability to store organic carbon (OC) for long term, reducing atmospheric CO<sub>2</sub> and providing benefits in terms of soil fertility. This property is specially related to soil texture. Stable soil OC content increase is related to percentage of fine soil particles ( $d < 50 \mu\text{m}$ ). In the study field, two different management zones have been identified by means of geoelectric scanning: one zone with high electrical conductivity (HEC) and one zone with low conductivity values (LEC) characterized by coarser texture. In order to estimate the OC mineralization kinetics and the OC stabilization in the two zones, a laboratory incubation test was set up. Soil respiration was assessed on untreated soil and on soil treated with one of the matrices derived from sewage sludge (SS): SS treated with CaO, SS treated with CaO and CO<sub>2</sub> and SS treated with CaO and H<sub>2</sub>SO<sub>4</sub>. The matrices were added in amounts necessary to provide 250 kg/ha of N. CO<sub>2</sub> production was measured during an incubation time of 84 days. HEC soil mineralization rate was higher in the early stages of incubation than LEC, but after about 21 days of incubation the trend tended to reverse and HEC soil showed a more pronounced slowdown in mineralization rate, suggesting an effect of organic matter protection. The mineralization percentage of OC added with SSs was between 30 and 35% higher in LEC with respect HEC soil, for all three amended theses. In summary, texture seemed to affect mineralization dynamics of the added organic matter, confirming the ability of the fine fraction to fix and protect it. Differences between management zones within the same field suggest the possibility of optimize the site specific SS distribution.

## **Antioxidant spray treatment as a sustainable alternative to dipping for freshly cut apples**

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Freshly cut apples are minimally processed value-added fruit products that are becoming increasingly popular in the market. They are commonly sold with the peel on, making them attractive for organic production. Dipping apple slices in an antioxidant solution is a very efficient and common technique used to inhibit browning, however, it has microbiological, economic, and ecological limitations: the dipping tank can be a source of cross-contamination; the concentration of active ingredients in the dipping solution decreases over time with multiple immersion cycles; a large amount of reagents must be consumed to prepare the antioxidant solution and disposed of when exhausted. Therefore, more sustainable production ways could be developed to reduce the environmental impact and costs. An alternative to dipping can be spraying an antioxidant solution on the surface of apple slices. This technique has several advantages over dipping: first, because the reservoir of antioxidant solution does not come into contact with the apples, microbial contamination is avoided; second, there is no need to dispose of excess solution because it can be produced in a tailored quantity; and finally, the titer of the solution remains stable over time, allowing constant efficacy from the first to the last production batch. In this study, browning inhibition was compared between apples treated with a conventional dipping procedure and with spraying over a 5-day storage period. Color, considered as a reference parameter, was measured colorimetrically, and the presence and abundance of dark spots was assessed visually. Although a high intra- and inter-batch variability due to the use of non-professional equipment was observed, based on changes in brightness ( $\Delta L^*$ ), hue angle ( $\Delta h$ ), chroma ( $\Delta C^*$ ), and dark spots, spraying was found to be as effective as dipping. Further investigation is needed in order to optimize the procedure for industrial application.

## Effect of foliar fertilizers from fish by-product on growth and development of Lettuce (*L. sativa*) in green-house conditions

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In Liguria Region, as in all coastal areas where fish represents a relevant part of human diet, a huge quantity of fish by-product is produced. In the contest of circular economy, fish by-product could be a resource instead of an environmental issue, thus producing materials with high added value. Such an example are fertilizers or biostimulants obtained from fish by-product that can be used to increase the sustainability of crops cultivation. Under the frame of project "FISH – Fertilizzante Innovativo Suolo e Habitat" (PSR Regione Liguria 2014 – 2020 Misura 16), the effect of foliar fertilizers obtained from fish by-product were evaluated on the growth and development of lettuce (*Lactuca sativa* L. 'Summerbel') in green-house conditions. Two consecutive experiments were carried out applying seven different treatments at the 22<sup>nd</sup>, 29<sup>th</sup> and 35<sup>th</sup> day after sowing. Tap water and a commercial product (FISH-MIX®, Biobizz) were compared to: two different hydrolyzed products (H and FISH n° 13), a basified-hydrolyzed product (BH), a hydrolyzed-fermented product (FH), and a basified-hydrolyzed-fermented product (FBH). Morphological (plant height and number of leaves) and physiological (leaf chlorophyll, flavonoids, anthocyanins content and the nitrogen balance index (NBI)) parameters were monitored during crop development. In addition, at the 39<sup>th</sup> day after sowing, the fresh and dry biomass of plants were evaluated. Results showed that plants treated with H and FBH products had a significant high number of leaves, plant height, and leaf chlorophyll content. In conclusion, fish by-products hydrolyzed and basified-hydrolyzed-fermented, tested in this experiment, have shown greater efficacy than controls. However, it will be necessary to continue the assessment of project FISH products in other experiments and on other plant species.



## **Sensory and electronic nose analysis of Noble goat's milk from Cilento (Italy)**

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The interest in extensive farming which is sustainable for the environment and animal welfare has recently increased. In such context, an approach that arises from these assumptions, aimed at enhancing the quality of dairy products, is the "Noble method", applied in Cilento area (PSR Campania 2014/2020-Measure 16.1) for zootechnical productions. The disciplinary of the "Noble method" prohibits the use of silage in the livestock feeding system, provides quantitative limits for the use of energy concentrates and dictates specific rules for the botanical composition of pasture forage (at least 5 different cultivars in the ration). Due to constraints related to the feasibility of extensive grazing, the "Noble method" appears to be a promising strategy especially for small ruminants, such as sheep and goat farms. In this investigation, sensory analysis was applied to test the differences between noble goat's milk and non-noble standard goat's milk. Then, E-nose, consisting of ten metal oxide semiconductor sensors was used to classify the two types of goat's milk, in terms of rearing and feeding system, and to test the discrimination feasibility of the device. The milk samples were taken in five different times over a period of six months (lactation period effect) from 18 individual goats. In terms of sensory differences, Noble milk scored higher for descriptors such as grassy and sweet aromatic, typical of an extensive pasture farmed milk, while regular milk scored higher for descriptors such as silage and malty. E-nose resulted capable of classifying the two milk types in terms of animal feed (87.78% correct classification of milk samples according to the feeding system). Since the "Nobile method" follows a production specification, it is necessary to control and guarantee the authentic quality of the dairy products, and the E-nose has proved to be a quick, reproducible and simple method for routine control analyses.

## **Changes in qualitative traits of rocket varieties (*Eruca sativa* and *Diplotaxis tenuifolia*) as influenced by biostimulants**

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Rocket species are largely consumed as baby leaf vegetables. They belong to the Brassicaceae family and can be considered as 'functional food' due to the abundance in several bioactive compounds, such as glucosinolates, phenolic compounds, anthocyanin, vitamins, among others. Nowadays, the consumer demand for nutrient-rich products with health benefits is growing steadily. Scientific evidence has shown the positive effect of plant biostimulants on crops and demonstrated their ability to improve quality traits and nutrient use efficiency. These characteristics make them suitable to be used as agricultural tools to increase the economic and environmental sustainability. This work is part of the project MIND FoodS HUB which encourages the agri-food research and promotes food quality and sustainable production. The study aimed to evaluate the effect of two commercial biostimulants (Megafol® and Actiwave®) on quality traits of two *Eruca sativa* Mill (E1 and E2) cultivar and two *Diplotaxis tenuifolia* (L.) DC (D1 and D2) cultivar. Rocket plants grown in plastic tunnel in the North of Italy during the summer season (June-July). The biostimulants products were applied two times during the cultivation at the recommended dosage. Rocket leaves were sampled and analysed at harvest. Changes in chlorophyll content and chlorophyll a fluorescence were evaluated non-destructively, whereas several compounds, including total and reducing sugars, sucrose, total glucosinolates, nitrate, carotenoids, anthocyanins, and phenolic content were determined. The nitrate content was generally lower in plants treated with Actiwave® compared to the control. At the same time, the total sugars increased in *Diplotaxis* genotypes while they were steady in *Eruca* varieties. Megafol® application induced an opposite effect on total sugars in D2 and E1 varieties whereas no changes were observed in D1 and E2. Results showed a variability in quality traits depending on the combined effect of biostimulants and cultivars.

## Towards the unraveling of the regulatory mechanisms of starch and prolamine accumulation in durum wheat

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The challenges that modern agriculture is called to face include the development of varieties with improved yields and quality. In wheat, grain quality strictly depends on the starch and protein content and composition, therefore the molecular mechanisms underlying their biosynthesis and accumulation in the endosperm are of great scientific and industrial interest. In recent years, although major key genes have been identified and cloned in many species, regulatory mechanisms involved in these processes remain unclear, especially in durum wheat (*Triticum turgidum*). This study is focused on 4 transcription factors belonging to three major plant families (AP2/ERF, bZIP and NAC), known to be involved in the regulation of starch biosynthesis and accumulation (and prolamins in some cases), and already identified in other plant species (such as maize, rice, *Arabidopsis thaliana* and *Triticum aestivum*): i) SPA (Storage Protein Activator), positively regulates both prolamins expression and starch biosynthesis in *T. aestivum*; ii) ZmbZIP22, identified in maize and rice where it plays a negative role in starch synthesis by reducing the amylose content; iii) ZmNAC34, identified in maize, induces a reduction of total starch against an increase in the amylose fraction in rice, and iv) AtWR11, identified in *A. thaliana* and also studied in rice and wheat, exerts the role of a negative regulator on starch biosynthesis in the endosperm. After having identified the orthologous genes in *T. turgidum* relying on sequence homologies and expression patterns, their role will be investigated through a functional genomic approach based on gene silencing by CRISPR-Cas9 in the durum wheat cv. Svevo. The produced mutant lines will be subjected to subsequent characterizations aiming to understand the role of these genes in the accumulation of starch and prolamins.

## Understanding effectors: a novel way to dismantle phytoplasmas?

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Apple Proliferation (AP) is one of the most important phytoplasmoses in Europe and widespread across several major apple growing areas in many countries. The disease, associated with the phytoplasma '*Candidatus Phytoplasma mali*' ('*Ca. P. mali*'), is transmitted by sap-sucking psyllids. Symptoms of the infection include foliar reddening and formation of witches' brooms, and lead ultimately to non-marketable fruits. Currently, controlling the vector, uprooting infected trees, and using healthy propagation material represent the only treatments to prevent the spread of the disease. In the last years, extensive research has shed light on various aspects of AP, although many remain to be understood. Among phytoplasmas' most interesting characteristics is the secretion of proteins, called effectors, in the host cells, which induce alteration of the host physiological processes and provide a fitness advantage to the bacteria. Research shows how these small peptides emerge as phytoplasma key molecules, able to determine many of the visible and invisible symptoms of the disease. It appears therefore clear how the research on bacterial effectors represents not only a crucial step towards a complete understanding of the disease but may also constitute a promising approach to implement a direct, efficient, and environmentally friendly cure against Apple Proliferation. In this framework, the molecular targets of '*Ca. P. mali*' effector SAP11-like, one of the best characterized phytoplasma effector to date, have been analysed. This protein targets at least two members of the TCP (Teosinte-Branched 1/ Cycloidea/Proliferating Cell Factors) transcription factor family, involved in a large array of plant-biological processes, such as development, growth, and defence responses. Analyses of the target TCP sequences in *Malus* varieties that display a natural tolerance towards the symptoms of AP revealed the presence of several non-synonymous mutations in the nucleotide sequences of these plants. These findings led to the hypothesis that the mutations could induce conformational changes in the proteins and, consequently, reduce the binding affinity with the effector, affecting its mechanism of action.

## Molecular genetic characterization of *Castanea sativa* Mill. trees in South Tyrol

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*Castanea sativa* Mill. is the third most frequent permanent crop in the Autonomous Province of Bolzano-Bozen (South Tyrol). The present research aimed to perform a molecular genetic characterization of chestnut trees collected all over the chestnut growing area of South Tyrol, focusing on ancient trees in order to characterize the local germplasm. Leaves from *C. sativa* trees were sampled and circumference of the stems at breast height was measured in six districts. The analysis of the samples was based on microsatellite markers (SSRs). Eleven SSR loci were amplified in multiplex polymerase chain reactions and the amplicons were separated and visualized in a fragment analysis. Population genetic analysis focused on the variability of microsatellite markers and on cluster analysis, in order to evaluate the relationships among the samples in dependence of the geographic area and the estimated age of the trees. The amplified loci generally displayed a high degree of polymorphism. The highest number of identical multilocus genotypes was detected for the marrone cultivar 'Südtiroler Gelbe'. Comparing the variability among the different districts, Valle Isarco and Val Venosta harbored the highest number of distinct chestnut tree genotypes. Homonyms, synonyms and misnomers were detected in the dataset. Cluster analysis evidenced two main groups and intermediate genotypes, showing a separation of chestnut and marroni genotypes; the former group characterized by high variability, and the latter group by high homogeneity. A certain degree of admixture may be hypothesized from the results of the cluster and parentage analyses. Furthermore, molecular genetic data on the trees of the age cohort of 350 years plus indicated that the marroni genotype was introduced later to the region. The study provided an overview on the genetic variability of *C. sativa* in South Tyrol and on the distribution of genotype groups in this area.

## Foliar treatments with wood distillate boost yield and nutritional properties of chickpea seeds

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In agriculture, synthetic fertilizers are increasingly being replaced by bio-based products. Among them, wood distillate (WD), also known as pyroligneous acid, is one of the most promising. WD is a by-product of the pyrolysis process using waste plant biomass as feedstock. Such a product is plenty of biologically active substances (i.e., sugars, polyphenols, acids, alcohols, and esters), improving plant fitness and yield performance. Here, the effect of WD foliar application on the growth and yield of chickpea (*Cicer arietinum* L.) plants was investigated. During the whole growing period, half of the plants was sprayed with 0.25% (v/v) WD solution (WD-treated plants), and the remaining half was left as control. After four months, several physiological and nutritional analyses were performed on the whole plants and seeds. Plant height and weight did not vary significantly, while seed diameter (+11.24%) and weight (+33.33%), as well as the content of starch (+45.92%), total soluble proteins (+12.91%), total polyphenols (+16.36%), and the antioxidant power (+28.43%) increased. Furthermore, also the content of essential free amino acids in seeds increased, except for lysine (-3.41%), phenylalanine (-10.50%), and methionine (-13.74%), which were slightly reduced. The concentration of potassium and magnesium was decreased in WD-treated seeds, although values were within the common ranges reported in the literature. These results clearly support the effectiveness of WD on boosting the yield and nutritional qualities in crop edible parts. From a general point of view, the use of WD could represent a suitable green solution to limit the environmental impact of agriculture and alleviate human malnutrition and famine.

## **Are two biostimulants better than one? Assessment of fulvic acids and protein hydrolysates action to overcome crops Fe-deficiency**

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Since the past century, Fe deficiency has been commonly considered a limiting factor in crop production. Indeed, due to its implication in plant growth, Fe deficiency can limit, amongst all, photosynthesis and sugar production, leading to low performance and stunted growth. Besides, owing to the biochemical correlation between soil properties and Fe availability - relying on the pH-value or the content in lime - Fe deficiency has constantly brought producers to meet challenges to optimize crop production and yields. Recently, biostimulants have been exploited as tools to address these challenges, promoting tolerance to abiotic stress, nutrient use efficiency and the bio-availability of nutrients themselves. In these terms, fulvic acids (FA) and more recently animal-derived protein hydrolysates (PHs) have been under study to characterize their mechanism of enhancing plant growth. Initially, the physical interaction in solution between FA and PHs was analysed by circular dichroism (CD) spectrophotometry and isothermal titration calorimetry (ITC). Furthermore, to achieve a better knowledge about the mechanisms of action of biostimulants, commercial FA and PHs products have been applied individually and together to treat cucumber Fe-starved plants under controlled conditions. Therefore, morpho-physiological traits, such as root length and biomass, SPAD index, root acidification and Fe<sup>3+</sup> reducing activities, have been recorded. Moreover, expression analyses of genes involved in Fe acquisition are in progress. Taken together, the results here presented could shed light on the modulation caused by FA and PHs of plant mechanism involved in Fe acquisition and give information for the optimization of new biostimulant formulations.

## The effect of strigolactones on salt stress mitigation

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Phytohormones are signaling molecules derived from plant biosynthetic processes that regulate plant growth and development. Moreover, they play a key role in mediating plant response to adverse conditions, acting both where they are synthesized and at other sites where they can be transported.

Strigolactones are emerging phytohormones involved in controlling root hyphal branching processes and plant communication with several rhizosphere organisms, including arbuscular mycorrhizal fungi. During the symbiosis, arbuscular mycorrhizal fungi obtain carbohydrates from their host plant and, in return, the plants obtain water and mineral nutrients (mainly phosphorous and nitrogen) from their fungal partners.

In recent years, research on SL has increased markedly as their potential activity in regulating many physiological and molecular pathways in plants to overcome abiotic stress has been discovered.

In the present work the effect of exogenous SL on tomato plants exposed to salt stress was tested. For this purpose, 15  $\mu\text{M}$  SL was exogenously applied to this crop subjected to stress conditions (150 mM NaCl) to evaluate the mitigator effect of this phytohormone.

Both photosynthetic parameters and antioxidant enzymatic activities were analysed and combined to an untargeted metabolomic approach for unravelling the molecular mechanism that leads to mitigating the stress.

The results indicated that SL counteracted the negative impact of salt stress on plants by maintaining the proper functionality of the photosynthetic machinery. Moreover, the enzymatic activities and the metabolomic approach revealed that SL activated both enzymatic and non-enzymatic antioxidant systems to deal with salt stress.



## Use of biostimulants to increase biomass production in Lavandin (*Lavandula × intermedia*) cultivated in Tuscan-Emilian Apennines

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Lavandin *Lavandula × intermedia* is one of the most important perennial aromatic shrub widespread in the Italian marginal areas and is mainly cultivated for essential oils production. Nowadays, in rural areas, biostimulants are gaining interest as a useful product to enhance biomass production, following the concept of sustainable agriculture. The present work aimed to evaluate the effects on biomass production of two biostimulants on lavandin crop cv "Grosso." The biostimulants evaluated were: FITOSIM® composed from hydrolysed animal epithelium and FITOSIM ALGA® formulated from brown marine algae. The trial was conducted in three farms located in the Tuscan-Emilian Apennine area (Northern Italy). The products were applied as foliar spray fertilization during the blooming phase (weekly, in two times) of the years 2020 and 2021. The crops were transplanted in three different years: at Preci farm in 2016, at Campazzo farm in 2013 and at Pedroni farm in 2009. In 2020, results showed that FITOSIM ALGA® increased the fresh weight of the inflorescence, moreover FITOSIM® and FITOSIM ALGA® treatments enhanced the stem and total fresh weights compared to the control. In 2021, treatments enhanced the inflorescence and total fresh weights, and the dry weight of the inflorescence, while FITOSIM ALGA improved only the total plant dry weight. Furthermore, the results showed that the year of transplant influenced the agronomic performances of the investigated biostimulants. In conclusion, this work suggests the ability of the biostimulants to enhance biomass production on lavandin crop. However, further investigation will be required to evaluate the most efficient number of treatments, the effect of the biostimulants on yield parameters and on the quality of the essential oils.

## Phenotyping of *Amaranthus hybridus* biotypes carrying two different allelic variants of acetolactate synthase (ALS)

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PhenAMAre project aims to provide an insight into the evolution of resistant biotypes under herbicide selection pressure. *A. hybridus* is a weed that infests summer crops and is diffused worldwide. Inhibitors of acetolactate synthase (ALS) are the herbicides most used for its control, especially in soybean. The repeated use of ALS inhibitors is causing the evolution of herbicide-resistant biotypes that are becoming a threat to sustainable agriculture. In this project, two biotypes of *A. hybridus* cross-resistant to ALS inhibitors, one including a point mutation W-574-L (TGG→TTG) and one including a double point mutation W-574-M (TGG→ATG) in the ALS gene, will be used for a comparative growth experiment using the ALSIA Plant Phenomics platform in Metaponto (<https://www.alsia.it>) in the spring 2022. Two heterozygous plants (ML) were individually reproduced to obtain the homozygous progeny MM and LL to be used for experiments. The activity of ALS enzyme extracted from homozygous MM, LL and WW was compared with an *in-vitro* activity assay to check if different allelic variants have a different level of resistance to ALS herbicides. Microsatellite analyses are in progress to estimate the genetic diversity among mother plants and the homozygous progenies. Three genotypes will be included in a phenomics analysis (WW, MM and LL), both untreated and ALS-inhibitors-treated. Plant growth will be monitored until flowering, evaluating several parameters (leaf area, stem diameter, plant height/width, growth rate, biomass, leaf tracking over time) and finally panicles will be collected, and seed production determined.

Eventual phenotype differences between the two biotypes might be useful for weed management, and to understand why an allelic variant can fix in a population even if it needs a double mutation event. Better understanding the effect of different allelic variants might also help to predict the effect of the same variants in other weed species.

## A LAMP (loop-mediated isothermal amplification) assay to detect the 574-Leu mutation endowing acetolactate synthase (ALS) resistance in three weedy amaranth species

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The UN Agenda 2030 and the European Green Deal through the “Farm to Fork” have the goal of promoting food security and sustainable agriculture. In this context, the reduction of pesticide use should be a priority. Currently, the chemical control remains the most efficient method to control weeds, but the repeated use of herbicides with the same site of action has led to the evolution of resistant weed biotypes. This is a threat for crop production sustainability, as farmers have to repeat the herbicide treatment or use chemicals with a worse ecotoxicological profile. Therefore, the on-site early detection of resistance might help stakeholders to optimize the use of agrochemicals. *Amaranthus* spp. biotypes resistant to acetolactate synthase (ALS) inhibitors, with the amino acid substitution triptofan-574-leucine in the ALS gene, have been reported in Italian soybean fields since 2007. We developed a loop-mediated isothermal amplification (LAMP) assay to rapidly detect plants of *A. retroflexus*, *A. hybridus* and *A. tuberculatus* carrying that specific point mutation. Candidate primer sequences were compared with a multi-species alignment of ALS: the most conserved primers among species were chosen. In the set-up step, a cleaved amplified polymorphic sequences (CAPS) assay was used to genotype susceptible and resistant accessions of each species. Homozygous (e.g., tryptophan-tryptophan and leucine-leucine) plants were used to test how the LAMP assay would perform. For the validation, the LAMP assay was used to predict the genotype of two accessions per each species, one phenotypically susceptible and one resistant. Then the same samples were genotyped with the same CAPS assay used for the set-up in a blind experiment and results were compared to estimate sensitivity, specificity and precision. This LAMP assay correctly detected the 574 mutants in the three *Amaranthus* species investigated, with reproducible results in lab conditions. The on-site feasibility is currently under investigation.

## **Plant-derived biostimulants differentially mitigate salinity stress in lettuce and tomato plants**

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Plants have evolved diverse tolerance mechanisms to cope with salt stress that are plant-species dependent. However, these adaptive strategies often prove to be inefficient in mitigating the rapidly increasing salinity. In this respect, plant-based biostimulants have gained increasing popularity because of their ability to alleviate the deleterious effects of salinity. Hence, this study aimed at evaluating the sensitivity of tomato and lettuce plants grown under high salinity and the possible protective effects of four protein hydrolysate (PH)-based biostimulants. Plants seeds were sown in pots filled with sand and sieved peat 1:1 (w/w) and grown under controlled conditions. Plants were set in a 2 × 5 factorial experimental design completely randomized with two salt conditions, Low salt (0 mM) and High salt (120 mM for tomato or 80 mM for lettuce), and four biostimulant treatments plus a Control with distilled water (C, D, H, P and Control). Salinity was imposed twice a week, five applications in total, whilst biostimulants were applied through foliar spraying once a week with a total of three foliar applications throughout the experiment. At 28 days after germination the plants were harvested. Our results showed that either salinity or biostimulant treatments affected the biomass accumulation of both plant species at different extents. A higher production of antioxidant enzymes (*i.e.*, CAT, APX, GPX and SOD) and proline was observed under salinity, differentially produced according to the biostimulant applied. Nonetheless, an overproduction of proline was observed in salt-stressed lettuce plants compared to tomato plants. Overall, our results suggest that tomato were constitutively more tolerant to salinity than lettuce plants. However, the effectiveness of biostimulants in alleviating high salt concentrations was more evident in lettuce. Among the four biostimulants tested, P and H showed to be the most promising for the amelioration of salt stress in both the plant species.

## The roles of brassinosteroids in root architecture and adaptation in *Arabidopsis thaliana* in limiting phosphate conditions

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Brassinosteroids (BRs) are steroid hormones that are essential for plant growth and development, playing an important role especially regarding cell division, elongation, and differentiation. In addition, BRs are also implicated in root hair induction and plant response to nutrient deficiency in soil. One of the most crucial factors is the process of BR transport and signalling in cells. It then results in a cascade of signals through the activity of several kinases that finally alter the activity of transcription factors, thus leading to altered gene expression. Key aspect of this study is the activity of BRs at the root level, with a resulting effect on root system architecture changes and on nutrient deficiency adaptation strategies, in particular regarding their cross-talk with other phytohormones like auxins. Moreover, BRs signalling has recently been shown to be involved in phosphorus and iron acquisition in studies using *Arabidopsis thaliana* as a model plant. Phosphate, a macronutrient found in large quantities in soil, is often not sufficiently bioavailable to ensure optimal plant growth, thus becoming another limiting factor for an optimal root system growth. In this respect, this study focuses on how these stimuli are perceived, and transmitted in the different root tissues, in the presence of altered phosphate availability and in different selected *Arabidopsis* genotypes, and how they affect root architecture and morphology. The development of a more extensive knowledge of the complex interplay between BRs signalling, cellular redox status and the surrounding microenvironment will certainly be beneficial for understanding the mechanisms of plant survival strategies and growth adaptation in non-optimal growth conditions, particularly as a valuable solution to increase crop yield and quality by minimizing the impacts on the environment.

## **Somatic embryogenesis is a sustainable tool for inducing biotic stress resistance in grapevine**

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Grapevine is one of the most economically important fruit crops worldwide. Several pathogens are known to affect the yield and quality of grape production, resulting in extensive use of plant protection products (PPPs). In the perspective of reducing the use of PPPs to shift toward a more sustainable agricultural system, the development of alternative protection strategies is highly desirable and it can be achieved by using New Plant Engineering Techniques (NPETs). These tools can be used in genetic improvement purposes such as inducing plant resistance to both biotic and abiotic stresses. The aim of this work is to use somatic embryogenesis as a method for grapevine genetic improvement, by exploiting its potential of inducing somaclonal variations in regenerated plantlets. Immature inflorescences of the red-grape variety 'Brachetto' were cultivated on dedicated substrates to stimulate somatic embryogenesis; the embryogenic calli were maintained in culture for several months, then the medium was supplemented with the resistance inducer Acibenzolar-S-methyl (commercial product BION®), widely used in agriculture against several pathogens, in order to prime defense responses in *in vitro* explants. Fifty-five independent somaclone lines were obtained, therefore a preliminary *in vitro* selection was performed by evaluating the expression levels of defence genes (e.g. pathogenesis-related proteins and stilbene synthases) commonly overexpressed in response to biotic stress. In 10 out of the 55 somaclones, several tested genes were upregulated in comparison with the mother plant, suggesting a constitutive activation of general responses to biotic stress. Further analyses will be conducted *in vitro* and *in vivo* on the selected somaclones to verify their ability to resist against fungal infections. The end goal would be to obtain grapevine plants with a higher level of tolerance/resistance to biotic stress without the use of techniques subjected to the restriction of European regulations for GMOs, such as transgenesis, genome editing or cisgenesis.

## Rearing mealworm larvae on substrates contaminated by foodborn pathogens

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The interest in edible insects rearing as a new valuable protein source of food and feed is significantly increasing. Despite the microbial safety of insect rearing is very important, very little research had been done on the effects of foodborn pathogens on their growth performance and welfare. In the present study mealworm (*Tenebrio molitor*) larvae were reared on organic wheat middling, spiked with *Escherichia coli* and *Staphylococcus aureus* at three contamination levels (1, 5, and 7 Log colony forming units per gram). For each contamination level and control (not contaminated wheat middling), six replicates of 100 third instar larvae each were placed in boxes and supplied with 0.04 g/larva/week of feed, until the occurrence of 50% pupation. Boxes were kept at  $28 \pm 1$  °C,  $60 \pm 5\%$  RH, and 24 h dark photoperiod. Last instar larvae and pupae collected were counted and individually weighted. No significant differences on the survival rate (ranged from 82 to 97%), development time (ranged from 73 to 81 days), last instar larval (ranged from 166 to 175 mg) and pupal weight (ranged from 144 to 149 mg) were observed among the different treatments. The results suggested that the contamination of feeding substrates with *E. coli* and *S. aureus* didn't affect the growth performance and the welfare of *T. molitor* larvae. Furthermore, other studies focusing on the microbial dynamics, in these rearing trials, showed that *E. coli* and *S. aureus* growth was remarkably inhibited in the larvae feeding substrates and in the larval gut. In conclusion, in this insect rearing system, the presence of *E. coli* and *S. aureus* seems to not represent a threat neither for the mealworm larvae nor for the safety of the insect-based foods.

## Characterisation of *Fusarium musae*, agent of crown rot disease of banana fruits

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Banana is a major export product and one of the most important staple crops after wheat, maize and rice. In banana production, important losses occur every year in terms of food and economic losses, especially in organic bananas where losses can reach up to 80% in some countries. One of the main causes of such a big loss is post-harvest diseases of which crown rot is the most devastating for banana fruits. Knowing the causative agents of crown rot disease it's fundamental to reduce its impact in the production of banana fruits. *Fusarium musae* is a novel species belonging to the *Fusarium fujikuroi* species complex isolated for the first time in 2011 and identified as one of the causative agents of crown rot disease in banana fruit that is its only known plant host. In order to study the threat posed by this novel species we characterised phenotypically and epidemiologically a collection of 12 strains of *F. musae* with different geographic origins. Infection assay on banana fruits showed that *F. musae* is more aggressive than its "sister species" *F. verticillioides* with which it has been misidentified for a long time. In addition, *F. musae* resulted less sensitive to widely used fungicides compared to *F. verticillioides* causing suspicion of some kind of intrinsic or induced resistance. Further studies will focus on genomics characterisation of this novel species in order to find the genes responsible for the infection of the fruit.



## Bioformulates as a promising and sustainable tool against wheat soil-borne pathogens

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Durum wheat (*Triticum durum* L.), one of the most relevant sources of food worldwide, is susceptible to the attack of fungal pathogens responsible for different severe diseases affecting the basal portion and the roots. Specifically, *Fusarium* Crown Rot (FCR) and *Fusarium* Foot Rot (FFR) are diffuse all over the world and the main pathogens are several *Fusarium* and *Microdochium* species that can act singly or in combination, affecting the roots, the collar and the first internodal portions of the culm. Both diseases are primarily linked with the presence of high concentrations of inoculum in the soil. Nowadays the main defense strategies against these diseases are based on agronomic practices aimed at reducing the predisposing causes and fungicide-based seed coating application. New strategies of defense involve the use of seed coating with beneficial microorganisms, a covering seed technique for precision agriculture able to improve seed performance and reduce production costs. The use of natural products can represent a valid alternative strategy to defend crops in the near future. There is a growing interest in the use of biocontrol methods that are based on the use of natural competitors aimed at suppressing the growth of pathogens. The organic seed coating technique with endophytic bacteria or Essential Oils (EOs) may represent a reliable and safe microbial-based strategy able to control soil-borne pathogens. To select potential biocontrol agents against FCR/FFR diseases, *in vitro* antagonistic effect of six different EOs (rosemary, thymus, lemon, basil, mint, carnation) and a total of 29 strains belonging to the genus *Lactobacillus*, *Pseudomonas*, *Bacillus*, *Stenotrophomonas* and *Paenibacillus* were tested against *Microdochium* and *Fusarium* assessing their fungistatic and fungicidal effects. Furthermore, their efficacy as seed coating using polyvinyl alcohol was tested. Further investigation on the three-way interaction plant-soil-microbe will help us to understand how to improve a seed coating strategy in wheat.

## Unraveling the capacity of the epiphytic bacterium *Stenotrophomonas rhizophila* Ep2.2 to protect tomato leaves against *Botrytis cinerea* infection

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Biological control agents (BCAs) such as plant growth-promoting rhizobacteria (PGPR) represent one of the most attractive eco-friendly solutions for sustainable plant protection. In the attempt to find novel BCAs able to protect tomato plants against the pathogens *Botrytis cinerea* and *Pseudomonas syringae* pv. *tomato*, we assessed the protection potential of four PGPR strains: *Pseudomonas fluorescens* M26, *Pseudomonas chlororaphis* M71, *Bacillus amyloliquefaciens* M123, and *Stenotrophomonas rhizophila* Ep2.2. The ability of these strains to inhibit *B. cinerea* and *P. syringae* pv. *tomato* was tested in vitro by dual culture assays. Two-compartment petri dishes were also used to reveal the inhibitory role of microbial volatile organic compounds (MVOCs). Our results showed that the four strains, although to different degree, were able to hamper pathogen growth through emission of MVOCs, with *P. chlororaphis* M71 and *S. rhizophila* Ep 2.2 being the most effective ones. However, these two strains did not show marked inhibitory effect in dual culture assays, especially against *P. syringae* pv. *tomato*. Leaves of tomato cv. Micro-Tom were used to evaluate the effectiveness *in planta* of the four PGPR strains when sprayed 48 h before *B. cinerea* inoculation. *S. rhizophila* Ep 2.2 strongly inhibited the pathogen and was then selected as the most promising BCA candidate. *S. rhizophila* Ep 2.2 had been isolated as epiphyte from an ornamental plant and identified by 16S rDNA sequencing. Although it is known that *S. rhizophila* is a plant associated bacterium, its plant protection potential has been overlooked so far. Proton Reaction - Mass Spectrometry (PTR-MS) coupled with Gas Chromatography - Mass Spectrometry (GC-MS) analyses are ongoing to characterize the blend of MVOCs emitted by *S. rhizophila* Ep2.2. Gene expression analyses on treated leaves will help to clarify whether *S. rhizophila* Ep 2.2 can stimulate the immune system to enhance natural defense mechanisms of tomato plants.

## Root and Collar Rot caused by *Phytophthora nicotianae* and *Phytophthora palmivora*, a new disease on Paulownia in Europe

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The genus *Paulownia*, characterized by fast growth, ability to re-sprout rapidly after cutting as well as a good tolerance to drought and high soil acidity, are trees native to China. *Paulownia* species are grown in managed plantings in several European countries for the production of wood and biomasses. The species of greatest commercial importance worldwide are *P. tomentosa*, *P. elongata*, *P. fortunei*, *P. catalpifolia*, and *P. kawakamii*. *Paulownia elongata* and *P. fortunei*. They are more thermophilic, suitable for biomass and wood production in warmer areas of southern Europe, such as Italy and Spain. In 2018, a severe disease characterized by wilting, stunting, leaf yellowing, and collapse of the entire tree as a consequence of root and crown rot, was observed in a commercial paulownia planting production. Around 40% of trees of a 2-year-old planting of *P. elongata* × *P. fortunei* in Calabria (Southern Italy). *Phytophthora* species were consistently recovered from basal stem bark, roots and rhizosphere soil of symptomatic trees and were identified as *Phytophthora nicotianae* and *P. palmivora*. The identification was performed on the basis of both morphological characteristics and phylogenetic analysis of rDNA ITS sequences. To confirm Koch's postulates, potted paulownia saplings were transplanted into infested soil and stem-inoculated by wounding. Both *Phytophthora* species were pathogenic and caused root rot and stem cankers. Even though *P. palmivora* was the only species recovered from roots of naturally infected plants, in pathogenicity tests through infested soil *P. nicotianae* was more virulent. This is the first report of *Phytophthora* root and crown rot of a Paulownia species in Europe. The use of healthy nursery plants is crucial for preventing this disease in commercial plantations. Strategies to prevent this emerging disease include the use of healthy nursery plants, choice of well-drained soils for new plantations, and proper irrigation management.

## Eco-sustainable approaches to understand and contain the kiwifruit decline syndrome in Italy

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During the last decades the Italian kiwifruit production has been threatened by the kiwifruit bacterial canker disease caused by *Pseudomonas syringae* pv. *actinidiae* (Psa). Nonetheless, since 2012 the kiwifruit orchards and their production are further decreased due to the emergency of a new complex disease known as Kiwifruit Vine Decline Syndrome (KVDS). The syndrome was reported for the first time in Verona province and since then it progressively spread in most of the Italian areas in which kiwifruit is cultivated. For example, in the Latium region, between 2017 and 2021 due to KVDS, 50,000 tons of kiwifruit have been lost among a cultivated area of 3.000 ha. The symptoms appear during summer (within June and July), by an evident reduction of the plant development, followed by a whole wilt of plant canopy. Starting from the browning and damaged root system, xylem necrosis, progressive loss of capillitium, hypertrophy, and separation of the cortical layer, the symptoms systemically quickly spread inducing the collapse and death of plants within the same season. Even though several studies have been carried out, different aspects related to the causes of KVDS are still unknowns. Since 2020, different studies have been planned in two experimental kiwifruit areas of Lazio region, where KVDS was recorded. A field trial is currently carrying out in two experimental kiwifruit orchards, different for management (organic and intensive) and soil characteristics. Eco-sustainable innovative treatments are being tested to evaluate their effect on the rhizosphere, vascular and epiphytic microbial communities and on the onset of KVDS symptoms. At present, the results showing a progressive changing of microbial communities. Moreover, further studies based on high throughput sequencing techniques and metagenomics have been planned to provide a more detailed qualitative and quantitative analysis of the microbial communities involved in the KVDS.

## Molecular assessment of the feeding behavior of the Brown marmorated stink bug *Halyomorpha halys*

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The Brown marmorated stink bug *Halyomorpha halys* is one of the most important invasive pest species in the world. It is native to north-eastern Asia and spread to many countries worldwide. In 2004 *H. halys* reached Europe, where it invaded most countries. By feeding on a variety of different fruits and vegetables, this species is causing high economic damage in the invaded areas. Although more than 300 host plants are known today, the exact feeding behavior of the Brown marmorated stink bug in nature still needs further investigations. This information could be applied to control this invasive species before the individuals start to enter the orchards and can be helpful to prevent the damage and yield loss caused by *H. halys*. The aim of this research project is to analyze the gut content of the brown marmorated stink bug. After extracting the DNA of the gut content of individuals collected through the season in different locations in South Tyrol, a DNA amplification will be performed with plant-specific primers. In this way, we will investigate the feeding behavior of this polyphagous pest species over the year. Our results will contribute to a better understanding of the behavior of *H. halys* in nature, will help to improve the control of this important pest species and can therefore lower the damage caused.

## Plant-derived foliar biostimulants vs. root-colonizing plant growth- promoting rhizobacteria: ameliorative effects salt-stressed tomato plants

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During the last years, one of the main concerns in agriculture and food production is related to the excessive salt concentrations in soils and irrigation water, which may affect plant growth and crop productivity. In order to mitigate these harmful conditions, plant biostimulants (PBs) may represent a powerful and sustainable tool. In this context, the present work was aimed at identifying the effectiveness of plant-derived and bacterial biostimulants in enhancing the growth and tolerance of tomato plants to salinity stress. Tomato seedlings were hydroponically grown in high (120 mM NaCl) and low (0 mM NaCl) salinity and they were either treated with 4 plant-derived protein hydrolysates biostimulants (*i.e.*, C, D, H, P) at foliar level or inoculated with the PGPR *Azospirillum brasilense*. The morphological and biochemical effects were assessed by analysing the biomass accumulation and root characteristics (*i.e.*, length, volume, surface area), enzymatic antioxidant activities (catalase, ascorbate peroxidase, guaiacol peroxidase, superoxide dismutase) and osmotic stress protection (proline). Furthermore, modifications in the metabolomic profiles of both leaved and root exudates were also investigated by UHPLC/QTOF-MS. Our results showed that both salinity and biostimulant treatments affected all the evaluated parameters. For instance, biomass accumulation decreased under high salinity, yet the microbial-based biostimulant treatment considerably improved root biomass and morphological traits, both in low and high salinity conditions. As expected, the antioxidant enzymes and proline production were enhanced in salinity stress at different level according to the biostimulant applied. The UHPLC/QTOF-MS approach allowed the identification of signature metabolomic patterns for the leaves extracts and root exudates, with inflected accumulation of compounds imposed by either biostimulant treatments or saline stress. Overall, results showed that *A. brasilense* and the biostimulant P were the most effective treatments to mitigate the deleterious effects induced by high salinity.

## **Gradual crop replacement strongly shapes soil fauna and microbial communities in long-lasting tree crops**

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This study investigates the long-term effect of crop conversion from grapevine to apple orchard on chemical and biological (biodiversity) soil quality parameters. In South Tyrol (Northern Italy), we selected an agricultural field characterized by three plots (within less than 1 ha) where grapevine was replaced by apple cultivation 100, 50, and 4 years ago, respectively indicated as A-1922, A-1970, and A-2016. This unique setting allows us to investigate the effect of gradual orchard replacement, avoiding other confounding environmental factors. We collected a total of 45 soil samples (15 replicates at each site). We assessed chemical-physical soil characteristics (e.g. pH, soil texture, soil organic matter, total nitrogen, total heavy metal content, and available mineral nutrient content). We investigated the soil biodiversity via DNA metabarcoding of the bacterial 16S rRNA gene, fungal ITS2 region and mesofauna COI gene. Overall, soil chemical composition could discriminate the plots and predict which history of conversion the samples went through (Anova on PCA components  $p < 0.01$ , LDA with 88.89% classification accuracy). These patterns were strongly reflected in the ecological organization of soil microbial and fauna communities. The alpha-diversity of the investigated communities showed significant changes across the sites (Kruskal Wallis  $p < 0.05$ ). Beta-diversity showed a strong effect of the conversion time on the bacterial, fungal and soil fauna communities' composition (PERMANOVA  $p$ -value  $< 0.05$  for all kingdoms). Co-occurrence ecological networks highlighted that the number and taxonomic diversity of bacterial keystone species increased according to the duration of apple cultivation and their interaction with the fungal communities. Conversely, the number of soil fauna species interacting with bacteria and fungi decreased linearly according to the oldest apple orchard. These results suggest to consistently include soil biodiversity among soil quality indicators, particularly in perennial tree crops, where the agricultural history of the land exerts a strong influence on soil biodiversity.

## HoloHerbarium: herbaria preserve plant microbiota responses

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The plant microbiota, composed by all microorganisms associated with plants, includes many organisms in most cases belonging to the kingdoms of viruses, bacteria and fungi. Research projects usually focus on the study of a host and a single symbiotic or pathogenic organism, but it is now clear that the health and fitness of plants is mainly influenced by all those interactions that are triggered within its microbiota. Recent research has also highlighted how agricultural practices can have a strong impact on the communities of microorganisms associated with plants, not only from a purely taxonomic but also functional point of view. The Holoherbarium research project, conducted at the Istituto per la Protezione Sostenibile delle Piante of the CNR of Turin in collaboration with the University of Turin and Muséum National d'Histoire Naturelle of Paris, focuses on the study of "ancient" plant microbiomes. In fact, the roots of herbarium plants contain molecular traces of their "ancient" microbiomes, in the form of DNA and proteins. Therefore molecular study of plants collected "before", and "during" the exponential "acceleration" of global changes should inform us on how and to what extent these changes have durably affected the plant microbiota, widely recognized as an essential component of plant health. The information gathered on microbial communities associated with past plants could be used in agro-ecology to restore potentially polluted agricultural land and to improve the agronomic characteristics of cultivated plants. Currently the research, focused on various samples of ancient roots, taken from Italian and French botanical collections, has highlighted the presence of arbuscular mycorrhizal fungi (AMF) in the cortical parenchyma of roots of stained herbarium samples of Segale cereale [Bianciotto et, 2022: "*Herbaria preserve plant microbiota responses to environmental changes*" Trends in Plant Sciences]. Ancient DNA extractions and NGS sequencing were performed on these samples. Preliminary analyzes confirmed the presence of symbiont fungal sequences (AMF). Meta-barcoding analyzes, currently underway, will allow a further study of the taxa composition of the various selected ancient plant microbiota.



## Characterization and valorization of the biodiversity of a collection of *Metschnikowia pulcherrima*-related strains for agri-food applications

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The updated EU rules on organic agri-food productions recommend the input reduction and preventive biological processes management. *Metschnikowia pulcherrima*-related strains represent an option as biocontrol agents (e.g. *M. fructicola* NRRL Y-30752, patent n. EP2269455A1). They are ubiquitous, aroma-producing yeasts, with antimicrobial activity against plant pathogens, mainly due to the pulcherrimin production, a brown-red iron-chelating pigment. Therefore, this study aimed to investigate the genotypic and phenotypic biodiversity of a collection of 108 strains from different origins, to select some of them for biotechnological applications. Several molecular fingerprinting techniques were used, starting from the type strains to identify the most discriminating tool, which was then applied to the whole collection. The phenotypic traits were evaluated by testing different enzymatic activities and the pulcherrimin production. Finally, the antimicrobial activity of selected strains was tested *in vitro* against undesirable microorganisms. A great biodiversity was acknowledged, especially regarding the genotyping and enzymatic activities. It was observed a strong inhibition towards *Botrytis cinerea*, a variable antagonism towards *Oenococcus oeni* and *Pediococcus parvulus* and none effect towards some yeasts. Further, since the bioprospecting of these yeasts is hampered by the lack of genome sequences available, the genomes of four type strains were sequenced, in collaboration with ISPA-CNR of Bari and Foggia (Ferrara, Capozzi, Mulè *et al.*, unpublished data), and *de-novo* assembled. A comparative genomic analysis carried out on the new genomes and those of the other three type strains in NCBI database showed that all sequences harbored variants of pulcherrimin-encoding genes. Furthermore, analysis of CAZymes unraveled up to 13,513 matches, including chitinases possibly involved in pathogen fungal cell wall degradation. The unexplored gene pool represents a substrate for further investigations to associate genotypic variants with phenotypes and these data suggest that the pulcherrimin-producing strains of *Metschnikowia* are suitable as biocontrol agents in agriculture.

## **Metagenomics unravel the microbial heritage of Italian typical fermented foods**

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In modern times, many traditional fermentation technologies have been industrialized, relying on the use of selected microbial starters to standardize the process and the fermented food properties. However, this leads to the global diffusion of few microbial strains and to a huge decrease of the microbial diversity in fermented foods. The project FOODMICROHERITAGE has the goal to characterize the microbiome of typical Italian fermented foods by using shotgun metagenome sequencing and to create a collection of microbial genomes of lactic acid bacteria (LAB) species and strains from artisanal fermented foods, to preserve and depict the microbial diversity of these traditional products. More than 200 Italian fermented foods were collected from different producers, including fresh, medium- and long-ripened cheeses and fermented meats. Metagenomics sequencing and Volatile Organic Compounds (VOCs) analysis were carried out. We identified microbial genes involved in the production of the typical sensorial properties of each fermented food, highlighting the presence of different LAB strains in each product. Microbial profiles of each food type are unique and can be used to track fermented foods origin and production according to traditional practices. Acknowledgements: This work was funded by a grant from the Italian Ministry of Foreign Affairs and International Cooperation to the project FOODMICROHERITAGE–Quality and authenticity protection of artisanal fermented foods through the characterization and conservation of their microbial and genetic heritage, CUP E79J21002000001.

## **Lactic Acid Bacteria resident in cheesemaking facilities drive the technological properties and impact on the cheese quality**

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The cheesemaking technology mainly relies on lactic fermentations by Lactic Acid Bacteria (LAB), that also contribute to shape the cheese sensorial profile during ripening. However, the importance of LAB goes beyond the technological aspect, since some metabolites produced by several LAB strains might also exert a positive effect on the health. In addition, LAB might inhibit foodborne pathogens or alternative microorganisms by competing for nutrients or by producing antimicrobial metabolites. We collected 94 environmental swabs from 16 cheese-making facilities in Southern Italy, besides ingredients, raw materials, and final products. Shotgun metagenome sequencing was carried out. Through the reconstruction and characterization of LAB Metagenome-Assembled Genomes, we observed a wide range of potential activities exerted by these microorganisms, supporting the idea that they might act as biopreservatives when developed in biofilms on industry surfaces. In addition, we observed that LAB strains from the production environment are specific to the cheese type, thus highlighting their potential role for the development of a distinctive sensorial profile. Acknowledgements: This work was supported by the project MASTER-Microbiome Applications for Sustainable food systems through Technologies and Enterprise, receiving funding from the European Union's Horizon 2020 research and innovation programme (GA 818368).

## Unculturable taxa of insects pests microbiomes: a focus on genera *Erwinia*, *Pantoea* and *Blattabacterium*

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*Candidatus* *Erwinia* *dacicola*, *Candidatus* *Pantoea* *carbekii* and *Blattabacterium* *cuenoti* are co-evolved symbiotic bacteria associated with *Bactrocera oleae* (olive fruit fly), *Halyomorpha halys* (brown marmorated stink bug) and *Periplaneta americana* (American cockroach), respectively. These bacterial endosymbionts are as-yet uncultivated, probably due to the adaptation to their hosts which has led to genome reduction; the uncultivability also determines the difficulty of describing these species in sufficient detail, and therefore they are referred to as *Candidatus* taxa. According to the List of Prokaryotic names with Standing in Nomenclature (LPSN), *Erwinia* includes 20 cultivable species with a validly published name, but 10 others are reported to have an invalid name, and *Ca. E. dacicola* is the only uncultured taxon. As for *Pantoea*, 18 species have a validly published name, but other 13 species with an invalid name; moreover 3 *Candidatus* species are reported, i.e., *Ca. P. carbekii*, *Ca. P. edessiphila* and *Ca. P. persica*, present in stink bugs of the family Pentatomidae. Regarding *Blattabacterium*, *B. cuenoti* is a valid species name, despite unavailability of a pure culture, and three other species have an invalid name and are uncultivated. In this complex framework, the aim of the present study was to clearly define the taxonomic status and the phylogenetic relationships among cultivable and as-yet uncultured species of *Erwinia*, *Pantoea*, and *Blattabacterium*, including also the species with an invalid name. This will provide: i) a robust basis for comparative genomic analyses, aimed at understanding the genomic background related to the free-living state and ii) the possibility to design reliable specific primers and/or probes to detect the uncultivated bacteria. It is also suggested that the whole genus *Blattabacterium* should be referred to as *Candidatus*, and that the species *B. cuenoti* be indicated as *Ca. B. cuenoti*, to clearly convey the information that it is an uncultivated taxon.

## **Assessment of Biolog Ecoplate™ method for functional metabolic diversity of aerotolerant pig fecal microbiota**

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In the last decades, gut microbiota and its role in mammal host development and health have been increasingly investigated. Metabolites produced by gut microbiota can affect intestinal homeostasis and immune system maturity and activation, and in turn, they can influence the health and growth performance of livestock. Therefore, a better understanding of the functional metabolic capability of the gut microbiota would be appreciated by the scientific community. In this study, the Biolog™ Ecoplates technology was applied for studying the metabolic potential of the aerotolerant microbial community of pig fecal samples, evaluating the interference of different storage conditions and cell concentrations. The length of time for which a fecal sample maintained detectable and unchanged microbial metabolic activity was also investigated. Two assays aimed to evaluate differences in the metabolic activities between fresh and snap-frozen fecal samples at different dilutions and at different lengths of times of preservation at -80 °C were carried out. The biodiversity and the predicted functionality of the entire bacterial community through a targeted metagenomic approach were also explored. The results highlighted that snap freezing of fecal samples preserved the metabolic activity of the microbial community when compared to fresh feces. Sample storage at -80 °C did not significantly affect the metabolic activity of the microbial community, which was stable for 150 days. Furthermore, the highest metabolic activity was detected with 1:2 to 1:5 dilutions of the stock suspension. Biolog™ Ecoplates technology is a rapid and useful method to explore microbial communities' metabolism in animal fecal samples contributing to investigate host animal physiology.

## Into the economics of organic agriculture: a topic modelling review of the scholars' interests

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Organic agriculture is experiencing a continuous growth in the world in terms of a variety of agricultural, environmental and economics indicators, such as utilized agricultural area, number of producers, or value of production as well as the impacts on yields, biodiversity, pollution, or farmer's safety (FiBL and IFOAM – Organics International, 2021). The importance of organic agriculture reflected in an ever-higher attention devoted by the scientific community to this method of production, which has led to the emergence of an abundance of scientific writing. Since the topic of organic agriculture interlaces a great variety of aspects of our society, scientific authors studied organic farming taking multiple perspectives, e.g., production (Kijlstra and Eijck, 2006), consumption (Rana and Paul, 2017), environment (Muñoz-Sánchez and Pérez-Flores, 2021). The contributors to organic farming have been proliferated also because experimental results can be hardly extended to different soil and climatic areas. Focusing on the economics of organic agriculture, the objective of our study is to figure out the major interests developed by scholars in this field and to explore whether and how these interests changed over time. To do this, we conduct an extensive review of the scientific literature dealing with the economics of organic agriculture. Because of the great amount of scientific material produced over the years, we use structural topic modelling (STM) to identify the main topics in the corpus of relevant abstracts retrieved from the Scopus database. STM (Roberts et al., 2014) is an unsupervised Bayesian generative topic model that allows not only to identify relevant topics from a corpus of text, but also to model the prevalence of these topics according to specific covariates, such as the year of publication, the geographical area where a study is conducted, the agricultural sector that is the object of the study, or the outlet of publication of the study. Exploiting these characteristics, we are able to investigate the trajectories of the economics scholars' interests in the organic sector or their spatial/product heterogeneity. This would allow to connect the trends in scientific economics interests to the wider organic debate (e.g., policies, production trends, issues), in addition to possibly identify areas that are still less explored.

## **Floristic-vegetational sciences and landscape ecology contributions for the accounting of agrobiodiversity and farmland ecological functionality: assessment of a certification scheme**

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The current widespread degradation of the environmental matrices underlying the agricultural systems undermines their capacity to sustain the ecological processes across rural land. Applied ecological sciences, like landscape ecology and phytosociology, concur to meet the need of scientific applied knowledge building related to the comprehension of the ecological patterns and trends underlying the farmland agroecological design and management. In this context, the herepresented project aims at validating a model for the accounting of the contribution that agroecological farmland management can bring to agrobiodiversity values and, as a whole, to the recovery of the ecological functionality of farmland. This work is a direct contribution to the development of the already existing Biodiversitas label (a certification scheme for farms developed by Polyculturae Association, in collaboration with University of Milan and Marche Polytechnic University), through its test, calibration and validation. The assessment model is focused on a result-based evaluation of a diversified set of practices. Analysis are being led at different spatial and time scales, on paired agroecology and conventional farms. Floristic, vegetational and landscape ecology data are being collected and processed and then matched with other environmental variables (soil, faunal and agronomic management data). A set of environmental indicators is being evaluated, aiming at identifying their fitness for project purposes. Reference indicators, their target values and significant thresholds, target species (bioindicators) and the indicators mutual interaction patterns will be the final output of this comparative assessment, aiming at their integration within the technical standards of the certification system. The project also envisages a subsequent qualitative and quantitative assessment of the ecosystem services interlinked with the studied ecosystem functions, aiming at highlighting the existing trade-offs and synergies between the evaluated practices. The first results of the preliminary analyses will be here presented.

## Microalgal biostimulant on lettuce: a possible solution to overcome drought stress

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According to the United Nations Food and Agriculture Organization (FAO), the world's population will reach 9.7 billion people by 2050, meaning that there will be an increase in food demand. Hence farmers are called to produce more food on the same actual available arable land, preserving the agricultural sustainability and at the same time working under uncertain scenarios affected by climate change. Baby leaf lettuce, *Lactuca sativa* L., is a species of high agronomic interest, in fact, has a marked importance in the sector of fresh-cut leafy vegetables. In Italy the areas destined for this crop are mainly concentrated in Lombardy, Veneto, and Campania; however, these areas are constantly expanding. Baby leaf lettuce is grown almost exclusively under protected environment (greenhouse) and therefore its irrigation water requirement is high. To reduce the irrigation water demand and at the same time to find agronomic solutions to overcome drought stress, biostimulant might be investigated in the sustainable and eco-friendly production of the baby leaf lettuce. In the present work, treatments with marine microalgae (*Isochrysis galbana*, *Nannochloropsis gaditana*, *Porphyridium* sp., *Spirulina* and *Tetraselmis suecica*) were carried out as a foliar spray application on baby leaf lettuce cv. "Oxana". The production of the crop was carried out at the University of Salerno, under greenhouse, and the crop was subjected to a strong drought stress during the last 20 days of the crop cycle. The results of the experiment showed that *Nannochloropsis gaditana* had an interesting biostimulant effect on the plant, improving different agronomic parameters such as leaf and total dry weights, while from a physiological point of view, the same treatment (*Nannochloropsis gaditana*) led to a higher crop water productivity compared to untreated plant. In addition, all investigated microalgae reported lower stomatic conductance values than the untreated plant, allowing a reduction of water consumption.



## Polyphenolic and terpenic composition of different lavender species and rosemary varieties: contribution to the knowledge of the officinal Mediterranean flora

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Rosemary (*Rosmarinus officinalis*) and lavender (*Lavandula* spp.) are well known Mediterranean plant species intensively used as officinal and spices mainly due to their large amounts of polyphenols and terpenes. Their phytochemical composition might change between species and/or varieties. Therefore, is important to select the most promising varieties to obtain bioactive compounds-enriched extracts for industrial applications. This study aimed at characterizing and compare the polyphenolic and terpenic composition in leaf extracts of different *Rosmarinus officinalis* varieties ('Alba', 'Arp', 'Ginger', 'Gorizia', 'Tuscan Blue' and 'Roseus') and species of *Lavandula* L. (*L. angustifolia*, *L. latifolia*, *L. intermedia* 'Alba', *L. stoechas* and *L. stoechas* 'Alba'). Leaves of plants cultivated in GEA park (Pistoia, Italy) were collected at the balsamic time (February and May/2021) and used to obtain extracts by ultrasound-assisted extraction, further analyzed by HPLC-DAD and GC-MS to identify and quantify polyphenols and terpenes, respectively. Hydroxycinnamic acid derivatives and flavonoids were the most abundant polyphenols in *Lavandula* leaf extracts. Regarding the comparison among species, *L. angustifolia*, *L. latifolia* and *L. intermedia* 'Alba' showed a higher total polyphenolic content (TPC), mainly characterized by caffeic and coumaric acids derivatives. On the other hand, despite lower TPC, *L. stoechas* was especially rich in luteolin and apigenin glucosides. All the varieties of *R. officinalis* had a very similar polyphenolic composition: presence of rosmarinic acid, flavonoids (luteolin, apigenin and quercetin glucosides), caffeic and carnosic acids derivatives, with the highest TPC being found for 'Roseus' and 'Arp', and the lowest for 'Ginger'. The further steps of the study will include the characterization of the terpenes present in these samples and the evaluation of their antioxidant activity. This study will contribute to expand the knowledge about the best varieties of rosemary and species of lavender to produce phytocomplexes that might be utilized for different medicinal, cosmetic and nutraceutical purposes.

## Urban Forests: science-based perspectives for air quality amelioration

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Among the ecosystem services provided by urban forests, the air quality amelioration is particularly relevant: the high level of air pollution in modern cities and the involvement of particulate matter (PM) in the spread of COVID-19 has exacerbated the air quality issue worldwide. For this reason, within the LIFE URBANGREEN project, a research was carried out to evaluate the amount of PM captured by 240 plants belonging to 17 species in two European cities (Rimini, Italy; Krakow, Poland), used as living laboratories. The accumulation of large ( $PM_{10-100}$ ), coarse ( $PM_{2.5-10}$ ) and fine ( $PM_{0.2-2.5}$ ) PM ( $\mu g\ cm^{-2}$ ) was determined with consolidated gravimetric techniques on 2160 leaf samples obtained from the basal, median and apical part of the crown of established trees and shrubs planted in street and in park, with repeated measurements throughout the growing season. On the same samples, the deposition ( $PM_{10}$  and  $PM_{2.5}$  ( $\mu g\ cm^{-2}\ day^{-1}$ )) was evaluated according to a model based on the wash-off rain effect. *Quercus ilex* accumulated more  $PM_x$  than the other species in Rimini, while in Krakow, the highest accumulators were *Pinus nigra* for  $PM_{10-100}$ , *Tilia cordata* for  $PM_{2.5-10}$ , and *Populus nigra* for  $PM_{0.2-2.5}$ . Only in Krakow the capture of some species was affected by the street or park growing condition. In both cities, a greater accumulation of  $PM_{10-100}$  was found in the basal leaves of the canopy. In Rimini, the total PM accumulation tended to increase throughout the year, while in Krakow, the opposite occurred. However, as the accumulation increased, the deposition decreased. The PM accumulation was reduced by rainfall and improved by the air PM concentration, while the wind speed effect was opposite, depending on the city. We believe that these findings can provide insights for the design of greener, healthier and sustainable cities.

## Leaves of *Arbutus unedo* L. as sources of phytocomplexes: does water stress affect the seasonal arbutin and phenolic content?

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*Arbutus unedo* L. is an evergreen species largely widespread in the Mediterranean basin. This plant is considered an underutilized shrub, well adapted to marginal lands subjected to different environmental stresses including drought. Its leaf extracts are an important source of polyphenols to be exploited in different industrial applications. Therefore, this study aimed to optimize the ultrasound-assisted extraction of *A. unedo* leaf extracts to obtain higher yields of polyphenols in a green way. In addition, a field experiment was performed to evaluate the effect of water stress on the seasonal leaf polyphenolic content and composition. To assess the physiological status of the plants, leaf water status, gas exchange, chlorophyll fluorescence were monitored on a seasonal basis. In addition, leaf mass per area and non-destructive measurements of epidermal flavonoids and chlorophyll were measured using a Dualex® device. The extraction conditions to enhance leaf polyphenolic contents in a green way were optimized using a response surface methodology, and higher yields were obtained using lower temperatures and time of extraction. Concerning the field experiment, the water shortage triggered the accumulation of these metabolites in summer till the beginning of the autumn. Therefore, the application of water stress in *A. unedo* seems to be a promising approach to obtain higher polyphenolic yields. Regarding the physiological responses, it is important to highlight that *A. unedo* plants could maintain good performances under water stress conditions, since the different leaf traits analyzed indicated a high photoprotection and drought tolerance. This information can be effectively employed to develop a sustainable agronomic strategy for the cultivation of this species in Mediterranean drought-stressed areas and to obtain polyphenolic rich extracts to be used in the pharmacological and nutraceutical sectors.

# The effects of climate change on grape growing and harvest properties in the wine producing region of South Tyrol, Italy, during 1996-2016

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Climate change is already affecting vine growing conditions and wine production in many regions worldwide. At the same time, consumer preferences and quality targets continue to determine cultivars and vineyard management practices. However, detailed information on how climate change is altering grape production is usually missing. The aim of this study was to quantify the influence of climate change on the viticultural production in South Tyrol, Italy, by relating historical vintage records (harvest date, the yield, and sugar content) to bioclimatic indices (i.e. GDD). For this purpose, a dataset containing vintage records for 8 cultivars (Pinot Noir, Chardonnay, Gewürztraminer, Lagrein, Pinot Gris, Sauvignon, Vernatsch, and Pino Blanc) across 548 different vineyard parcels (135 ha), covering the time period 1996-2016 and an elevational range between 202 m and 764 m a.s.l. was used. Climate information was assessed by calculating bioclimatic indices from interpolated temperature maps over the same period. The Huglin Index, the Winkler Index, the Average Growing Season Temperature (GSTavg), and the Cool Night Index (CNI) have increased by 93 GDD, 172 GDD, 0.9°C, and 2.4°C, respectively from 1996-2016. Harvests have shifted towards earlier dates until 2003, however, from then on, no clear trend could be observed. Moreover, our results indicate that the harvest date for the analysed cultivars usually occurs after a thermal time of about 1600 GDD ( $R^2=0.65$ ) has been reached. Grape sugar levels have experienced a noticeable increase (0.09 °KMW per year;  $R^2=0.67$ ), indicating that adaption strategies via adjusted vineyard management practices, e.g., harvest retardation or yield regulation, may have enabled wineries to better exploit grape ripeness under warmer growing conditions. This study provides first insights on how climate change is impacting important grape growing and harvest properties and thus, may help to develop adaptation strategies aimed at the preservation of high-quality wines and their typicity.

## The evolutionary history of *Phaseolus vulgaris* as revealed by chloroplast and nuclear genomes

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Knowledge about the origin, evolution and expansion of crop species is crucial for their conservation and exploitation. *Phaseolus vulgaris* has a unique evolutionary history, with the wild form originated in Mesoamerica and subsequently introduced into South America, leading to the formation of two South American wild gene pools in North Peru and Ecuador and in South Andes. However, the debate on common bean origin is still open. Indeed, recent studies proposed the so called "Pseudovulgaris" hypothesis on the origin of common bean, that indicates the formation of the North Peru and Ecuador gene pool as occurred much earlier than that of *P. vulgaris* species and, thus of the diversification of Mesoamerican and Andean gene pools. In this case, the North Peru-Ecuador population represents a different species, named *P. pseudovulgaris* (*P. deboucki*) and it shared a common ancestor with the Mesoamerican and Andean groups, that remains to be discovered or has become extinct. Here, by analyzing the phylogeny of *P. vulgaris* we aim to better investigate the *P. vulgaris* origin and verify the different hypotheses. A wide sample that represents the entire geographical distribution of the wild forms of the species was genetically characterized for chloroplast genome diversity. A concatenated sequence of 3,231 chloroplast informative sites was used to build a phylogenetic tree. Moreover, 39 *de novo* chloroplast genomes were assembled and used to provide a temporal frame of the divergence for the analyzed genotypes, suggesting that the separation between the Mesoamerican and the North Peru-Ecuador gene pools occurred 0,15 Mya. Nuclear data, from the resequencing of a sample of ten accessions, were used to corroborate results. Overall, analyses of nuclear and plastid data support monophyletic and Mesoamerican origin of common bean.

## Evaluating rye and squarrose clover cover crops for no-till organic tomato production

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The omission of tillage operations i.e., no-till, is part of conservation practices that have gained increasing popularity since the 1970s in conventional agriculture. No-till has been recently tested in organic farming as an agroecological way for soil management in combination with cover crops. The choice of cover crop is key to addressing the challenges arising with this technique, often related to weed and nutrient management. A field plot trial was set up in 2019-2020 at the Centre for Agri-environmental Research "Enrico Avanzi" in Pisa (Italy) to evaluate the effects of three cover crops and their residues management on weed suppression, soil N availability and tomato yield. Rye (*Secale cereale* L.) and squarrose clover (*Trifolium squarrosum* L.) pure stands and their mixture in half rates, were either turned into the soil or flattened via roller-crimper before tomato transplantation. Weedy plots with no cover crop preceding tomato served as a control. At termination time, rye and the mixture had equally the highest dry residues per unit area. The presence of a living cover crop decreased winter-spring weed abundance measured in dry biomass. Rye and the mixture controlled weeds better than squarrose clover. Weed control continued during the season till harvest with a notable performance by the mixture in no-till. Weed community composition seemed also to be influenced by tillage regime and cover crop choice. Tomato plant growth and yield measured in fruits number and fresh weight were affected by the cover crop. Tomato plants following squarrose clover and the mixture produced more than the control, while probably N immobilization from rye residues hindered tomato growth and yield. Our work showed that legume-cereal cover crop mixture such as rye-squarrose clover can be a good choice for no-till to be successful in Mediterranean organic vegetable systems-

## Multi-parametric evaluation of mechanical termination of cover crops: experimental procedures and preliminary results

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The termination of cover crops in conventional low-till systems is mostly conducted mechanically in combination with herbicide treatments. The optimal selection and combination of different tools may lead to a significant reduction of herbicide use for cover crop termination, or even to avoid it. The multidisciplinary project X-Cover aims at studying the efficiency of the mechanical termination conducted with different combinations of tools and to estimate their economic viability (in terms of fuel consumption and mechanization costs) when compared to herbicide treatments. This paper presents the experiments designed to obtain this information through field tests on cover crops of vetch (*Vicia villosa*) and barley (*Hordeum vulgare*) conducted at different stage of development by means of an instrumented, modular prototype with three interchangeable set of tools (harrow with anchors, disc harrow, crimper roll). A suite of on-the-go proximal sensing techniques and direct measurements conducted at sampling points are used to characterise the different treatments through multiple parameters, as the termination efficacy (dead/ alive over ground biomass ratio; water stress of plants); effects on the top soil layer (soil roughness, soil water content, aggregate distribution, soil penetration resistance); energy requirements and associated costs (draft power, fuel consumption). The measurements methods are discussed along with some illustrative preliminary results.

## Study the bioethology of a quasi social parasitoids as a first step in biological control

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Biological control is an important component of integrated pest management (IPM) because it mainly attempts to manage invasive pests through the introduction of natural enemies such as predators, parasitoids or pathogens. Due also to the increase in limitation of the use of phytosanitary products imposed by law for their disproportionate use in the past and side effects in the environments, biological control has been recently re-adopted. Xylophagous insects are among the most difficult plant pests to control, especially with traditional chemicals, as they spend most of their life cycle inside natural wood or wood packing materials. *Psacotheta hilaris hilaris* (Pascoe) (Coleoptera, Cerambycidae, Lamiinae, Lamiini), the yellow-spotted longhorn beetle that causes damages to plants in the family Moraceae, is one of the exotic beetles imported into Europe in the last decades and whose distribution is slowly expanding. For its control, parasitoids assume a key role, becoming the subject of numerous studies. However, potential biocontrol agents must be screened prior to release, paying particular attention to their bioethology, to optimize their application. The quasi-social parasitoid *Sclerodermus brevicornis* Kieffler (1906) (Hymenoptera: Bethyridae) is able, thanks to its morphology, to enter the galleries of xylophages in search of the host larva, and several females are able to cooperate to paralyze and ovideposit on larger hosts. Furthermore, as it is very difficult to observe *S. brevicornis* in nature as its lifecycle is spent mostly inside wood galleries, studies of bioethology in laboratory finalized to balance the number of parasitoids with the hosts (intraspecific competition) and the interaction with other parasitoids (intraguid parasitization) become an essential starting point for the use as control agents.



## Evaluation of different “Attract and Kill” techniques against olive fruit fly *Bactrocera oleae* (Rossi) [Diptera: Tephritidae].

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The main pests for the olive groves are the olive fruit fly *Bactrocera oleae*. The larvae feed on the olive fruit and cause important economic losses. The control of this pest relies on broad spectrum insecticides applications, such as dimethoate, which are recently (EU Reg. 2021/155 of 9 February 2021) revoked. In this context, the aim of this study was to evaluate the efficacy of eco-friendly control techniques in different condition of olives production. The survey was carried out in the olive season 2020 and 2021 in the Lake Garda area on two experimental olive groves. Two “*Attract and Kill*” techniques were evaluated: “bait spray” Spintor™Fly (hydrolysed protein as lure and spinosad as insecticide, “*Mass trapping*” with plastic cone trap lured with sexual pheromones and ammoniacal as attractive, and deltamethrin as insecticide inside (Flypack®). The plots (3-4 ha) were set-up following EPPO standards (PP 1/280(1)). The adult flies were sampled through crossbar traps (ISAGRO®), while the infestation level was evaluated twice a week from August to harvest date observing the presence of sterile stings, larvae, pupae, and exit holes. The two investigated years were characterized by very different yields: 14,0 and 0,1 Tons olives/ha produced respectively in 2020 and 2021. Spray bait had significant reduction of the olive fruit fly population in the late autumn only in 2021 compared to the untreated plot. Although the number of catches was different over the two years in the treated and untreated plot, the infestation rate was significant lower in the treated plot. “*Mass trapping*” showed a similar population density reduction in both years, but the traps were efficacy only in 2020 where higher olive production was observed. The use of “*Attract and kill*” techniques in integrated olive fly management with high and low olive production were discussed.

## Predicting the potential distribution of *Toumeyella parvicornis* Cockerell in the Mediterranean basin

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*Toumeyella parvicornis* (Cockerell, 1897) (Hemiptera, Coccidae), is a soft scale insect native to North America, recently introduced in Italy and in France where it has spread causing harmful infestations on plants belonging to *Pinus* genus. In Europe, *T. parvicornis* has been reported mainly on stone pine (*Pinus pinea* L.) and less often on maritime pine (*Pinus pinaster* Aiton). Desiccation and yellowing of the needles represent some of the main symptoms of its presence. Furthermore, the pest releases copious quantities of honeydew followed by the growth of black moulds on pine twigs. Some aspects of its biology such as the several overlapped generations, the high fecundity and possibility to reproduce as sexual as by parthenogenesis may contribute to make it a successful invasive species when in a suitable environment.

Hence, in order to predict the pest spreading pattern and to plan management strategies, it is crucial to learn which could be the areas with the highest risk of invasion by this pest, interlinked with the geographical distribution of the host species across all the Mediterranean basin. Our aim was to obtain a map of the potential distribution of *T. parvicornis* in the Mediterranean basin, combining the available occurrence points data of the pest and bioclimatic variables. We resorted to the Maxent Species Distribution Model to obtain a predictive map of environment suitability showing different risk rates areas of potential occupation, based on the favourable environment for the pest. Our results showed that the areas with the highest suitability for the species are located on the coastal areas, where are also distributed mostly of the Mediterranean pines. This correspondence suggests a high risk of a large-scale diffusion and it gives a useful information to implement management strategies of this damaging pest.

## The use of biochar to mitigate the risk of a realistic exposure of pesticides applied to *Brassicaceae* crops

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The massive use of Plant Protection Products useful for preventing plant diseases of horticultural species, often occurs with the adoption of unsustainable agronomic practices resulting in different environmental impacts. In light of this, the present work assesses the use of biochar, applied to improve the yield of cauliflower crops, as a sustainable agronomic practice to reduce the leaching of pesticides. Moreover, the use of biochar with one biological phytosanitary treatment was also compared. We investigated the potential of two biochar, one based on wood waste "Wood biochar" and another composed of agricultural and forestry waste, "Mix biochar". Five experimental lines were set up in a greenhouse; each line was composed of four replicates of *Brassica oleracea* L. var. botrytis. The biochar was applied in two lines with an amount equal to 5% of the total soil volume. The other lines were composed of plants planted "Without biochar"; a "Control" line and a line treated with the biological insecticide Spinosad and the fungicide Trichoderma. The pesticides used in the other lines were Azoxystrobin fungicide plus Spinosad. The treatment was applied for nebulization two times and the percolation water was collected from each replicate to analyse the agrochemicals four times: 10 and 30 days after the first treatment and 13 and 21 days after the second one. The highest residue of pesticides was found especially after the second treatment with a maximum concentration of Azoxystrobin > 3000 ng/L in the line where biochar was not been applied. This quantity was ten and six times higher than that one observed in wood and rainbow biochar respectively. However, at the final time (21 days after the second treatment) the lower amount of this pesticide was observed just in the biochar Wood line with a concentration of 109 ng/L. Otherwise, for Spinosad, a suggested better retain capacity was shown from biochar biochar Mix with respect to biochar Wood. Finally, the Trichoderma line exhibited the highest concentrations of Spinosad at the final time of monitoring. In conclusion, the results show that the presence of biochar does not always increase the absorption capacity of pesticides in soil. This ability can vary depending on the type of biochar used and probably can reflect the general conditions to which the plant is subjected.

## An Uncertainty Hovers in Forestry

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Tree height is a fundamental measure in forestry being used to describe local stand fertility, forest ecological structure or to estimate forest biomass. Error in height measure necessarily affects the accuracy of derived estimates of dendrometric or forest parameters (i.e. carbon stock). Ordinarily, tree height is not directly measured but obtained through hypsometer. Hypsometer operates angular and distance measurements that are used to derive tree height by trigonometric computation. This method is affected by errors mainly related to the identification of the actual tree apex (often difficult to be recognized in dense stands) and to the measurement process itself. For operative reasons hypsometric measurement is performed with no fixed support thus introducing a variability due to operator's hand motion in collimation that affects angle measures. Additionally, other operational conditions: user-to-target distance, terrain slope, stem curvature, crown shape, affect measures introducing errors that are hardly to be detected and minimized.

In the proposed study, a method for estimating uncertainty of tree height measurements was proposed based on the variance propagation law. The latter can provide estimates of the theoretical variance of indirect measures (tree height) considering the known ones of those measures directly obtained by hypsometer (angles and distances). Some simulations were performed involving several combinations of terrain slope, tree height and distances by modelling the behaviour of height uncertainty and its sensitivity to such parameters. Simulations proved that height uncertainty could vary between 0.5 m and 5 m.

The results have been finally summarized in graphs that are expected to be operationally used to properly calibrate ground campaigns and to make users aware of the final uncertainty of their measures. Authors are, in fact, convinced that a higher consciousness about forest data uncertainty can drive to more reliable forest deductions.

## **Effect of cover crops introduction in a long-term vegetable succession**

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Maintaining an appropriate level of soil organic matter is essential for sustainable agricultural productivity. However, in the last decades, the agricultural areas have faced progressive soil impoverishment. Excessive use of mineral fertilizers, tillage operations, as well as too short and frequent crop cycles have progressively degraded the soil quality, especially in the horticultural production systems. In this context, a three-year study was established at the University of Padova experimental farm, with the aim of investigating the effect of winter cover crops (CCs) introduction in a long-term horticultural succession. This latter has been subjected to a fertilization trial (nitrogen supplied at different doses with organic matrices and/or mineral fertilizer) since 2005. The study, which is still ongoing, began in 2020 with the introduction of a CCs mixture (triticale-pea) in half of each experimental plot maintaining a spinach cash crop in the other half as a comparison. During the following spring-summer season, cabbage and green bean have been cultivated as cash crops. In the first cycle, the total CCs biomass was not affected by the fertilization management ( $3.9 \text{ Mg ha}^{-1}$  on average), even though the triticale produced more biomass ( $3.5 \text{ Mg ha}^{-1}$ ) than peas ( $0.4 \text{ Mg ha}^{-1}$ ). The fertilization management instead influenced one of the following cash crops (cabbage). Neither the cabbage nor the pea was affected by the CCs or the spinach presence in the winter season. The CCs presence, however, has determined a lower soil nitrates content ( $6.4 \text{ mg Kg}^{-1}$ ) compared to the spinach ( $11.4 \text{ mg Kg}^{-1}$ ). These preliminary results suggest that the short-term effect of CCs introduction in a long-term vegetable succession does not impact the cash crops yield, but decreased the soil nitrates content potentially leachable.

## Stomatal dynamics in grapevine: new traits for optimizing resources-use efficiency?

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In the field, grapevine is subjected to a series of constantly mutable environmental stimuli occurring over variable temporal time frames (e.g., whole season, days, hours and seconds). Non-synchronous responses between the photosynthetic machinery and stomatal movements can lead to reduced intrinsic water-use efficiency owing to slow stomatal closure and a limitation in CO<sub>2</sub> uptake following a significant sluggishness in stomatal opening. We showed that genotypic variation exists in the dynamics of stomatal movement in grapevine to fast changes in vapor pressure deficit and light conditions and that these responses can be associated with seasonal traits such as carbon isotopic signature in the field. Similarly, stomatal opening during broader timeframes (e.g., early morning) showed significant genotypic variation and may be targeted for the fine-tuning of specific management scheduling (e.g., irrigation) and for breeding new varieties (or optimizing scion x rootstock interactions) with preferable diurnal gas-exchange patterns. Indeed, in situations of limited water availability, high temperatures and elevated vapor pressure deficit, prioritizing early morning photosynthesis may be a promising escape strategy where carbon uptake is ensured during the less stressful portion of the day. Further work will focus at better determining the interactions between different environmental stimuli (e.g. light signals) and stomatal movements in grapevine.

## **The role of Trees Outside Forests in the cultural landscape of the Colline del Prosecco UNESCO site**

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The multifunctional role of Trees Outside Forest (TOF) is largely recognized in scientific literature, but they are still rarely considered in forest inventories and planning, with consequent underestimation of their role and amount. In addition, their cultural role has rarely been considered both at scientific and management level. TOF characterize many European cultural landscapes, including the one of the Colline del Prosecco, inscribed in 2019 in the UNESCO World Heritage List. One of the reasons of the inclusion, in fact, is the landscape mosaic made of vineyards interspersed with small woodlands and tree rows. This paper focuses on two types of TOF, Small Woods and Linear Tree Formations (TOF NON A/U). Their detailed mapping and the performing of different spatial analysis, allowed to assess their role and to provide data for future monitoring and for local forest planning. Results confirmed that TOF NON A/U are one of the main features of the UNESCO site landscape: despite the limited overall surface (1.95% of the area), 931 different patches have been identified. Spatial analysis highlighted the key landscape and ecological roles, acting as intermediate features between large forest patches, and also an important role for hydrological protection (they can be found also in slopes above 80% of inclination). The study provided a detailed map and database of one of the main features of the Colline del Prosecco UNESCO site cultural landscape, verifying the multifunctional role of TOF NON A/U and the necessity to include them into local forest planning, but also suggesting their inclusion in national forest inventories.

## Responses of five *Quercus* species exposed to drought and flooding in a common-garden

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The frequency and intensity of drought and flooding events are expected to increase due to climate change effects. *Quercus* spp. is severely exposed to environmental changes across the Mediterranean basin, although several species are well adapted to drought conditions. The present study aimed to assess the responses of *Quercus ilex*, *Quercus coccifera*, *Quercus macrolepis*, *Quercus trojana* and *Quercus cerris* to drought and flooding towards understanding potential limitation during forest restoration processes. Southern Italy populations of oak species widely distributed throughout the Mediterranean range were selected for common-garden experiments. Under experimental conditions, 21 days of drought followed by 24 hours of flooding were performed on seedlings of 5 oak species. Measurements of biomass (e.g., leaf number, seedling height, total leaf area) and ecophysiological traits (e.g., relative water content, chlorophyll content, chlorophyll fluorescence) were carried out to determine the intensity of water stress. While anatomic traits of xylem (morphology, hydraulic diameter, theoretical specific conductivity, vessel number for mm<sup>2</sup>) and metabolic and proteomic characteristics were determined to discriminate species responses. The data collected aim to determine whether functional variations in stress responses are performed by seedlings of different species, and, if any, how the single species reacts to constrained conditions. The combined analysis of functional and structural traits for detecting stress signals in oak seedlings was complemented by monitoring the available water in pots to assess plant performance under limiting moisture conditions and, consequently, to estimate the evolutionary potential of oaks under changing environments scenarios.



## **Assessing resilience components in maritime pine provenances grown in common gardens**

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Knowledge acquisition on the response of tree species to drought in the Mediterranean hotspot is an important step to guide adaptation strategies to climate change impacts, e.g., assisted migration. We assessed the resilience components – i.e., resistance, recovery, and resilience – to drought in 2003, in five provenances of maritime pine planted in four common gardens in Sardinia and analyzed the influence of climate variables on these indices. Genetic variation influenced growth rates but not the components of resilience. Among the provenances, Corsica was the most productive, while Tuscany the least. Portugal showed the best performance in warmer common garden sites. The resilience components were, instead, influenced by environmental conditions at the common garden sites. In the dry sites, trees showed the lowest resistance but the highest recovery values. However, two sites, which had the lowest stand density, recorded opposite trend during the drought year, probably influenced by moderate thinning. Resilience components were related to climate variables in different ways. Resistance and resilience had a similar pattern, both being positively related to precipitation and negatively to maximum temperature, while recovery showed an opposite trend. This indicates a noticeable adaptation of maritime pine to drought conditions, though the age factor should be considered. Despite only minor differences among provenances were found, environmental conditions at the common gardens and their management were important in determining tree growth. This study suggests the provenance of Corsica may provide appropriate material for forest plantations in Mediterranean conditions with mitigation purposes.

## Assessment of biochar amendment in nitrate capture, soil quality, N-metabolism, and yield of *Brassica oleracea* L. var. *botrytis*

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Nitrogen (N) is an essential nutrient for plant growth and yield. In agriculture, the massive use of nitrogen fertilizers results both in a low quality of market agriculture products, and pollution of surface and groundwater by nitrates. In recent years, the slow release of nitrate through the application of carbon-rich amendment (i.e. biochar) has been suggested, as a sustainable strategy for increasing agronomic and environmental benefits. Numerous studies have reported the positive effects of biochar in increasing nitrogen use efficiency and plant growth. However, the impact of the biochar application in soils treated with fertilizer and agrochemicals on *Brassica oleracea* L. var. *botrytis* is unknown. The objective of this research was to evaluate the effects of two biochar types on yield, growth, and inflorescence quality of cauliflower, treated under N fertilizers and pesticides. This study also investigated soil quality and the nitrogen compounds dynamics in environmental matrices. A greenhouse experiment was conducted, during cauliflower growing season, to examine the effects of biochar amendment (*wood* and *mix* biochar) in combination with N fertilizers and pesticides. A control test was performed without treatments. Soil physical-chemical properties and nitrate quantification in percolation water were investigated in samples collected at the end of the experiment and 10 days after each agriculture practice, respectively. Furthermore, biochemical and agronomic parameters were studied on cauliflower plant tissues at the marketable yield stage. The analyses are occurring. The results of soil morphological structure by scanning electron microscopy show the increase of nutrients in biochar macroporous structures. Soils amended with *mix* biochar were characterized by the increase in total nitrogen (N) and carbon (C) content compared to treatments with *wood* biochar and fertilizers. Finally, the first results revealed that plant growth and chlorophyll content increased in biochar amended soil compared to untreated.

## Biochar and *Cannabis sativa* L. as alternative for integrated bioenergy production and restoration of potentially toxic elements contaminated soils

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The increased consumption of non-renewable resources has significantly contributed to severe environmental pollution. Bioenergy production can supplant fossil fuels consumption but often subtracts valuable land for food or feed production. However, this is not the case when soils contaminated by potentially toxic elements (PTE) are used to cultivate bioenergy plants. In these soils, the growth of selected plants may be favoured by adding amendments, like biochar, which may improve soil fertility and reduce PTE bioavailability. To verify this, softwood-derived biochar was added at 3% rate to a mining soil contaminated with Sb (2175 mg·kg<sup>-1</sup>), Zn (3149 mg·kg<sup>-1</sup>), Pb (403 mg·kg<sup>-1</sup>), As (343 mg·kg<sup>-1</sup>), and Cd (12 mg·kg<sup>-1</sup>), to evaluate its influence on soil fertility and functionality (i.e. soil respiration and enzyme activities), PTEs mobility and bioavailability. The *Cannabis sativa* L. productivity (biomass and seeds), PTEs uptake and bioaccumulation were also assessed. Biochar addition increased cation exchange capacity (CEC) and the soil organic matter content, though the dissolved-organic-carbon decreased by 2.2-fold in the amended soil. Biochar reduced PTEs mobility, leading to a slight increase (between 1.02- and 1.05-fold) of their residual fraction. At the same time, soil respiration and enzyme activities (i.e., urease and alkaline phosphomonoesterase) increased in the amended soil, as did biomass and seeds production of *C. sativa* (i.e., 1.7- and 2.0-fold respectively). With few exceptions, biochar addition increased the PTEs concentration in roots, while PTEs in shoots and seeds decreased or were not affected. The PTEs removal efficiency by *C. sativa* increased in plants grown in the amended soil, though their translocation from roots to shoots decreased (except for Sb and Pb). Taken together, these results suggest that *C. sativa*, in combination with softwood biochar, could be used to produce biomass and seeds and favour the functional restoration of PTE contaminated soils.

## **Particulate matter emissions from agricultural activities: the maize case study**

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Soil can be resuspended in the atmosphere due to wind or mechanical disturbances, such as agricultural activities (sowing, tilling, etc.), producing fine and coarse particulate matter (PM). Agriculture is estimated to be the third PM<sub>10</sub>-emitting sector in Europe, emitting more than the transportation sector. However, very few emission figures are available for the different cropping operations, e.g., currently the emission factors (EF) proposed by the European environmental agency for tilling operations are based on few studies, none of which has been made in the Mediterranean area. Moreover, the soil Emission Potential is extremely variable, since is influenced by factors such as humidity, texture, chemical composition, and wind speed. Thus, within the project funded from the CRT Foundation "Emissioni di polveri sottili dalla filiera del Mais" the emission of some important agricultural operations (tilling, sowing, harvesting) were studied through field and laboratory experiments, to calculate EF of different agricultural soils used for maize cropping in North-Western Italy. In laboratory studies, a Soil Resuspension Chamber (SRC) was developed to estimate the impacts of soil moisture and physico-chemical characteristics on soil susceptibility to emit PM<sub>10</sub> during cropping operations (the Emission Potential). Although PM<sub>10</sub> emitted from agricultural operations is not identified as a problem, PM has been recognized as priority pollutant and carrier of noxious substances such as Potentially Toxic Elements (PTE). Thus, in soil emitted PM<sub>10</sub>, PTE were observed to be up to 16 times more concentrated than in the original soil, evidencing a possible cause of concern for operator's safety during agricultural activities.

## Land-use change from arable lands to orchards with grassed floor improves soil quality: a study case in Emilia-Romagna Region

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Soil is increasingly recognized to be an important provider of a wide range of ecosystem services for local and global societies. However, soil is a non-renewable resource on a human life scale and, therefore, maintaining the soil in good condition is of vital importance. The main aim of the present contribution was to investigate the influence of land-use change from arable land to fruit orchards on soil organic carbon (SOC) pools and on the soil microbial community activity. In southeast part of Emilia-Romagna Region (Italy), we selected a ten- and fifteen-years-old peach orchards, a twenty-years-old pear orchard, a thirty-years-old kiwi orchard and an arable land as reference. In the orchards, soil was covered by natural grasses and pruned wood materials were shred and left on the soil surface. Within each field, soil samples were collected from 0–15 cm depth, and the SOC pool amounts and the soil microbial biomass and its activity were measured. The increased SOC concentration and C stock in soils due to the land-use change from an arable land to fruit orchard would suggest the capacity of the selected orchards cultivation systems to enrich soil of organic matter. Noteworthy was the accumulation of the most stable SOC pool. The investigated land-use change increased the soil suitability for the microbial community which showed a higher C use efficiency. The accumulation of stable C forms and the improved microbial metabolism could be attributed to the high input of fresh and readily utilizable organic matter coming from both plant residues and floor weeds. Overall, the present contribution indicated that fruit orchard where soil was covered by natural grasses and pruned wood materials were shred and left on the soil surface should be promoted for improving soil quality and functioning.

## **Life cycle assessment (LCA) of different aquaculture systems: preliminary results from the SIMTAP project**

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Aquaculture is a major contributor to the growing demand in seafood even if it implies environmental concerns related to the consumption of feed, the emission of nutrients and organic compounds into the water and, sometimes, the consumption of pesticides and antibiotics. In this study, LCA was applied to assess the environmental impact related to seabream and seabass farming of different offshore plants located in Central Italy. The selected functional unit (i.e., the reference unit of the study to which all the inputs and outputs should be referred) was 1 ton of fish at the fish farm gate. The system boundary includes the production of feed and other production factors consumed (e.g., fuel, electricity), the rearing operations and all the emissions related to the process (e.g., phosphorous and nitrogen emissions due to the metabolism of the fish during the entire production cycle). For Gilthead Sea bream and European sea bass, the environmental results, for most of the impact categories considered, showed that aquafeed is the main hotspot. For the Climate Change impact, aquafeed impact ranges from 60 to 75% of the total, while for freshwater and marine eutrophication the contribution of feed is second only to that of the emissions of P and N compounds, respectively. The analysis highlighted a strong relation between aquafeed conversion rate, amount of N and P emitted and, consequently, the impact on eutrophication. In a context where the sustainability of production process must be improved, studying new sustainable fish diets is nowadays urgent. Following the example of the SIMTAP system, such new diets should be characterized by limited transportation impact (use of locally produced raw material and diets consumption) and maximizing the use renewable energy and protein sources (e.g., solar power and microalgae).

## Smart irrigation strategies for tomato: a Mediterranean case study

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The increasing weather anomalies (temperature increase and precipitation alterations), outline future scenarios of water shortage for the Mediterranean. It is urgent to increase water use efficiency and crop yields by applying improved irrigation strategies. In this study, a smart irrigation system was adopted to investigate the effect of different management strategies on processing tomato under water deficit conditions. An open field experiment managed by CREA-PB and DAGRI was conducted on processing tomato (Var. Durpeel F1) during the 2021 crop season. Four different irrigation treatments with drip irrigation were tested: full irrigation (Fi) - (100%), deficit irrigation (Di) - (80%), regulated deficit irrigation (RDi) - (60-80-60%) and farmer level deficit irrigation (FLDi) - (60%). The trial was monitored by a smart Decision Supporting System (DSS) based on IoT, artificial intelligence and machine learning, connected to field sensors for soil moisture and weather station for a predictive optimization of irrigation under a controlled water regime. Di, RDi and FLDi have shown significant yields decrease of 42.02, 50.22 and 78.82 %, respectively, compared to Fi. No significant differences were observed for the Irrigation Water Productivity (IWP= Yield ha<sup>-1</sup> /irrigation amount) related to the fresh yield, among Fi, Di and RDi. FLDi showed lowest values among the others. No significant differences in canopy cover were observed between treatments. In conclusion, despite the decrease in yields, maintaining a high WP level can be considered as part of an irrigation strategy in case of water scarcity for processing tomatoes cultivation in Mediterranean regions. This study demonstrates that Fi, Di, and RDi managed by DSS have brought the most promising results. A final consideration is that the deficit irrigation strategies require deep knowledge of crop response to drought stress, and this may explain the complications observed for FLDi in this first year of experimentation.

## **Ecofriendly humic-like substances from the upscaling of municipal biowaste as biobased fertilizers**

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The management of municipal biowaste (MBW) constitutes a well-known environmental and economic burden for society, due to increasing population, human urbanization, and consumption habits. Currently, MBW is treated in equal parts by controlled fermentation, incineration and landfilling. Fermentation yields compost and anaerobic digestate with a very low market value.

The LIFE EBP project (LIFE19 ENV/IT/000004) addresses environmental problems in municipal biowaste management suggesting the conversion to new biobased products (BPs) for agricultural and chemical industry starting from the chemical hydrolysis of the anaerobic fermentation digestate.

This upscaling of the digestate to be used as soil fertilizer, plant biostimulant and anti-pathogen agents in agriculture is being studied through lab-scale, greenhouse and field experiments to obtain high added-value products to replace commercial fossil-based products and related GHG emissions.

In the present study, BPs were studied as fertilizers in greenhouse test in two plant models (lettuce and spinach) and their effect on soil activity and crop production was defined through soil enzymatic activity, morphometric measures and crop production and quality.



## **Ruminant enteric Methane Emissions of a High-Input and a Low-Input Dairy Production System**

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Ruminant enteric methane emissions are critical contributors to climate change. The aim of this study was to quantify and compare methane emissions of a low-input and a high-input dairy production system that have been monitored simultaneously for three years in Dietsheim/Bruneck, Northeast Italy. The high-input system is characterized by year-round indoor housing of Simmental cattle that are fed on a concentrate-based ration, while the low-input system consists of the breed Tyrol Grey cattle which is held predominantly on pasture during the vegetative season and fed on a forage-based ration. The ruminant enteric methane emissions have been estimated according to different equations using dry matter intake, energy corrected milk yield, or the fatty acid profile of the milk as predictors. This allows evaluating the enteric methane emissions of both systems over the year on an animal level and per liter milk.

## **The conservation of endangered livestock breeds: an agricultural economics perspective**

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Since 90s, European Union acknowledges the importance of agricultural biodiversity with the adoption of payments, in the Common Agricultural Policy, aimed at improving the conservation status of agricultural breeds, habitats and landscape. Specifically, the conservation of traditional endangered livestock breeds contributes to the long-term resiliency of agricultural activities. However, traditional livestock breeds provide farmers a lower income in terms of production of milk and meat. Moreover, the direct payments provided by the CAP often do not compensate the differences in productivity and thus can not be considered a sustainable and definitive solution for the conservation of these breeds in the long term. Certainly, scholars can contribute the conservation of these endangered livestock breeds through the development of new conservation approach. Nonetheless, for this to be possible, the involvement of breeders and final consumers is essential. In the frame of the INNOVA Project, related to the preservation of the Varzese-OttoneseTortonese (V.O.T.) cattle, this research aims to explore the drivers and constraints for the conservation of an endangered native cattle breed from the breeders as well as consumers side. The first part of the research has been implemented through in-depth interviews with all the cattle breeders members of the V.O.T. Association (n= 10). These interviews were aimed at detecting breeders' characteristics, knowledge and acceptance of reproductive technologies, constraints and drivers related to endangered cattle breeding and possibilities for future improvement. Results from this study give a hint to support breeders in the development of more sustainable conservation strategies for endangered native breeds. Moreover, our findings highlight the possible role of agricultural economics in the development of successful biodiversity future interventions.

## **Breeding for quality and health: genetic parameters for somatic cells and milk traits in the local cattle producing PDO “Parmigiano Reggiano” cheese**

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Increased milk yield is a primary goal in dairy cattle breeding because of its preeminent relevance in determining herd profitability. Nevertheless, the quality of the milk is equally important as the level of production, especially in breeds such as the Reggiana due to the impact on the quality of the dairy products, such as the PDO “Parmigiano Reggiano” cheese. In this sense the Somatic Cell Count (SCC), commonly recorded according to the Italian official milk recording system, provides some information about milk quality, and is the most common indicator of udder health status, considering its high positive genetic correlation with mastitis. In most countries, the genetic evaluation of udder health is based on the logarithmic transformation of SCC into Somatic Cell Score (SCS) to achieve normality of distribution. This study aimed at calculating heritability, genetic and phenotypic correlations among SCS and milk traits in the Reggiana breed. About 32500 test-day records of 8500 cows (9900 animals in the pedigree) were used. Heritabilities were in the order of 7-22% for milk yield and fat and protein production, and values less than 7% in the case of SCS. Negative genetic correlations were found between milk yield and total solids, supporting the antagonistic relationships between both kinds of traits reported in the literature. Genetic and phenotypic correlations between SCS and milk yield were positive, which could mean that increasing milk productivity leads to a detriment of udder condition and, subsequently, increases SCS level in milk. Fat and protein yield were negatively correlated with SCS, which could mean that selection focused on increasing the percentage of solid content could also be beneficial for udder health. The study highlighted the importance of accounting for udder health when selecting for production and quality in dairy cattle.

## **Non-genetic factors affecting milk differential somatic cell count in specialized and dual purpose dairy cows**

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In addition to somatic cell count (SCC), milk differential somatic cell count (DSCC) has been proposed in recent years to monitor cow's udder health status. Milk DSCC (%) represents the proportion of polymorphonuclear neutrophils out of the total SCC and, according to the literature, values > 65 % indicate inflammation of the mammary gland. The present study aimed at investigating the DSCC variability using test-day data (TD) collected in multibreed herds and from cows belonging to i) a specialized dairy (Holstein) and ii) a dual purpose breed (Simmental) and sampled in North of Italy. Only herds with both breeds present were kept and restrictions on SCC allowed outliers elimination. For each TD, DSCC\_n (cells\*1,000/mL) was calculated by multiplying SCC (cells\*1,000/mL) and DSCC. Finally, 112,748 TD from 12,009 Holstein and 3,830 Simmental cows in 266 herds were available to estimate the least square means of DSCC and DSCC\_n through a linear mixed model. The fixed effect of breed, parity (1, 2, 3 and ≥4), lactation stage (12 classes), and sampling season were considered together with the first-order interaction between breed and lactation stage, breed and parity and parity and lactation stage. The random effects were: cow, herd-test-date, and residual. Estimates of the two breeds were significantly different ( $P < 0.001$ ) being DSCC equal to  $64.19 \pm 0.06$  and  $62.54 \pm 0.10\%$  and DSCC\_n equal to  $296 \pm 2.52$  and  $210 \pm 4.40$  cells\*1,000/ml in Holstein and Simmental, respectively. Overall, both traits were greater in Holstein than in Simmental cows, regardless of parity and lactation stage. However, in late lactation estimates of the two breeds were similar ( $P > 0.05$ ). Preliminary results suggest that the conventional DSCC threshold might be partially adjusted to better fit the udder health status in Simmental breed.

## **Associations between milk mineral profile and blood energy metabolites in Holstein cattle**

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The study of milk fine composition – in which the mineral profile is the most unexploited – is gaining interest as it mirrors the cows' health and metabolic changes that occur especially in the early lactation period. This study aimed at assessing the potential associations between 15 macro- and micro- mineral elements and a set of metabolic disorders indicators, specifically: i) ultrasound (US) measurements of liver (triacylglycerol liver content [pTAG], portal vein area, portal vein diameter [PVD] and liver depth) and ii) blood energy metabolites (non-esterified fatty acids [NEFA],  $\beta$ -hydroxybutyrate [BHB], glucose, cholesterol and urea). Milk and blood were analysed with official analytical procedures and US liver measurements were taken from 300 Holstein cows in the first 120 days of lactation (DIM). Minerals traits were analysed with a linear mixed model including as fixed effect cows' DIM, parity, milk yield, and the above-mentioned metabolic disorder indicators, and herd/date as random effect. The macro-mineral category exhibited the highest number of significant associations ( $P < 0.05$ ). Potassium was positively and linearly associated with urea, negatively with BHB and with a negative quadratic pattern for NEFA and PVD. Phosphorus was linearly positively associated with glucose and urea, negatively with BHB and with a quadratic trend for PVD. Sulphur highlighted a linear negative association with BHB and pTAG, and a negative quadratic trend with PVD. Significant associations were found for some essential micro-minerals and contaminants. Our results indicate the existence of associations between the milk mineral profile and the hepatic/haematic measurements confirming the role of milk as a source of information mirroring the animal health status, which could contribute to the development of an animal health barometer to be used at the population level.

## **Improving animal welfare by means of automatic monitoring of goat vocalizations: development of a bleating recognition filter**

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Ensuring high levels of animal welfare is fundamental for a better and more sustainable use of resources in animal farming. In fact, high levels of animal welfare are related to higher immune response, lower risk of disease, lower use of drugs and antibiotics, and, therefore, fewer drug residues in the environment, thus leading to better global health. The VOCAPRA project aims to study goats' vocalizations in order to improve human-animal communication and provide farmers with an early warning tool to detect poor animal welfare conditions and intervene to meet the animals' needs. To this aim, the development of a filter, i.e. a non-linear supervised algorithm, was initially required for distinguishing goat vocalizations from all other sounds. For a whole year starting from April 2021, we recorded sounds in four goat farms, using 18 acoustic sensors. We selected 150 audio tracks and classified them as: noise (n=60), adult goats' vocalizations (n=50), and kids' vocalizations (n=40), based on the prevailing sound. Then, the tracks classed as adult goats' vocalizations were edited to isolate only bleats. Using a sliding window technique, each track was then split into smaller tracks, whose acoustic characteristics were extracted and subjected to a random forest classifier algorithm, obtained from the sklearn.ensemble library, to quantify the number of bleats. On the basis of the number of bleats we then assigned a score from 0 (no bleats) to 1 (only bleats) to each smaller track. The algorithm was subjected to k-validation training (k=5) using for each k the 80% of all classified audio tracks, while the remaining 20% was used to test the algorithm. We reached an algorithm accuracy of 82%, suggesting the possibility of using it to select goats' vocalizations', which is the first step for developing an IT tool for monitoring goat welfare based on vocalizations.

## **Fertility and management parameters in a tool for the simplified evaluation of global warming potential related to the milk production**

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For improving sustainability of the dairy sector, farmers and technicians need to have reliable and easy tools to identify the strategies that make the best use of resources, minimizing the environmental impact. In this sense, the overall efficiency of the farms should be considered, including fertility and management parameters of the herd. The aim of the study was to develop an equation for the evaluation of global warming potential (GWP), related to milk production, starting from results of the Life Cycle Assessment analyses and technical variables about management of the herd and land area. Data of fertility and management of the herd were provided by Associazione Nazionale Allevatori della Razza Frisona, Bruna e Jersey Italiana (ANAFIBJ). Statistical analysis was carried out using SAS 9.4 software. A GLM procedure was performed, on 25 dairy farms, to build an equation suitable to estimate the GWP of milk production at farm level (kg CO<sub>2</sub>eq/kg FPCM, fat and protein corrected milk). From the results obtained, the best equation to estimate GWP of 1 kg of FPCM, included the following variables: % alfalfa on the total land, dairy efficiency, milk protein content, primiparous cattle restocking, lactation duration, pregnancy rate, FPCM/hectare, number of lactation/cow, amount of feed self-produced, individual daily milk production, % cornsilage/DMI and interpartum. The average value of GWP, for the 25 farms, was 1.45 kg CO<sub>2</sub>eq/kg FPCM (SD 0.25). The tool enables to have findings quickly, by providing a method easily applicable on farm scale, without the need for tabulated data or empirical formula. The tool allows farmers and technicians to compare different management strategies to enhance sustainability. In addition, it allows using already existing data concerning fertility and management of the herd, which means finding a clear vision of milk production sustainability at national level and its trend over the time.

## **Could Automatic Milking Systems reduce dairy farms greenhouse gases emissions?**

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Dairy farms produce GreenHouse Gases(GHG), although emission intensity is reducing from the 50s to today. A further improvement is possible and required. Therefore, the aim of this study is to analyse the contribute of Automatic Milking Systems (AMS) in increasing environmental sustainability of farms. A Life Cycle Assessment (LCA) analysis was done on two farms (A and B) in northern Italy. Then, LCA was rebuild, assuming farms changes including AMS: 8 scenarios were created supposing changes in Milk Production (MP), increasing it by 5% (MP5) and 15% (MP15), in Fat and Protein Content (FPC), considering no increase and increase respectively of 0.10% and 0.06%, in Feed Purchases (FP) and in Energy Consumption (EC), increased by 1.8 kWh and 2.44 kWh per 100 liters of milk. Improvement in FPC slightly reduced GHG emissions from 0.04% (A), to 0.06% (B). On the contrast, FP affected emissions negatively: due to growing of protein purchases, emissions increased from 0.86% (A, MP5) to 3,59% (B, MP15). Also EC slightly affected GHG emissions: between the lower and the higher EC, they only varied by 0.07% (A) and 0.15% (B). Overall, in both farms, AMS resulted in a reduction of GHG emissions from a minimum of 3% (MP5) to a maximum of 9% (MP15). This result is mainly due to the increase in MP, able to compensate higher FP and EC, even in the worst scenario. In conclusion, AMS appears to reduce the environmental impact of dairy farms, without considering any influence on the reduction of somatic cell count, which could hopefully further decrease GHG emissions of farms.



## **Application of Selective Dry Cow Therapy protocol based on total and differential somatic cell count to reduce antimicrobial use at dry off**

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The large majority of farmers use intramammary antimicrobials (AT) on all cows at the beginning of dry period in order to prevent future mammary infections and to stop eventually ones in act (Blanket Dry Cow Therapy, BDCT). In 2022 in Italy BDCT was banned with the aim of reducing antibiotic resistance. One solution may be Selective Dry Cow Therapy (SDCT), which consists in the selection of cows that cannot be treated at dry off (NT). The aim of the study was to evaluate a SDCT protocol based on somatic cell count (SCC) and differential ones in milk at the end of lactation. According to the SDCT protocol, the last test milk in the 30 days before dry off of NT had to be <100.000 SCC/ml for primiparous and <200.000 SCC/ml for multiparous cows, with the sum of neutrophils and lymphocytes <69,3 % of SCC. 847 milk samples from three farms of Lombardy were analysed in 2020 and 2021. 239 cows were monitored at the end of lactation and at the beginning of the subsequent one and 73 were selected and treated only with teat sealant (NT). Results obtained highlighted that at the end of lactation, AT had higher SCC compared to NT (5.17 and 4.66  $\log_{10}$  SCC/ml for AT and NT respectively,  $P<0.01$ ). At the beginning of lactation (during the first 60 days) milk SCC of AT and NT was pared to 4.76 and 4.87  $\log_{10}$  SCC/ml, pointing out the same mammary condition of the two groups ( $P=0.78$ ). SCC was, on the average <100.000 SCC/ml, both for AT and NT. SDCT protocol, therefore, is useful to select AT and NT, with a subsequent reduction of antimicrobial use of 30%, with no consequences on farmers' profits and mammary welfare.

## **Importance of calf management in dairy farms: an Italian investigation**

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In dairy farms, calves' management is often neglected, as unproductive animals. The aim of this study is to observe the situation of dairy calves management in 118 dairy farms located mainly in northern Italy (92.3%) using a questionnaire. SAS® software was used to perform statistical analysis and calculate averages, frequencies, correlations and multiple correspondence analysis. About 60.7% of farms have herds with at least 101 lactating cows and 85.6% have intensive breeding methods, 75.2% without grazing. The 42.3% of farms take care of calves during milking, the 40.5% after the end of the same and only 22.9% declared the absence of disease. Calf area in about 69.0% of farms is outside and 31.0% is inside but only the 8.85% dedicated an area only for calves. The 46.6% of farms use single housing, 22.0% multiple box, 31.4% use different types of housing and the 29.5% keeps calves in single housing beyond the limit established by law. The 54.4% of farms separate cow-calf within 1 hour from birth. The 97.4% of farms always administer colostrum and the average time of administration is  $3.03 \pm 4.38$  hours, only 45.2% of farms administer colostrum within 2 hours of life and the 69.2% of farms provide maternal colostrum. Analysis of multiple correspondence showed that farms using mainly maternal colostrum generally tend to separate the calf from the mother later (>1 hour) and to provide colostrum less promptly (>2 hours). These parameters appear to be correlated with a birth to weaning mortality rate lower than 5%, maybe because staying with the mother, calf takes with breastfeeding more colostrum useful for immunity. Young animals represent the future of the herd, so their health and management are essential to obtain healthy and consequently productive adult animals.

## **Towards an in-flight assessment of gaseous and particulate emissions of livestock farming: development of a prototype UAV-based system**

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Quantifying and reducing greenhouse gases (GHGs) emissions in livestock systems is a highly debated topic. International policies are supporting mitigation strategies to reduce the environmental impact of agricultural practices. Besides, the assessment of air quality in livestock buildings for ensuring human and animal safety and welfare is poorly addressed and no real-time monitoring systems are currently available. In this framework, automated and low-cost tools enabling a continuous monitoring of air concentrations of gases and particulate in livestock buildings, manure and feed stores would represent a significant advancement in the sector. Given that drones are increasingly used for air quality monitoring in several fields (e.g. atmospheric chemistry research, industrial emission monitoring), the aim of our project was to assess the feasibility of a UAV-based system for real-time measurements of air pollutants at farm level. We present a first attempt to develop an integrated prototype system for gas and particulate monitoring using portable self-engineered measurement units at ground and on a small UAV, to detect emission hotspots and to provide real-time graphic alerts by means of a web-app. The system embeds low-cost commercial sensors for GHGs (CH<sub>4</sub>, CO<sub>2</sub>), NH<sub>3</sub> and particulate (PM<sub>2.5</sub>, PM<sub>10</sub>) into customized portable units located at ground and on a rotor-based drone. The sensors were calibrated in a specialised laboratory and the system was tested in a commercial dairy farm to assess the feasibility of the project. Ground measurement units were located inside and close to the external boundaries of the cattle building, while simultaneous flights were carried out in the top atmospheric boundary layer up to 30 m a.g.l. Gas and particulate concentration measurements were timestamped and georeferenced with centimeter accuracy. The results confirmed the feasibility of the project at farm level, although further research is required to validate field measurements with reference instruments and techniques.

## **Exploitation of the GEE platform to Apennine grasslands monitoring**

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Grasslands in mountainous and marginal areas provide a wide variety of ecosystem services, in addition to agricultural goods and disaster protection. In the Apennines, extensive grassland systems face a gradual reduction in exploitation or abandonment. To successfully manage these ecosystems, it is important to define the proper stocking rate of livestock by estimating nutritional quality and pasture production. Optical satellite sensor imagery, which is increasingly available and free of charge also thanks to the Copernicus program, provides an important contribution to this estimation. As satellite imagery has become more accessible, platforms such as Google Earth Engine (GEE) have been developed for fast, simple, and powerful data processing. GEE enables global data analysis by exploiting a large catalog, as well as disseminating results to a wide audience through a simple web link. Our study is developed as part of the VISTOCK project (<https://vistock.toscanallevatori.it/>), supported by GAL START TOSCANA, which involves the integrated use of grazing herd management innovative technologies, known as Virtual Fencing collars and precision livestock tools. Our contribution involves the development of a code on the platform GEE, which allows, starting from the coordinates of a single point, the computation of several vegetation indices (VI) for different buffer areas, the automatic time series visualization through interactive plots, and the downloading of the data in .csv format. Thus, the VI are correlated to observed data (e.g. LAI, fPar, biomass and forage quality) taken along two grazing seasons (2020–2021). With our study, a code for pasture quality monitoring activities will be presented and disseminated freely. Future perspective will be the possibility of integrating the information on pasture quality and production directly from the field via tablet or smartphones to support farmers in setting a rational grazing in semi-extensive pastures according to the forage supply during the season.

## Sewage sludge amendment for rice: a potential alternative to inorganic fertilizer

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In a context of circular economy, municipal solid waste can be adequately treated producing sewage sludge (SS). SSs are rich in organic and inorganic plant nutrients, and exert beneficial effects on soil properties. A greenhouse experiment was conducted on rice plants (*Oryza sativa* L. var. Sirio CL) to assess the effects of three different SSs: SS1, treated with CaO, SS2, treated with CaO and CO<sub>2</sub>, and SS3, treated with CaO and H<sub>2</sub>SO<sub>4</sub>. SSs were compared with unamended soil (control) and mineral fertilizer (urea, 4 g/pot). Physiological and biochemical analyses were carried out on leaves at tillering initiation, booting and panicle emergence. In the first stage, all SSs increased chlorophylls (Chls) content (+49%), and SS3 also in the following stages. At first sampling, antioxidant capacity was 2-, 3- and 2.5-fold higher than controls in SS1, SS2 and SS3, respectively. This result seems related to the increased ascorbic acid (2-fold higher than controls) only in SS1 and SS3, whilst in SS2 other antioxidant molecules could be involved. At the end of the experiment, biomass production significantly increased in all SSs (+38%) than controls, while in SS2 and SS3 even compared to urea (+39%). Likewise, a positive correlation ( $r=0.95$ ) was found between the total amount of nitrogen and biomass in plants grown on SS2 and SS3, which increased their total nitrogen content of 73% and +60% compared to controls and urea, respectively. Apparently, none of SS treatments induced any oxidative stress, as shown by the measurements of malondialdehyde by-products and photosynthetic efficiency ( $F_v/F_m$ ). In summary, the observed increase of antioxidant activity could help plants to face out potential environmental stress during the transition from vegetative to reproductive stage. Moreover, the increased Chls content may have determined an enhanced photosynthetic activity, inducing a significant increase of total biomass at the end of the experiment.

## Grafting with non-suckering rootstocks stimulates stress resilience of European hazelnut (*Corylus avellana* L.) to drought

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Physiological and molecular processes underpinning water stress responses in European hazelnut (*Corylus avellana* L.) still need deeper investigation; in particular, no studies addressing the influence of non-suckering rootstocks in the regulation of such responses are available for this species. In order to fill these gaps, we contrasted changes in gas exchanges and stem water potential ( $\Psi_{\text{stem}}$ ) of two hazelnut genotypes, the commercial San Giovanni cultivar (SG) and the non-suckering rootstock, Dundee (D), and in their corresponding heterograft (SG/D), during a drought stress treatment followed by recovery. Already in well-water conditions, stomatal conductance ( $g_s$ ) was significantly higher in SG than in SG/D and D plants. However, along with drought progression,  $g_s$  values dropped less and more gradually in SG/D and D than in SG plants. The rootstock effect in influencing the stomatal regulation of SG scions was also evident after re-watering. Indeed, unlike  $\Psi_{\text{stem}}$ ,  $g_s$  recovery followed different dynamics based on genotype or grafting. In D,  $g_s$  recovered completely in 7 d, while in SG, it never reached the pre-stress levels, even after 14 d. However, when SG was grafted onto D roots,  $g_s$  recovery did occur and was completed in 10 d. We hypothesized that the observed differences could rely on changes either in endogenous metabolic signals - particularly in abscisic acid (ABA) concentrations - or in leaf stomata density and/or dimensions. We therefore analyzed ABA levels in roots and leaves from well-irrigated, severely stressed and recovered SG, D and SG/D hazelnuts, and we integrated these data with analysis of proline content and key candidate genes. Additionally, we compared the stomatal dimension and density of SG and D leaves with those determined on the heterograft. The collected results provide first evidence that grafting with a non-suckering rootstock may represent a strategy to improve resilience of hazelnut cultivars to drought.

## Biochar soil amendment for the urban tree establishment phase: What consequences for tree physiology, soil quality and environment?

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Trees play a pivotal role in the urban environment alleviating the negative impacts of urbanization. Indeed, local governments promoted strong tree planting policies. However, the poor soil qualities and neglect tree maintenance (e.g., irrigation and fertilization) can seriously mine the plant health status during the tree establishment phase. Among soil conditioners, the use of biochar to sequester CO<sub>2</sub> and, at the same time, improve soils properties, thanks to its influences on the soil water-holding capacity, soil enzymes activities and NPK contents, is an emerging research field. Therefore, the study aimed to assay firstly the physiological responses of a commonly used urban tree species (*Tilia hybrida* "Argentea") to 1.5 % biochar amendment, and secondly, the ability of biochar amended plants (CB) to tolerate a period of drought. Moreover, the potential benefits of biochar soil amendment to CO<sub>2</sub> sequestration were calculated. Thanks to the improved soil properties, CB plants reported a higher nutritional status through the growing season than controls (Cnt). This condition, in addition to a better water balance, helped CB trees to tolerate the potential environmental constraints during the establishment phase (e.g., high temperatures and water shortage), showing superior leaf photosynthetic performances. The improved plant and soil conditions in CB treatment contributed to increase the total tree biomass compared to controls. In addition, the total sequestered CO<sub>2</sub> in the CB treatment (plant and soil) increased by 133 % than Cnt. This study highlighted the dual functions of the biochar soil amendment promoting the sequestration of CO<sub>2</sub> and, at the same time, ameliorating the soil properties resulting in an enhancement of plant physiological responses to environmental constraints. The use of biochar at the tree planting, especially in an urban environment, is a feasible and environmentally sustainable strategy to improve the success during the tree establishment phase.

## Copper extraction potential of citrate and malate in vineyard soils

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Fungi are one of the most important pests limiting agricultural production. To control fungi mainly copper (Cu)-based fungicides are used. This has led to an increasing Cu accumulation in soils in the last century. Copper in soil affects the soil quality, microorganisms, nutrient bioavailability, as well as plant health. To counteract this, plants release root exudates such as malic and citric acid into the rhizosphere. These organic acids can alter Cu uptake by forming complexes and thus influencing Cu availability. Therefore, we aimed to evaluate the Cu mobilization potential of malic and citric acid in three vineyard soils with differing Cu content. Copper was extracted for 24h through shaking with different combinations of citric acid, malic acid, as well as  $\text{KNO}_3$  as background electrolyte and  $\text{NaN}_3$  for preventing microbial degradation. The extraction solution was analyzed for Cu content, as well as for the organic acid content. Both malic and citric acid presented the potential to mobilize Cu. The two acids in combination were revealed to have the strongest extraction capability, followed by citric acid and malic acid alone. This is most likely due to differences in the carboxyl groups. The mobilization potential also decreased with decreasing soil pH, higher soil organic matter content and higher clay content. Moreover, both organic acids have shown higher degradation than adsorption to soil particles, indicating a strong influence of microorganisms. Citric acid displayed higher adsorption than malic acid, most likely due to its additional carboxyl group. It can therefore be concluded that soil pH, organic matter content, soil texture, type of organic acid and microorganisms are the most important factors affecting the Cu mobilization. To better determine the influence of root exudates, it would be recommendable to also investigate other important exudates.



# **Groundwater contamination by glyphosate, glufosinate ammonium, and AMPA: occurrences and sources in hilly vineyards**

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Glyphosate [N-(phosphonomethyl)glycine] and glufosinate ammonium [ammonium dl-homoalanin-4-(methyl) phosphinate] are broad-spectrum, nonselective, post-emergence herbicides extensively used in various applications for weed control in aquatic systems and vegetation control in non-crop areas (Barker and Dayan 2019). Aminomethylphosphonic acid (AMPA) is the major degradation product of glyphosate found in plants, water, and soil. Numerous toxicological studies reported negative effects of glyphosate, its metabolite AMPA, and glufosinate ammonium to mammalian and aquatic organisms (Geetha, 2021). Both glyphosate and AMPA have been detected in several groundwater samples in Europe (EEA, 2020), U.S (Battaglin et al., 2014), Canada (Van Stempvoort et al., 2016), Argentina (Demonte et al., 2018; Okada et al., 2018) and China (Geng et al., 2021). Nevertheless, very little is known about the influence of land use and the impact of crop type on surface and groundwater contamination by glyphosate, AMPA, and glufosinate ammonium (Medalie et al., 2020). In the present work the occurrences and the main drives of glyphosate, AMPA, and glufosinate ammonium in the groundwater of a hilly vineyards located in north-west of Italy were evaluated. Groundwater monitoring results showed frequent detection and concentrations above EQS for groundwater for glyphosate and AMPA, while glufosinate ammonium was never detected. Such monitoring results were not expected as the modelling estimations under the local pedoclimatic conditions, using FOCUS PEARL model, suggested their low risk of leaching to groundwater. Furthermore, the wells' locations and their uses showed the most frequent and highest concentrations in wells located in farmyards, if compared with wells located in vineyards, indicating a non-crop site use of glyphosate in the farmyard as the most likely contamination source.

## **Variable-rate fertilization of durum wheat (*Triticum durum* Desf.) based on geophysical mapping**

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Proximal soil sensors are receiving strong attention from several disciplinary fields, and this has led to a rise in their availability in the market in the last two decades. The aim of this work was to validate agronomically a zone management delineation procedure from electromagnetic induction (EMI) maps applied to two different rainfed durum wheat fields. The k-means algorithm was applied based on the gap statistic index for the identification of the optimal number of management zones and their positions. Traditional statistical analysis was performed to detect significant differences in soil characteristics and crop response of each management zones. The procedure showed the presence of two management zones at both two sites under analysis, and it was agronomically validated by the significant difference in soil texture (+24.17%), bulk density (+6.46%), organic matter (+39.29%), organic carbon (+39.4%), total carbonates (+25.34%), total nitrogen (+30.14%), protein (+1.50%) and yield data (+1.07 t ha<sup>-1</sup>). In addition, on site 2 has been evaluated crop response to Variable rate techniques (VRT) was compared to uniform nitrogen application (UA) on the whole field. The application of VRT resulted in a reduction of 25% nitrogen fertilizer with the same level of yield respect to UA. Grain protein content, as well as gluten content and N content, were significantly higher in VRT than in UA. Because of lower nitrogen input and higher levels of N removal, VRT reached a higher nitrogen use efficiency than UA, and this indicates a lower environmental impact and a higher economic profitability.

Moreover, six unmanned aerial vehicle (UAV) flight missions were performed to investigate the relationship between five vegetation indexes and the EMI maps. The results suggest performing the multispectral images acquisition during the flowering phenological stages to attribute the crop spatial variability to different soil proprieties.