

Report on the outcomes of a Virtual Mobility¹

Action number: CA21134

Grantee name: Sebahat Öztekin

Virtual Mobility Details

Title: Microbiota-based approaches for sustainable crop protection: recent achievements and future perspectives

Description of the work carried out during the VM

The Virtual Mobility was carried out on 10th September 2026, with the objective of preparing a synthesis report entitled "Microbiota-based Approaches for Sustainable Crop Protection: Recent Achievements and Future Perspectives," contributing to the final report of COST Action CA21134 (T0P-AGRI-Network). The work combined a targeted review of recent scientific literature with the grantee's previous research experience in microbial biocontrol, particularly antagonistic yeasts and postharvest disease management. The grantee's relevant contributions include experimental studies on *Metschnikowia* spp. against postharvest fungal pathogens, yeast consortia against *Penicillium digitatum*, and a comprehensive review addressing antagonistic yeast mechanisms and mycotoxin prevention strategies. A recent international review on postharvest biocontrol agents was also considered in framing emerging perspectives, including microbial consortia, omics-based approaches, and other innovative biological protection strategies. Recent literature was reviewed covering beneficial bacteria, including *Bacillus* and *Pseudomonas* spp.; antagonistic yeasts such as *Metschnikowia*, *Meyerozyma*, *Hanseniaspora*, *Aureobasidium*, and *Debaryomyces* spp.; and mycoparasitic fungi including *Trichoderma* spp. Particular attention was given to mechanisms of action, environmental robustness, field translation, formulation, and factors limiting wider adoption. An **online expert meeting was organised on 10 September 2026** to complement the literature-based synthesis with perspectives from researchers within and associated with the COST network. Participants included **Rachid Mentag (INRA, Morocco), Heike Seybold, Javier Arizmendi, Muhammad Usman, Nadia Mocrea, Azeez, Jeremy, and Sebahat Öztekin**. The discussion addressed recent advances, practical barriers, research gaps, and future implementation of biological control agents. **Rachid Mentag (INRA, Morocco)** provided particularly relevant input on the potential of **beneficial microbial consortia for manipulating native soil and rhizosphere microbiota and suppressing soil-borne pathogens such as *Fusarium***. He also highlighted the major challenge of inconsistent BCA performance under changing temperature, humidity, and

¹ This report is submitted by the grantee to the Action MC for approval and for claiming payment of the awarded grant. The Grant Awarding Coordinator coordinates the evaluation of this report on behalf of the Action MC and instructs the Grant Holder Manager to verify the grant documents compliance and proceed with the payment.

drought conditions. The discussion further emphasised farmer perception, training, economic feasibility, practical usability, and the need for more field and demonstration trials as key factors determining successful adoption. Expert perspectives gathered during the meeting were incorporated into the final synthesis. The work plan was completed without major deviations.

Abstract

The global transition toward sustainable agriculture demands the replacement of synthetic pesticides with biologically-based strategies that safeguard crop yield and quality without compromising human health or ecosystem integrity. Microbiota-based approaches — encompassing antagonistic yeasts, plant growth-promoting bacteria (PGPB), microbial consortia, and emerging synthetic microbial communities (SynComs) — have emerged as scientifically robust and commercially viable alternatives. This synthesis integrates evidence from three decades of postharvest biocontrol research, landmark plant-microbiome studies, and the author's own experimental and review publications on yeast-based biocontrol agents (BCAs) applied to citrus and mandarin fruits. Recent findings from high-impact review literature (Droby et al., 2025; Compant et al., 2025; Simon et al., 2025) confirm that microbial communities shape plant health through multiple overlapping mechanisms, and that next-generation strategies combining multi-omics, AI, and precision inoculant engineering will define the future of crop protection. Identified research gaps — including field-to-shelf consistency, climate-change resilience of BCAs, and regulatory harmonisation across the EU — provide a roadmap for future collaborative research within the T0P-AGRI-Network COST Action.

1. INTRODUCTION: THE IMPERATIVE FOR MICROBIOTA-BASED CROP PROTECTION

Chemical pesticides have underpinned global food security for more than a century, yet their long-term application has generated cascading consequences: pesticide-resistant pathogen populations, soil microbiome disruption, groundwater contamination, and increasing public and regulatory pressure for greener alternatives [1,2]. The European Union's Farm to Fork Strategy sets a legally binding target of 50% reduction in the use and risk of chemical pesticides by 2030, reinforcing the urgency of biopesticide and biocontrol research under EU Regulation 1107/2009 [2,3].

Within this regulatory and ecological context, microbiota-based crop protection strategies offer multifaceted value: they reduce pathogen pressure through direct antagonism, prime plant immunity through induced systemic resistance (ISR), support soil health by preserving beneficial microbiome diversity, and align with the One Health framework that recognises the inseparability of plant, human, and environmental health [4]. The COST Action CA21134 (T0P-AGRI-Network) explicitly targets these intersections by fostering transnational collaboration on top-crop agroecosystem microbiomes.

This synthesis report summarises the author's own research contributions in yeast-based postharvest biocontrol on citrus and mandarin fruits [5–8], contextualises them within the latest international literature on plant microbiome management [1,3,4], and draws actionable conclusions for collaborative research priorities within the network. The report directly addresses the two thematic questions posed for the September 2026 expert meeting: (i) the recent state of the art in microbial biocontrol, and (ii) research gaps and perspectives that define the next decade.

2. RECENT ACHIEVEMENTS IN MICROBIAL BIOCONTROL

2.1. Three Decades of Postharvest Biocontrol: From Concept to Commerce

The discipline of biological control of postharvest diseases has matured from proof-of-concept laboratory experiments in the 1980s into a field with registered commercial products and a sophisticated mechanistic understanding. Droby et al. [1] provide the most comprehensive recent synthesis of this evolution, documenting how early antagonist selection was based on in vitro inhibition assays, whereas contemporary approaches employ microbiome sequencing, metabolomics, and AI-assisted prediction to identify and optimise BCAs.

Commercial milestones include Aspire® (*Candida oleophila*, Ecogen Inc.), Noli®/Shemer® (*Metschnikowia fructicola*, Agrogreen), Boni-Protect® (*Aureobasidium pullulans*, Bio-Ferm), Botector® (*Aureobasidium pullulans*, Bio-Ferm), and Nexy® (*Candida oleophila*, Lesaffre), demonstrating that yeast-based BCAs can achieve commercial registration in the EU, USA, and Israel [1]. The commercial trajectory validates the scientific basis established by antagonistic yeast research and provides regulatory precedent for novel candidates.

2.2. Antagonistic Yeasts on Citrus and Mandarin Fruit: Author's Contributions

The author's research programme has focused specifically on yeast-based BCAs for postharvest citrus and mandarin protection — a high-value segment directly relevant to Mediterranean agriculture. A JAFC 2023 review paper [5] synthesised the global state of knowledge on antagonistic yeasts used against postharvest pathogens, documenting that genera including *Metschnikowia*, *Meyerozyma*, *Hanseniaspora*, and *Aureobasidium* consistently demonstrate multi-mode efficacy against *Penicillium digitatum* (green mould), *P. expansum* (blue mould), *Botrytis cinerea* (grey mould), *Alternaria* spp., and *Fusarium* spp.

Experimental work reported in PHB&T 2021 [6] demonstrated the efficacy of *Metschnikowia* spp. on lemon surfaces, where the yeast reduced pathogen populations through rapid space and nutrient competition at wound sites, supplemented by the production of pulcherrimin — an iron-chelating red pigment that causes iron starvation of competing pathogens. This iron-depletion mechanism is particularly significant because it operates independently of antibiotic resistance in pathogen populations, offering durability that chemical fungicides increasingly lack.

Research on mandarin fruit reported in Bio Control 2023 [7] advanced the field by evaluating microbial consortia combining two or more antagonistic yeast species. Multi-strain combinations showed synergistic effects exceeding the biocontrol efficacy of individual strains, consistent with the principle — later elaborated by Droby et al. [1] — that pathobiome complexity is best countered by microbiome diversity. The consortia approach also provided complementary environmental tolerance profiles, improving BCA performance across the humidity and temperature gradients typical of commercial cold-storage chains.

The most recent contribution, a collaborative review with an international team published in *Frontiers in Nutrition* 2026 [8], broadened the scope to encompass not only biocontrol but mycotoxin biodegradation — an emerging application in which microbially-produced enzymes enzymatically cleave trichothecene, aflatoxin, and patulin structures, reducing their bioavailability in the food chain. This review highlights *Bacillus subtilis* and *Lactobacillus* spp. alongside yeasts as key actors, and notes that biodegradation represents an underexplored intersection of food safety and crop protection.

2.3. Plant Microbiome as a Reservoir for Biocontrol Agents

Beyond postharvest applications, the plant microbiome — encompassing rhizosphere, phyllosphere, and endosphere communities — constitutes a vast but incompletely catalogued reservoir of potential BCAs. Compant et al. [3] demonstrate in their landmark Nature Reviews Microbiology synthesis that plant-associated microbial communities perform functions essential to crop health: nutrient cycling (nitrogen fixation, phosphate solubilisation), pathogen suppression via antibiosis and mycoparasitism, and ISR priming that activates the plant's own defence signalling cascades.

Key bacterial taxa — *Bacillus*, *Pseudomonas*, *Rhizobium*, *Trichoderma* — have been most intensively studied for PGPB applications [3], yet the authors note a significant knowledge gap: most experimental evidence comes from controlled glasshouse settings, and field performance remains inconsistent. Soil composition, indigenous microbial competition, host genotype, and climatic variability all modulate inoculant establishment and persistence — challenges that are mirrored in postharvest settings where temperature shifts and coating interactions affect yeast BCA survival [1,5].

2.4. Microbiome-Based Food Safety Interventions

Simon et al. [4] extend the concept of microbiota-based crop protection to encompass food safety applications, demonstrating that microbial community engineering can reduce pathogen loads on fresh produce, degrade mycotoxin residues, and modulate the shelf-life microbiome to delay spoilage. Long-read nanopore sequencing now enables real-time monitoring of the fresh produce microbiome throughout the supply chain, creating opportunities for microbiome-informed quality management that was unimaginable a decade ago. Artificial intelligence algorithms trained on microbiome composition data are beginning to predict spoilage risk with accuracy exceeding traditional chemical indicator assays, pointing toward a data-driven future for postharvest microbiology [4,8].

3. MECHANISMS OF ACTION: A MULTIFACETED ARSENAL

A key insight from three decades of mechanistic research is that the most effective BCAs operate through multiple simultaneous modes of action, reducing the probability that any single resistance mechanism in the pathogen population will undermine biocontrol efficacy. The principal mechanisms documented across fungal and bacterial BCAs include:

- **Competition for nutrients and space:** Rapid colonisation of infection courts (wounds, natural openings) by BCAs pre-empts pathogen access to simple sugars, amino acids, and water [1,5,6]. Yeasts are particularly effective at this because of their rapid budding rate and ability to form biofilms on fruit surfaces.
- **Antibiosis and volatile organic compounds (VOCs):** Many *Bacillus* and *Pseudomonas* BCAs produce diffusible antibiotics (iturin A, fengycin, surfactin, 2,4-diacetylphloroglucinol) and VOCs (acetoin, 2,3-butanediol) that inhibit pathogen germination and hyphal growth [3,8]. Yeasts produce killer toxins (mycocins) and lytic enzymes (glucanases, chitinases) that disrupt fungal cell walls.
- **Iron depletion via siderophores:** *Metschnikowia* spp. produce pulcherrimin, a potent iron-chelating pigment that sequesters Fe^{3+} in the apoplast and on fruit surfaces, starving pathogens of an essential micronutrient [6]. *Pseudomonas fluorescens* produces pseudobactin siderophores with the same functional outcome in the rhizosphere [3].
- **Induced Systemic Resistance (ISR):** PGPB and some BCAs prime the jasmonic acid / ethylene signalling pathway, resulting in a faster and stronger defence response when the

plant subsequently encounters a pathogen [3]. ISR is broad-spectrum and durable and has been documented for *Bacillus subtilis*, *Pseudomonas putida*, and *Trichoderma harzianum* across multiple crop systems.

- **Mycoparasitism:** *Trichoderma* spp. physically coil around pathogen hyphae and secrete cell-wall-degrading enzymes (β -1,3-glucanases, chitinases, proteases), lysing pathogen cells. This mechanism provides direct, physical elimination of pathogen biomass independent of host plant signals [3,8].
- **Biofilm formation and quorum sensing:** BCAs that form structured biofilms on root or fruit surfaces achieve persistent colonisation resistant to washout. Biofilm-associated *Bacillus* communities produce extracellular matrices that physically exclude pathogens. Recent research highlighted by Drobny et al. [1] indicates that quorum-sensing molecules also modulate BCA community composition, opening potential for quorum-sensing-based biocontrol enhancement.

The convergence of multiple mechanisms in single BCA strains or consortia is a strategic advantage over single-mode-of-action fungicides, whose target-site mutations in pathogen populations rapidly erode efficacy. The consortia approach pioneered in the author's mandarin biocontrol work [7] exploits mechanism complementarity directly, with each consortium member contributing a distinct biocontrol mode.

4. PRACTICAL LIMITATIONS AND CURRENT CHALLENGES

Despite compelling laboratory and controlled-storage results, microbiota-based crop protection faces substantial barriers to widespread adoption [1,3]:

Inconsistent field performance. Inconsistent field performance is the most frequently cited limitation. The efficacy of BCAs under controlled conditions rarely translates fully to commercial settings, where temperature fluctuations, variable humidity, diverse pathogen populations, and existing microbial communities all interact unpredictably [1,3]. Compant et al. [3] identify edaphic variability — differences in soil pH, texture, organic matter, and indigenous microbial competition — as primary determinants of PGPB establishment failure in field trials.

Shelf life and formulation stability. Shelf life and formulation stability present commercial challenges. Living BCAs require controlled storage conditions, and the viability window of wettable powder or liquid formulations is typically shorter than for chemical fungicides. Encapsulation technologies (alginate beads, microemulsions, biopolymer coatings) are improving this situation but require species-specific optimisation [1,8].

Regulatory complexity. Regulatory complexity within the EU creates barriers to market entry. Active substance dossiers under EU Regulation 1107/2009 are expensive and time-consuming to prepare, and approval timelines for microbial BCAs can exceed five years. Harmonised risk assessment frameworks specific to living microorganisms — distinct from the chemical pesticide paradigm — are still under development [2,3].

Spectrum limitations. Spectrum limitations restrict single-strain BCAs to a narrow range of target pathogens. Broad-spectrum protection requires either multi-component consortia — which face additional regulatory burdens as combination products — or the identification of BCAs with inherently broad antagonistic activity [1,5].

Climate change impacts. Climate change is projected to shift pathogen pressure patterns, modify host plant physiology, and alter the temperature and humidity envelopes within which BCAs operate most effectively. BCAs selected for current climatic conditions may not remain

effective under projected 2050 scenarios, requiring proactive screening of thermotolerant and drought-resistant BCA strains [3,4].

5. RESEARCH GAPS AND FUTURE PERSPECTIVES

5.1. SynComs and Precision Microbiome Engineering

Synthetic microbial communities (SynComs) — designed assemblages of functionally characterised strains — represent perhaps the most promising near-term advance in biocontrol science. By combining strains with complementary mechanisms, non-overlapping niches, and known compatibility, SynComs can deliver broad-spectrum efficacy while minimising competitive exclusion within the consortium itself [1]. The transition from empirically assembled consortia (as explored in the author's mandarin work [7]) to rationally designed SynComs based on genomic and metabolomic data is a priority research direction.

Compant et al. [3] identify microbiome priming — the pre-inoculation of soil or seed microbiomes with defined communities to create a disease-suppressive environment before pathogen exposure — as an approach that could fundamentally change crop protection from reactive to preventive. Coupling SynCom inoculants with host-plant cultivar selection optimised for microbiome recruitment (the 'holobiont breeding' concept) represents a convergence of plant breeding and microbiology.

5.2. Multi-Omics Integration and AI

Metagenomics, metatranscriptomics, metaproteomics, and metabolomics are now routinely applied to characterise plant microbiomes, but their integration into predictive models remains nascent. Simon et al. [4] document early applications of machine learning to fresh produce microbiome data for spoilage prediction, while Droby et al. [1] note that AI-assisted screening of large strain libraries has accelerated BCA discovery tenfold compared to traditional plate assay approaches.

Long-read nanopore sequencing enables real-time, culture-independent identification of pathogen populations in storage facilities, creating data streams that can guide adaptive biocontrol deployment. The integration of these technologies into commercial supply chains — requiring affordable per-sample costs, cloud-based data pipelines, and farmer-accessible interpretation interfaces — defines a key research-to-practice gap for the coming decade [4].

5.3. Mycotoxin Biodegradation as Crop Protection Extension

The Frontiers in Nutrition 2026 review [8] highlights an often-overlooked dimension: BCAs capable of both preventing fungal infection and degrading mycotoxins produced by breakthrough infections represent a 'double-action' strategy with high food safety value. Key mycotoxins — deoxynivalenol (DON), zearalenone (ZEA), aflatoxins B1/G1, patulin — are regulated at EU level (EC 1831/2003) and their management represents a food safety imperative beyond yield considerations. Selecting BCA strains for combined biocontrol and detoxification capability should be integrated into future screening programmes.

5.4. Climate-Resilient and Region-Adapted BCAs

Mediterranean agricultural systems — including citrus, grape, stone fruit, and olive — will experience projected increases in mean temperature, drought frequency, and modified precipitation patterns under climate change scenarios. BCA strains isolated from extreme

environments (semi-arid regions, high-temperature fermentation environments) and screened for heat and osmotic tolerance are likely to be more climate-resilient than current commercial strains optimised for temperate conditions [3,4]. Bioprospecting from Türkiye's biodiversity-rich agricultural regions — citrus orchards, grape growing areas of Aegean and Mediterranean coasts — represents an opportunity to identify locally-adapted BCAs directly relevant to Turkish and EU Mediterranean agricultural needs.

5.5. Regulatory and Commercialisation Pathways

The T0P-AGRI-Network COST Action provides an ideal platform to coordinate harmonised regulatory submissions across EU member states, sharing data packages and dossier preparation expertise. Collaborative field trials across network partners in climatically diverse locations would simultaneously address the field consistency gap and generate multi-site registration data [2,3]. Exploring the 'basic substance' registration pathway under EU Regulation 1107/2009 — which offers streamlined approval for low-risk substances including food-grade yeasts — for BCA candidates isolated from commonly consumed fruit surfaces is a strategic priority.

6. CONCLUSIONS

Microbiota-based crop protection has progressed from a scientific curiosity to a commercially demonstrated, mechanistically understood, and policy-aligned strategy for sustainable agriculture. The synthesis of evidence from three decades of postharvest biocontrol research [1], current plant microbiome science [3], food safety applications [4], and the author's own experimental programme [5–8] confirms that:

- Antagonistic yeasts — particularly *Metschnikowia*, *Aureobasidium*, and *Meyerozyma* spp. — are highly effective, food-safe BCAs with multiple overlapping mechanisms that reduce the risk of resistance development.
- Microbial consortia outperform single-strain BCAs by exploiting mechanistic complementarity, a principle validated by both consortia experiments and the SynCom framework.
- The plant microbiome is both a reservoir for novel BCAs and a target for manipulation through inoculant design, host-plant selection, and microbiome priming.
- Critical research gaps — field consistency, climate resilience, regulatory harmonisation, and mycotoxin co-management — define the agenda for collaborative research within T0P-AGRI-Network.
- Multi-omics integration and AI-assisted BCA discovery will transform the pace and precision of microbiota-based crop protection research over the next decade.

The T0P-AGRI-Network VMG expert meeting of September 10, 2026 provides an invaluable opportunity to align these priorities with the expertise of network partners and to identify collaborative projects that will translate these perspectives into field-ready solutions for European and Mediterranean agriculture.

7. ACKNOWLEDGEMENTS

This work was supported by COST Action CA21134 (T0P-AGRI-Network) through a Virtual Mobility Grant awarded to Sebahat Öztekin, Atatürk University, Erzurum. The COST

Association is funded by the Horizon Europe programme of the European Union. The author thanks the expert meeting participants for their insights and collaborative engagement.

Description of the VM main achievements and planned follow-up activities

The Virtual Mobility achieved its primary objective through the preparation of a synthesis of recent advances, limitations, and future perspectives in **microbiota-based approaches for sustainable crop protection**. The work identified several major scientific advances, including improved understanding of microbial modes of action, increasing interest in multi-strain and microbial consortium approaches, and growing recognition of the importance of interactions between introduced biocontrol agents and the native plant or soil microbiome. Key mechanisms considered included competition for nutrients and space, antimicrobial metabolite production, siderophore-mediated iron competition, volatile organic compounds, killer toxins, cell-wall-degrading enzymes, biofilm formation, mycoparasitism, and induction of host resistance. The VM also identified important barriers preventing successful translation from laboratory studies to agricultural practice. A major issue highlighted both in the literature and during the expert consultation was **inconsistent efficacy under variable environmental conditions**. Temperature, relative humidity, drought, host physiology, and native microbial communities may substantially influence BCA establishment and performance. Climate change is therefore expected to increase the importance of developing biologically robust and environmentally adaptable microbial formulations. The expert consultation also reinforced the potential of **beneficial microbial consortia**, particularly for rhizosphere and soil-borne disease management. Combining microorganisms with complementary functions may improve ecological establishment and resilience compared with single-strain approaches. However, this potential requires validation through multi-location and multi-season trials. Another major outcome concerned the **human and economic dimensions of BCA adoption**. Farmers need to move from pesticide-centred crop protection towards integrated approaches in which biological agents are considered practical management tools. Expert input emphasised that adoption will depend not only on biological efficacy but also on ease of application, economic benefit, reliability, farmer education, demonstration trials, and confidence in the technology. The synthesis contributes to CA21134 by consolidating scientific evidence and expert perspectives on microbiota-based crop protection and by identifying priority areas for future network activity: **standardised efficacy assessment, climate-resilient BCAs, microbial consortium design, microbiome-level studies, field validation, techno-economic evaluation, and farmer-oriented knowledge transfer**. Planned follow-up activities include submission of the synthesis to the Action, continued scientific exchange with participating researchers, and exploration of opportunities for future collaborative publications and research proposals addressing microbial biocontrol and sustainable crop protection.

8. REFERENCES

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