

Review Article

BIOCONTROL POTENTIAL AND MECHANISMS OF ACTION OF PLANT ASSOCIATED YEAST

Abstract

Yeasts, single-celled fungi, have established themselves as effective biocontrol agents against various plant pathogens, demonstrating mechanisms such as nutrient and space competition, secretion of antimicrobial compounds, and induction of plant resistance. Their genetic stability, resilience to harsh conditions, and ease of cultivation on low-cost media enhance their applicability in agriculture. This review explores the diverse modes of action employed by yeasts, including biofilm formation, production of lytic enzymes, volatile organic compounds, and mycoparasitism, all contributing to their biocontrol efficacy. Notable registered yeast species like *Candida oleophila*, *Aureobasidium pullulans*, *Metschnikowia fructicola*, and *Saccharomyces cerevisiae* showcase commercial potential in managing plant diseases, with various products already available in the market. Additionally, yeast strains have been shown to enhance plant growth, improving crop vigour and yield. The commercial applications of yeast-based bioproducts highlight their potential as sustainable alternatives to chemical pesticides in agriculture, emphasizing their importance in integrated disease management strategies.

Introduction

Yeasts are single-celled microorganisms classified under the kingdom Fungi. They can be either facultative anaerobes or obligate aerobes, and typically reproduce by budding. Yeasts have long been utilized in industries such as baking and brewing, and are also involved in the preparation of various foods like idli and dosa. Additionally, some yeasts exhibit medicinal properties, making them useful as alternative treatments for human ailments such as diarrhea, common flu, and respiratory disorders. Certain yeasts, such as *Saccharomyces boulardii*, are probiotic in nature and contribute to lowering cholesterol levels and enhancing immune function in humans. In the plant environment, yeasts are predominantly found in the phylloplane, rhizosphere, and soil.

Why Yeast?

Yeasts possess several beneficial characteristics that make them excellent candidates for use as biocontrol agents (Agirman et al., 2023).

- i) Their unicellular morphology offers numerous practical applications, including ease of cultivation in fermenters, adhesion properties, and biofilm formation (Rossouw et al., 2018).
- ii) Yeasts are genetically stable and effective even at low concentrations.
- iii) They can endure harsh environmental conditions and are resistant to postharvest chemical treatments such as fungicides, pesticides, and phytohormones.
- iv) Yeasts are effective against a wide range of pathogens affecting various crops.
- v) They are easy to culture on low-cost media and are not highly demanding in terms of nutrient requirements.
- vi) Yeasts can be formulated into stable products with extended shelf life.
- vii) They are simple to store and distribute.
- viii) Yeasts do not produce toxic metabolites harmful to humans.

Modes of Action of Yeasts Against Plant Pathogens

Yeasts employ several mechanisms to combat plant pathogens, including competition for nutrients and space, secretion of antimicrobial compounds, production of lytic enzymes and volatile organic compounds, direct parasitism, and the induction of plant resistance (Freimoser et al., 2019).

1. Competition for Nutrients and Space

Nutrient and space competition is a critical factor in microbial ecology and is considered one of the primary modes of action for biocontrol yeasts. Iron is a particularly important nutrient for biocontrol yeasts, and competition for iron plays a key role in their antagonistic activity (Spadaro & Droby, 2016). In *Aureobasidium pullulans*, the siderophore fusarinine C (fusigen) has been identified as a compound with antibacterial activity, suggesting its role in iron competition within ecological niches (Wang et al., 2009). Gore-Lloyd and colleagues (2019) found that wild-type *Metschnikowia pulcherrima* colonies exhibited a distinctive red pigmentation due to the production of pulcherriminic acid, which complexes with iron. Mutant *M. pulcherrima* colonies, lacking this pigment, showed reduced iron competition and lower antifungal activity, as demonstrated by decreased inhibition (80%) of *Botrytis caroliniana* mycelium compared to the 98% inhibition observed with the wild-type

strain. This iron-deprivation mechanism is thought to be a key factor in *M. pulcherrima*'s antagonism of fungal plant pathogens. Furthermore, *Saccharomycopsis schoenii* lacks components of the sulfur assimilation pathway, likely acquiring methionine from its prey (Junker et al., 2019). Additionally, Bencheqroun et al. (2007) demonstrated that apple amino acids were significantly depleted at wound sites treated with *Aureobasidium pullulans* strain Ach1-1, particularly in combination with *Penicillium expansum*, compared to untreated wounds. The addition of exogenous amino acids reduced the efficacy of strain Ach1-1 in controlling *P. expansum*, suggesting that amino acid competition plays a role in biocontrol efficacy.

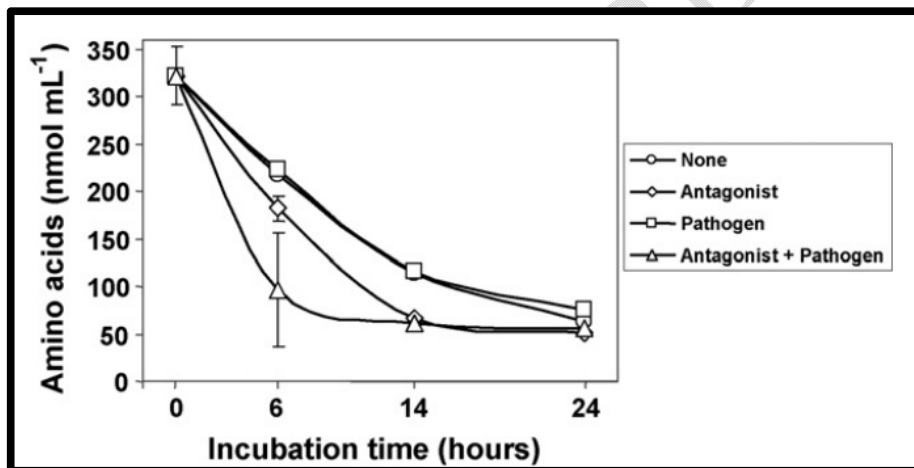


Fig 1 :Assimilation of apple amino acids by the antagonist and the

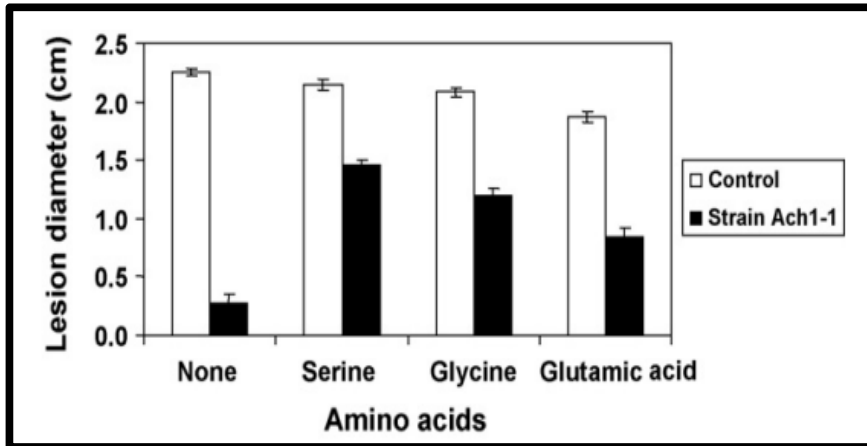
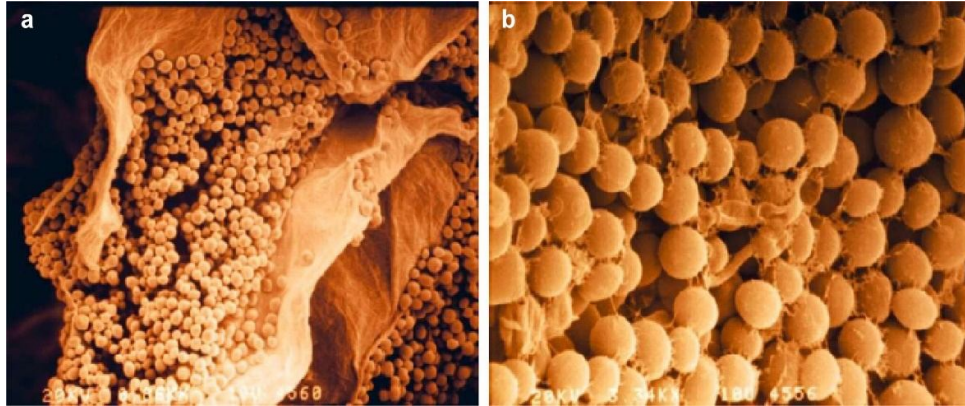


Fig 2 :In situ competition for nutrients

Biofilm Formation

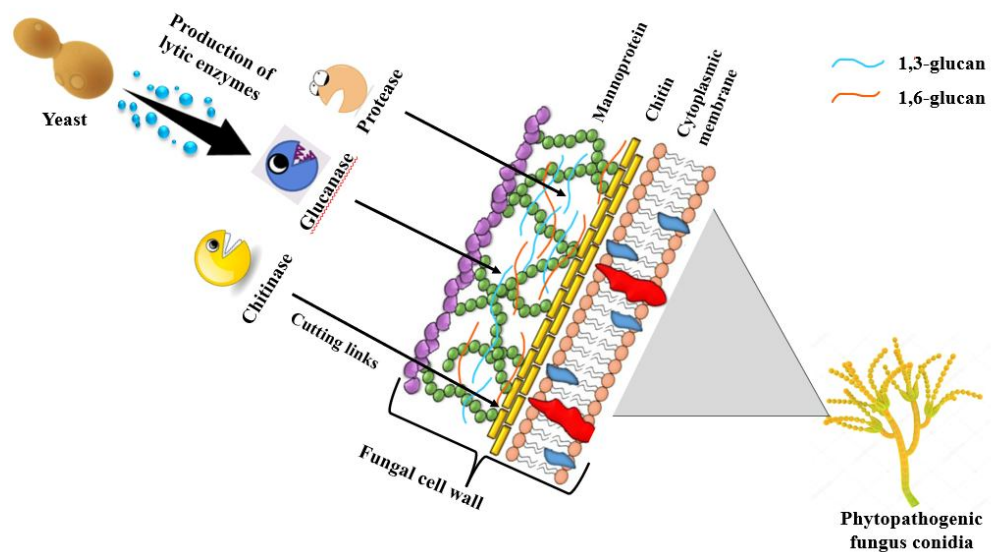
Biofilm formation is an effective and intricate strategy employed by microbial communities to compete for space on various surfaces. These biofilms can consist of a single microbial species or multiple species coexisting in consortia (Costa-Orlandi et al., 2017). The process begins with microbial cells adhering to a surface, followed by modifications in the cell wall, secretion of an extracellular matrix, and, in certain cases, the development of hyphae or pseudohyphae (Cavalheiro & Teixeira, 2018). In *Saccharomyces cerevisiae*, biofilm-forming cells have been shown to be significantly more effective than their planktonic counterparts in colonizing apple wound surfaces, which in turn enhances their ability to control the spread of blue mold caused by *Penicillium expansum* (Ortu et al., 2005).

Fig:3 :Biofilm Formation



1. Secretion of lytic enzymes

The secretion of enzymes that degrade cellular components is indeed a common feature in various host-pathogen interactions. This process is often a key virulence strategy employed by pathogens to gain access to nutrients, evade host immune responses, and cause damage to host tissues. Some of the lytic enzymes produced by yeast are:



2. Fig:4 :Secretion of lytic enzymes

a. Chitinases

The secretion of chitinolytic enzymes is a valuable strategy for biocontrol agents, particularly in fighting fungal pathogens. These enzymes degrade chitin, a key component of fungal cell walls, enhancing the effectiveness of biocontrol agents against plant diseases (Zajc et al., 2019). Various yeast genera, including *Aureobasidium*, *Candida*, *Debaryomyces*, *Metschnikowia*, *Meyerozyma*, *Pichia*, *Saccharomyces*, *Tilletiopsis*, and *Wickerhamomyces*, exhibit chitin-degrading activity, allowing them to target and dismantle fungal cell walls (Pretscher et al., 2018). Furthermore, the breakdown of chitin produces chito-oligosaccharides (CHOS), which can stimulate plant immune responses, thereby enhancing plant defenses against fungal infections (Langner & Gohre, 2016).

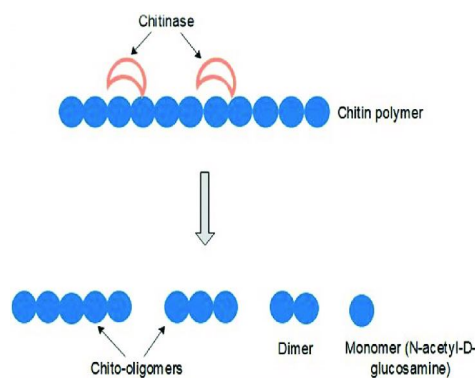


Fig:5 :Chitinases

b. Glucanases

Glucans are key components of fungal cell walls, and exoglucanases are enzymes that cleave the bonds between 1,3-glucan and 1,6-glucans, playing a crucial role in cell wall modification and adhesion (Xu et al., 2013). In *Pichia anomala*, two exo- β -glucanase genes, PaEXG1 and PaEXG2, have been linked to biocontrol activity against *Botrytis cinerea* in fruits, with the deletion of these genes resulting in a marked decrease in biocontrol efficacy (Friel et al., 2007). Additionally, Lopes et al. (2015) found that six isolates of *Saccharomyces cerevisiae* exhibited antifungal

properties against *Colletotrichum acutatum* by secreting exoglucanases in citrus.

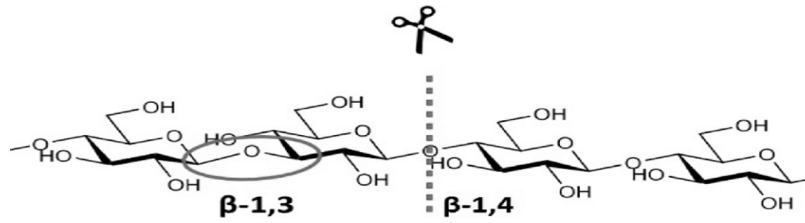
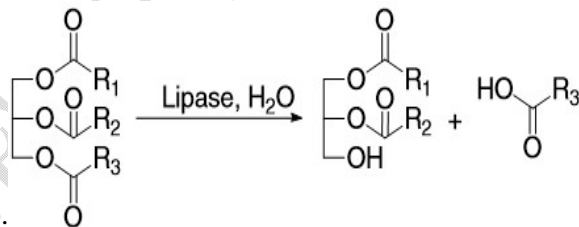


Fig 6 :Glucanases

c. Lipases

Lipases are enzymes that catalyze the hydrolysis of lipids, converting them into free fatty acids, glycerol, and other alcohols. Lipolytic activity is commonly observed during the screening of extracellular enzymatic activity in yeast and yeast-like strains (Arroyo-Lopez et al., 2008). Additionally, lipase activity has been identified in various pathogenic yeasts, including species of *Candida*, *Cryptococcus*, and



Malassezia (Sommer et al., 2016).

Fig 7 :Lipases

d. Proteases

Proteases are enzymes that cleave alpha peptide bonds between naturally occurring amino acids. In biocontrol yeasts, protease activity may be detected at later stages of biocontrol, suggesting its limited role in this process (Bar-Shimon et al., 2004). The alkaline serine protease Alp5 from *Aureobasidium pullulans* has been shown to inhibit spore germination and reduce germ tube length in several fungal species, including *Penicillium expansum*, *Botrytis cinerea*, *Moniliniafructicola*, and *Alternaria alternata* (Banani et al., 2014). Furthermore, protease activity has also been observed in the

genera *Metschnikowia*, *Pichia*, and *Wickerhamomyces* (Pretscher et al., 2018).



Fig 8 :Characterization of extracellular lytic enzymes produced by the yeast biocontrol agent *Candida oleophila*

3. Toxin production

Toxins are proteins, often glycosylated, that bind to specific receptors on the surface of target microorganisms, leading to their destruction through a targeted mechanism. Yeasts are generally not recognized as significant producers of secondary metabolites compared to filamentous fungi and bacteria, which may contribute to the perception that they pose fewer biosafety risks in industrial and biotechnological applications (Agirman et al., 2023). An example of a bioactive compound produced by the biocontrol yeast *Pseudozyma flocculosa* is flocculosin, a low molecular weight cellobiose lipid with the potential to inhibit plant pathogenic fungi (Teichmann et al., 2011). *Aureobasidium pullulans* synthesizes a variety of polymers, lipids, volatiles, enzymes, and secondary metabolites (Prasongsuk et al., 2018). Notable yeast strains that produce toxins against pathogens include *Saccharomyces cerevisiae* (K1, K2, K28, Klus, KHR, KHS), *Pseudozyma flocculosa* (flocculosin), and *Candida nodaensis* (PYCC 3198) (Luska et al., 2015; Teichmann et al., 2011; da Silva et al., 2008). A significant agricultural breakthrough was the use of killer yeast strains, specifically *Pichia fermentans* strains 27, 28, and 56, to control the postharvest pathogen *Penicillium digitatum* on citrus, with strain 27 demonstrating a protection efficiency of 93.6% (Perez et al., 2016).

Fig 9

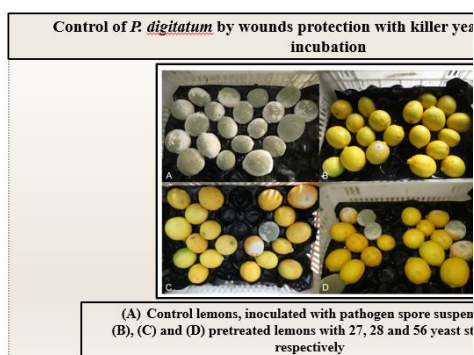
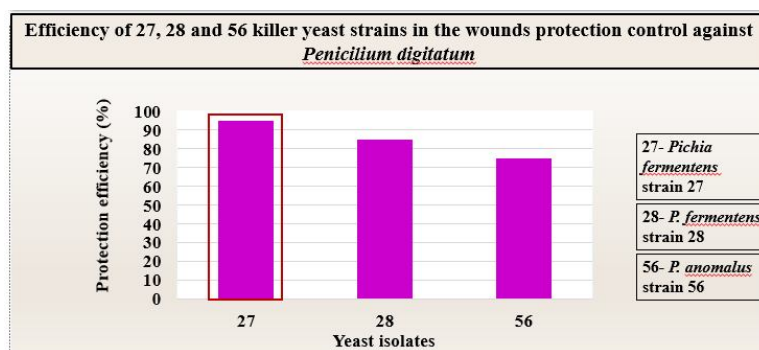


Fig:10

4. Volatile organic compounds (VOCS)

Volatile organic compounds (VOCs) are low-water-solubility compounds, typically under 300 Da, produced during the metabolism of organisms. They encompass a variety of chemical types, including hydrocarbons, alcohols, and phenols (Morath et al., 2012). *Aureobasidium pullulans* effectively inhibits several fungal pathogens like *Botrytis cinerea*, both in vitro and in planta (Di Francesco et al., 2015). The biocontrol of table grapes by food yeasts such as *Wickerhamomyces anomalus* and *Saccharomyces cerevisiae* is primarily due to VOC production (Parafati et al., 2015). Inhibition of *B. cinerea* germination has been associated with 2-ethyl-1-hexanol (Huang et al., 2012). In a 2022 study by Natarajan et al., 12 of 45 yeast isolates showed effective inhibition of *Aspergillus flavus* growth, with *S. cerevisiae* strain YKK1 achieving up to 92.1% inhibition of mycelial growth and 98.1% reduction in aflatoxin B1 production. Gas chromatography-mass spectrometry analysis identified several antimicrobial compounds, including 1-pentanol and benzothiazole, which significantly reduced fungal growth and aflatoxin synthesis.

Fig 11 :Strains of *Saccharomyces cerevisiae*

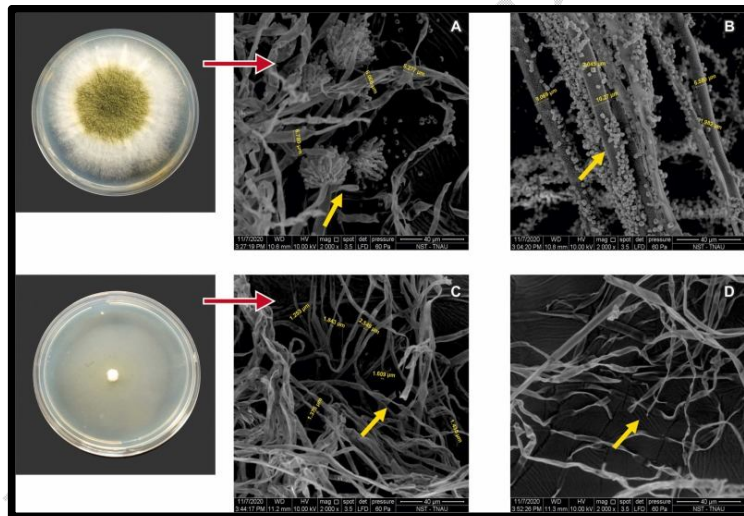
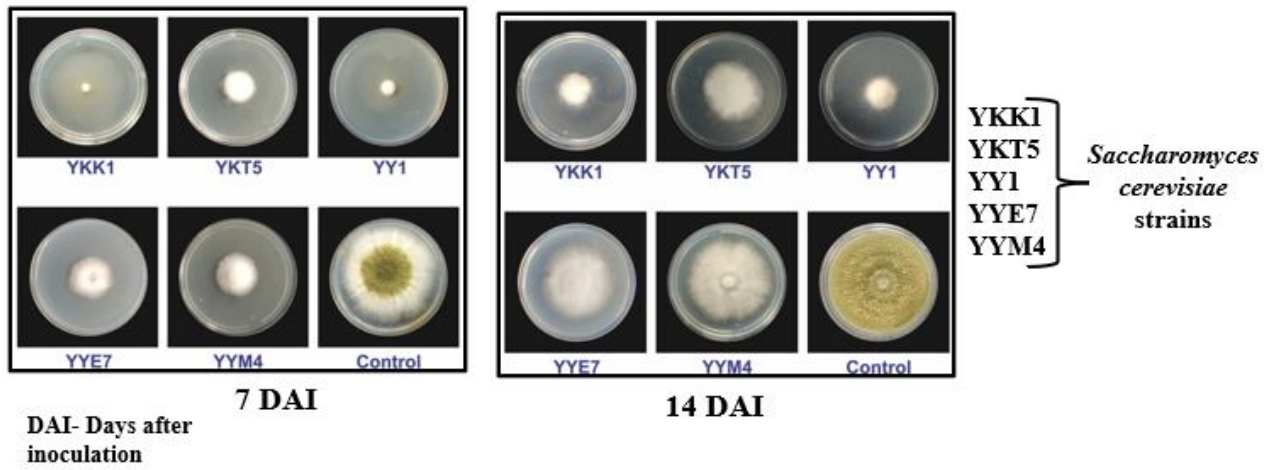


Fig 12 :Effect of VOCs on fungal morphology

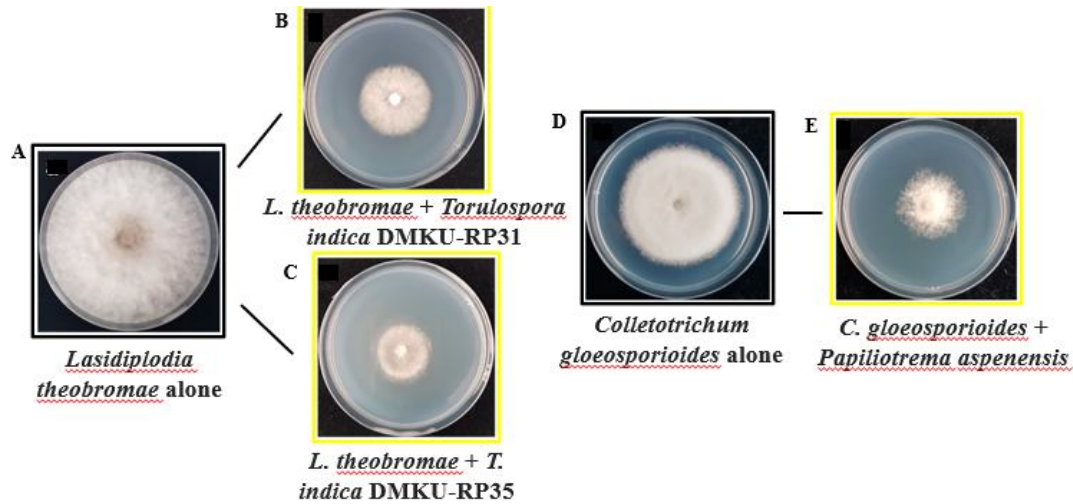
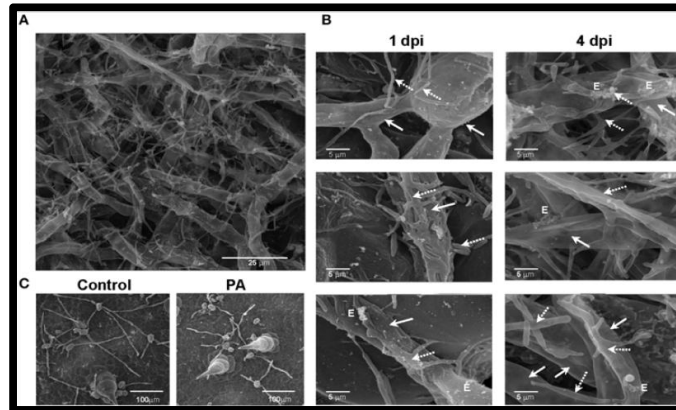


Fig:13 :Aflatoxin synthesis

5. Mycoparasitism

Mycoparasitism is a lifestyle in which one fungus engages in parasitic interactions with another. *Pichia guilliermondii* has been observed to exhibit mycoparasitic behavior towards the hyphae of the plant pathogenic fungus *Botrytis cinerea* (Wisniewski et al., 1991). *Pseudozymaaphidis* parasitizes the powdery mildew pathogen *Podosphaeraxanthii* and *B. cinerea* (Gafni et al., 2015). The genus *Saccharomycopsis* comprises predatory yeasts that directly feed on other microorganisms, including their prey, and has been studied for its biocontrol activities against various *Penicillium* species (Junker et al., 2019).

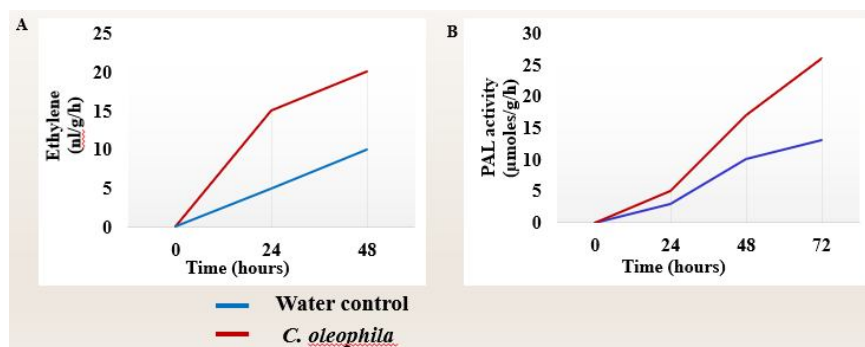
Fig:14 :Mycoparasitism



6. Induction of resistance

Biocontrol yeasts can induce systemic resistance in plants against various pathogens (Lee et al., 2017). *Saccharomyces cerevisiae*, *Rhodosporidium paludigenum*, *Candida saitoana*, *C. oleophila*, and *Metschnikowia* species trigger an innate immune response, enhancing resistance against phyllosphere pathogens in fruits. The innate immune response and systemic resistance induced by *Candida oleophila* are linked to several factors, including the overproduction of reactive oxygen species (ROS) in the plant (De Miccolis Angelini et al., 2019). Additionally, yeasts like *Candida laurentii*, *Cryptococcus flavescens*, and *Rhodotorula glutinis* can be combined with resistance inducers such as salicylic acid or rhamnolipids in integrated disease management strategies (Yan et al., 2014). The application of *Candida oleophila* cell suspensions to grapefruit peel tissue boosts ethylene biosynthesis, phenylalanine ammonia lyase activity, and phytoalexin accumulation, thereby stimulating host defense (Droby et al., 2002).

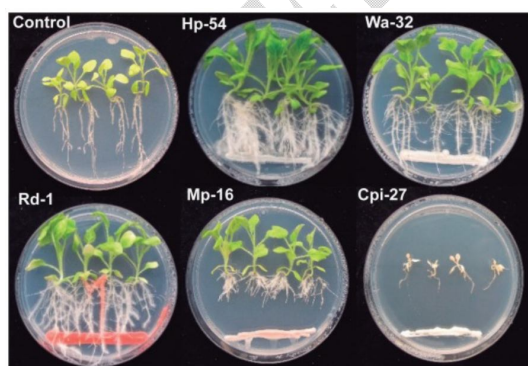
Fig:15 :Induction of resistance



Plant growth promotion by yeast

Yeasts are promising agents for promoting plant growth. In vitro studies have demonstrated that co-culturing *Nicotiana benthamiana* seedlings with specific yeast strains enhances their growth, leading to healthier and more robust development, which is crucial in the early stages of plant growth. Inoculating tobacco and lettuce seedlings with particular yeast strains also boosts their vigor, increasing the likelihood of successful establishment when transplanted, ultimately improving crop yields (Fernandez et al., 2020). Moreover, introducing yeasts such as *S. cerevisiae*, *Debaryomyces hansenii*, and *Lachancea thermotolerans* into maize-growing soil has shown to promote the vigor of maize plants, indicating their positive effect on growth and development (Fernandez-San Millan et al., 2020).

Fig:16 :Plant growth promotion by yeast (stage 1)



Hp-54 - *Hyphopichia pseudoburtonii*
 Wa-32- *Wickerhamomyces anomalus*
 Rd-1 - *Rhodotorula dairenensis*
 Mp-16- *Metschnikowia pulcherrima*
 Cpi-27- *Candida pimensis*
 Cv-15- *Candida vanderwaltii*

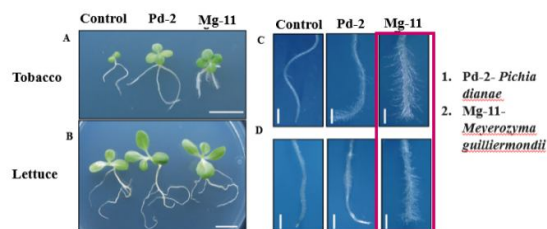


Fig 17 :Plant growth promotion by yeast(stage 2)

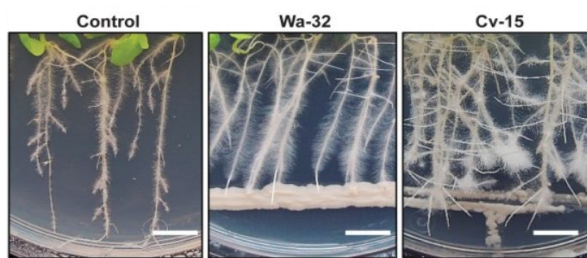


Fig:18 :Plant growth promotion by yeast(stage 3)

Registered biocontrol yeast species

a. Candida oleophila

C. oleophila was the first yeast developed as a commercial plant protection agent, leveraging its competitive advantages for nutrients and space. This marked a significant advancement in yeast-based biocontrol in agriculture. Its efficacy against plant pathogens stems from mechanisms such as producing hydrolytic enzymes, volatile compounds, biofilm formation, and inducing plant resistance (Huang et al., 2011; Wisniewski et al., 2007). Strains I-182 and O have been commercialized as Aspire and Nexy, respectively, with strain O gaining approval in Europe in 2013 (European Food Safety Authority, 2015).

b. Aureobasidium pullulans

The saprophytic fungus *A. pullulans*, found in various environments, shows biocontrol activity, particularly two registered strains: DSM 14940 (CF 10) and DSM 14941 (CF 40). These strains are effective against fireblight and postharvest diseases, formulated into the wettable powder product Blossom-Protect (EFSA, 2013). They are also registered for controlling postharvest diseases in apples (Boni-Protect) and are under study for use against storage diseases in strawberries and plums (Weiss et al., 2006; 2012).

c. Metschnikowia fructicola

Metschnikowia species, particularly *M. fructicola* and *M. pulcherrima*, are primarily found in the phyllosphere and exhibit significant biocontrol potential against postharvest diseases. Their antifungal mechanisms include nutrient competition and the secretion of glucanases and chitinases (Gore-Lloyd et al., 2019). *M. fructicola* isolate NRRL Y-30752, discovered in Israel, has been registered to prevent postharvest diseases in crops like sweet potatoes and carrots, with EFSA approval for use in stone fruits, strawberries, and grapes (Eshel et al., 2009; EFSA, 2017).

d. Saccharomyces cerevisiae

Saccharomyces cerevisiae, known for its applications in biotechnology, has demonstrated antifungal activity, especially strains like BY4741 and others that inhibit plant pathogens such as *Aspergillus* and *Fusarium* (Hilber-Bodmer et al., 2017; Armando et al., 2013). These strains employ mechanisms like secreting hydrolytic enzymes and volatile compounds. The commercial product Romeo, containing *S. cerevisiae*, is used to induce systemic resistance against downy mildew in crops (EFSA, 2015; Freimoser et al., 2019).

Commercial Applications

Several yeast-based bioproducts are currently registered, including Blossom Protect, Biotector, and BoniProtect, all containing *A. pullulans* strains for various plant diseases. Blossom Protect targets fire blight and other rots in fruits, while Biotector is effective against graymold in grapes and strawberries. BoniProtect prevents fungal diseases in orchards. Julietta is a fungicide with *S. cerevisiae* designed for strawberries and tomatoes, while Nexy, containing *C. oleophila*, is used against mold in apples and pears. Noli features *M. fructicola* for postharvest decay, and Romeo, made from *S. cerevisiae* cell walls, prevents powdery mildew in multiple crops (Kowalska et al., 2022).

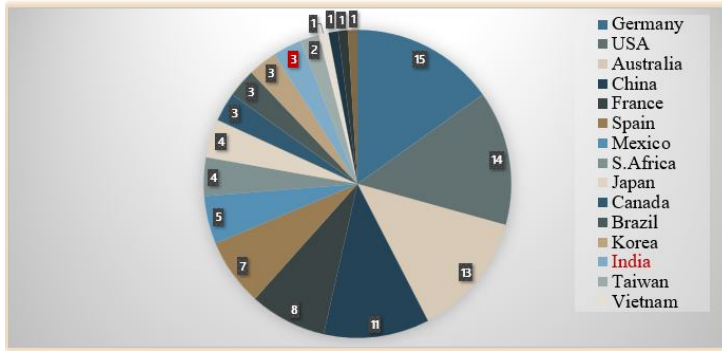


Fig:19 :Patents on Yeast-Based Products for Plant and Fruit Disease Biocontrol

Patents on Yeast-Based Products for Plant and Fruit Disease Biocontrol

Germany leads in the number of patents for yeast-based products, followed by the USA and Australia, while India holds 3% of these patents. From 2009 to 2021, the Derwent Innovation database reported 163 patents related to yeasts as biological control agents, highlighting their growing recognition in sustainable agricultural disease management. Together, Germany, the USA, Australia, and China account for 53% of all yeast-related patents worldwide. Of the 163 patents, 73.68% specified the genus or species of the yeast, while 26.31% simply noted "yeast" among the components. These products often contain multiple microorganisms, with 32.89% featuring *M. fructicola*, 11.18% containing *Candida* species, and a mix of *C. oleophila*, *M. fructicola*, and *P. anomala*. Other included yeasts are *Pichia* (9.86%), *Rhodotorula* (7.89%), and *Cryptococcus* (5.92%), either alone or with *Rhodotorula*. Only 1.97% involved *Debaryomyces* (Hernandez et al., 2021).

Fig:20 :Comparative overview



Case study

In a 2016 study by Zhimo et al., researchers investigated the biological control of post-harvest fruit diseases in India using antagonistic yeasts. They isolated 29 yeasts from various sources and identified three strains—*Candida tropicalis* YZ1 (CtYZ1), *Saccharomyces cerevisiae* YZ7 (ScYZ7), and *Candida tropicalis* YZ27 (CtYZ27)—which showed broad antagonistic activity against pathogens in vitro. The researchers applied these yeast strains at concentrations of 1 to 4×10^{-8} colony-forming units per milliliter (Cfu/ml) to artificially inoculated wounds on banana fruits with *Colletotrichum musae*. The results demonstrated that CtYZ1, ScYZ7, and CtYZ27 reduced the mean lesion diameter by 85.5%, 88.7%, and 91.9%, respectively, while a fungicide (Carbendazim at 1.0 g/l) reduced lesions by 75.8% compared to the control group. These findings indicate the effectiveness of the yeast strains in controlling fungal growth on fruit.

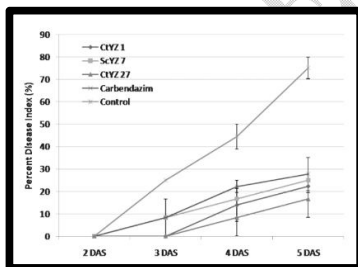


Fig 21 :Banana

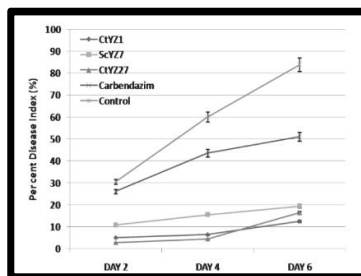


Fig 22 :Litchi

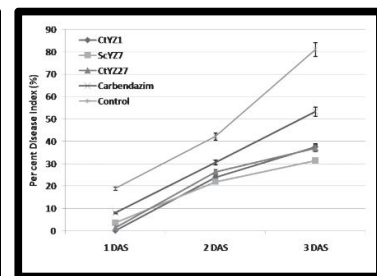


Fig 23 :S

The 2020 study by Into et al. assessed the antagonistic activities of 83 yeast strains against various rice pathogenic fungi, including *Pyricularia oryzae*, *Rhizoctonia solani*, *Fusarium moniliforme*, *Helminthosporium oryzae*, and *Curvularialunata*. Fourteen strains demonstrated inhibitory effects, with *Torulaspota indica* DMKU-RP31, DMKU-RP35, and *Wickerhamomycesanomalus* DMKU-RP25 effectively inhibiting all tested pathogens. The researchers explored mechanisms behind these antagonistic effects, including the production of volatile organic compounds (VOCs), fungal cell wall-degrading enzymes, and biofilm formation. Additionally, in greenhouse tests, these yeast strains suppressed rice sheath blight caused by *R. solani* by 60.0% to 70.3%, compared to an 83.8% suppression with 3% validamycin. These findings suggest the potential of these yeast strains as biocontrol agents for rice sheath blight.

Treatment	Lesion Height(cm)	Disease Incidence(%)
Control (negative control)	0d	0d
<i>Rhizoctonia solani</i>(positive control)	23.8 ± 1.6	25.9 ± 2.3
<i>R. solani</i> + <i>Torulaspota indica</i> DMKU□RP31	7.2 ± 1.4	7.7 ± 1.4
<i>R. solani</i> + <i>T. indica</i> DMKU□RP35	8.3 ± 1.5	8.8 ± 1.4
<i>R. solani</i> + <i>W. anomalus</i> DMKU□RP25	8.1 ± 1.0	8.7 ± 0.8
<i>R. solani</i>+ 3% Validamycin	3.9 ± 0.5	4.2 ± 0.6

Table:1 antagonistic activities

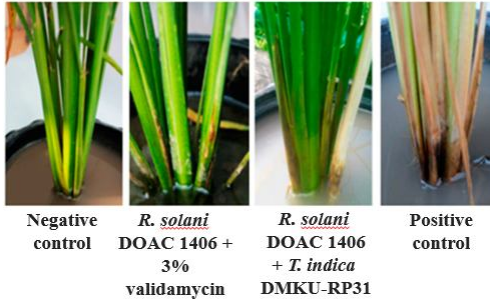


Fig 24 :Rice sheath blight disease lesions 15 days after *R. solani*DOAC 1406 inoculation

In 2017, Lee et al. studied the isolation of leaf-colonizing yeasts from pepper plants in a major pepper production area of South Korea and their potential for controlling *Xanthomonas axonopodis* infections. Using a semi-selective medium with rifampicin to inhibit bacterial growth, they isolated 838 yeast strains from the leaves. Notably, *Pseudozyma* strain RGJ1, applied as a foliar spray at a concentration of 10^{-8} cfu/ml, significantly protected pepper plants against *X. axonopodis* in greenhouse trials, highlighting its potential as a biocontrol agent against bacterial diseases. Additionally, strain RGJ1 unexpectedly conferred protection against several viruses, including Cucumber mosaic virus and Pepper mottle virus, suggesting broad-spectrum disease control capabilities. Co-culture assays showed no direct antagonism between strain RGJ1 and *X. axonopodis*, indicating that disease suppression likely results from induced resistance in the pepper plants. Molecular analyses revealed the expression of induced resistance marker genes, specifically *Capsicum annuum* Pathogenesis-Related proteins CaPR4 and CaPR5, associated with enhanced plant defense responses, suggesting that strain RGJ1 primes the plants for better pathogen resistance.

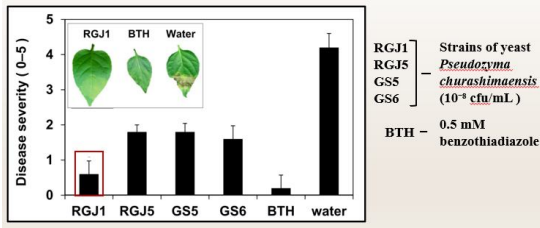


Fig 25 :Strains of yeast *Pseudozyma*

Fig:25 The development and commercialization of yeast-based plant protection

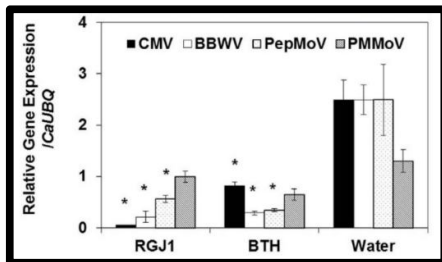
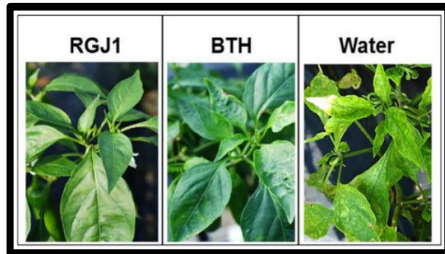


Fig:26 RGJ1 primes the plants for better pathogen resistance



Reasons for non-popularity for yeast-based plant protectants

The development and commercialization of yeast-based plant protection products face several challenges, including:

1. **Lack of Mechanistic Understanding:** Insufficient knowledge of yeast interactions with plant pathogens hinders product development.
2. **Registration Hurdles:** The complex and costly registration process for biocontrol products poses challenges for smaller companies.
3. **Collaboration Gaps:** The need for expertise from various fields often leads to difficulties in forming necessary partnerships.
4. **Limited Commercial Potential:** Yeast products may lack the appeal of conventional pesticides, making adoption difficult in the agricultural sector.
5. **Market Acceptance:** Farmers and consumers may be resistant to adopting biocontrol methods.

6. **Long Development Timelines:** Extended product development periods can deter investment and commercial interest.

Conclusion

Yeast serves as a promising biocontrol agent for sustainable pathogen management in agriculture, effectively targeting various pathogens. This versatility allows it to address multiple issues, making it particularly valuable for crops affected by different pathogens. Yeast can outperform traditional fungicides, leading to less frequent chemical applications, which conserves resources and minimizes environmental impact. As a natural alternative to chemical pesticides, yeast-based biocontrol reduces risks of soil and water contamination and protects non-target organisms. Overall, yeast-based strategies offer a resilient and sustainable method for controlling plant pathogens, avoiding pesticide resistance and long-term ecosystem harm.

Future Prospects:

1. **Understanding Modes of Action:** Improved knowledge of yeast mechanisms can lead to targeted strategies.
2. **Consortium Design:** Combining complementary yeast strains can enhance disease defence.
3. **Large-Scale Production:** Advances in yeast cultivation can increase accessibility and affordability.
4. **Industrial Collaboration:** Partnerships between research institutions and industries can boost product development.
5. **Biological Innovation:** Genetic modifications of yeasts may enhance their biocontrol efficacy for specific applications.

Reference

Agirman, B., Carsanba, E., Settanni, L., and Erten, H. 2023. Exploring yeast-based microbial interactions: The next frontier in postharvest biocontrol. *Yeast*. 5: 34-68.

- Armando, M. R., Dogi, C. A., Poloni, V., Rosa, C. A. R., Dalcerro, A. M., and Cavaglieri, L. R. 2013. *In vitro* study on the effect of *Saccharomyces cerevisiae* strains on growth and mycotoxin production by *Aspergillus carbonarius* and *Fusarium graminearum*. *Int. J. Food Microbiol.* 161(3): 182-188.
- Arroyo-López, F. N., Querol, A., Bautista-Gallego, J., and Garrido-Fernández, A. 2008. Role of yeasts in table olive production. *Int. J. Food Microbiol.* 128(2): 189-196.
- Banani, H., Spadaro, D., Zhang, D., Matic, S., Garibaldi, A., and Gullino, M. L. 2014. Biocontrol activity of an alkaline serine protease from *Aureobasidium pullulans* expressed in *Pichia pastoris* against four postharvest pathogens on apple. *Int. J. Food Microbiol.* 182: 1-8.
- Bar-Shimon, M., Yehuda, H., Cohen, L., Weiss, B., Kobeshnikov, A., Daus, A., Goldway, M., Wisniewski, M., and Droby, S. 2004. Characterization of extracellular lytic enzymes produced by the yeast biocontrol agent *Candida oleophila*. *Curr. Genet.* 45: 140-148.
- Bencheqroun, S. K., Bajji, M., Massart, S., Labhilili, M., El Jaafari, S., and Jijakli, M.H. 2007. *In vitro* and *in situ* study of postharvest apple blue mold biocontrol by *Aureobasidium pullulans*: evidence for the involvement of competition for nutrients. *Postharvest Biol. Technol.* 46(2): 128-135.
- Cavalheiro, M. and Teixeira, M. C. 2018. *Candida* biofilms: threats, challenges, and promising strategies. *Front. Med.* 5: 28.
- Chappell, C. R. and Fukami, T. 2018. Nectar yeasts: a natural microcosm for ecology. *Yeast.* 35(6): 417-423.
- Costa-Orlandi, C. B., Sardi, J. C., Pitangui, N. S., De Oliveira, H. C., Scorzoni, L., Galeane, M. C., Medina-Alarcón, K. P., Melo, W. C., Marcelino, M.Y., Braz, J. D., and Fusco-Almeida, A. M. 2017. Fungal biofilms and polymicrobial diseases. *J. Fungi.* 3(2): 22-35.
- da Silva, S., Calado, S., Lucas, C., and Aguiar, C. 2008. Unusual properties of the halotolerant yeast *Candida nodaensis* Killer toxin, CnKT. *Microbiol. Res.* 163(2): 243-251.

- De Miccolis Angelini, R. M., Rotolo, C., Gerin, D., Abate, D., Pollastro, S., and Faretra, F. 2019. Global transcriptome analysis and differentially expressed genes in grapevine after application of the yeast-derived defense inducer cerevisane. *Pest Manag. Sci.* 75(7): 2020-2033.
- Di Francesco, A., Ugolini, L., Lazzeri, L., and Mari, M. 2015. Production of volatile organic compounds by *Aureobasidium pullulans* as a potential mechanism of action against postharvest fruit pathogens. *Biol. Control.* 81: 8-14.
- Droby, S., Vinokur, V., Weiss, B., Cohen, L., Daus, A., Goldschmidt, E.E., and Porat, R. 2002. Induction of resistance to *Penicillium digitatum* in grapefruit by the yeast biocontrol agent *Candida oleophila*. *Phytopathol.* 92(4): 393-399.
- Eshel, D., Regev, R., Orenstein, J., Droby, S., and Gan-Mor, S. (2009) Combining physical, chemical and biological methods for synergistic control of postharvest diseases: a case study of black root rot of carrot. *Postharvest Biol. Technol.* 54: 48–52.
- European Food Safety Authority (EFSA). 2013. Conclusion on the peer review of the pesticide risk assessment of the active substance *Aureobasidium pullulans* (strains DSM 14940 and DSM 14941). *EFSA. J.* 11: 3183.
- European Food Safety Authority (EFSA). 2015. Conclusion on the peer review of the pesticide risk assessment of the active substance *Candida oleophila* strain O. *EFSA. J.* 10: 2944.
- European Food Safety Authority (EFSA). 2015. Peer review of the pesticide risk assessment of the active substance *Saccharomyces cerevisiae* strain LAS02. *EFSA. J.* 13(12): 4322.
- European Food Safety Authority (EFSA), Arena, M., Auteri, D., Barmaz, S., Bellisai, G., Brancato, A., Brocca, D., Bura, L., Byers, H., Chiusolo, A., and Court Marques, D. 2017. Peer review of the pesticide risk assessment of the active substance *Metschnikowia fructicola* NRRL Y-27328. *EFSA. J.* 15(12): 5084.
- Fernandez-San Millan, A., Farran, I., Larraya, L., Ancin, M., Arregui, L.M., and Veramendi, J. 2020. Plant growth-promoting traits of yeasts isolated from Spanish vineyards: Benefits for seedling development. *Microbiol. Res.* 237: 126480.

- Freimoser, F. M., Rueda-Mejia, M. P., Tilocca, B., and Migheli, Q. 2019. Biocontrol yeasts: Mechanisms and applications. *World J. Microbiol. Biotechnol.* 35: 1-19.
- Friel, D., Pessoa, N. M. G., Vandenbol, M., and Jijakli, M. H. 2007. Separate and combined disruptions of two *exo-β-1, 3-glucanase* genes decrease the efficiency of *Pichia anomala* (strain K) biocontrol against *Botrytis cinerea* on apple. *Mol. Plant-Microbe Interact.* 20(4): 371-379.
- Gafni, A., Calderon, C. E., Harris, R., Buxdorf, K., Dafa-Berger, A., Zeilinger-Reichert, E., and Levy, M., 2015. Biological control of the cucurbit powdery mildew pathogen *Podosphaera xanthii* by means of the epiphytic fungus *Pseudozyma aphidis* and parasitism as a mode of action. *Front. Plant Sci.* 6: 132-150.
- Gore-Lloyd, D., Sumann, I., Brachmann, A. O., Schneeberger, K., Ortiz-Merino, R. A., Moreno-Beltrán, M., Schläfli, M., Kirner, P., Santos Kron, A., Rueda-Mejia, M. P., and Somerville, V. 2019. Snf2 controls pulcherriminic acid biosynthesis and antifungal activity of the biocontrol yeast *Metschnikowia pulcherrima*. *Mol. Microbiol.* 112(1): 317-332.
- Hernandez-Montiel, L. G., Droby, S., Preciado-Rangel, P., Rivas-García, T., González-Estrada, R. R., Gutiérrez-Martínez, P., and Ávila-Quezada, G.D. 2021. A sustainable alternative for postharvest disease management and phytopathogens biocontrol in fruit: Antagonistic yeasts. *Plants.* 10(12): 2641.
- HersHKovitz, V., Ben-Dayán, C. L., Raphael, G., Pasmanik-Chor, M. E., Liu, J. I. A., Belausov, E., Aly, R., Wisniewski, M., and Droby, S. 2012. Global changes in gene expression of grapefruit peel tissue in response to the yeast biocontrol agent *Metschnikowia fructicola*. *Mol. Plant Pathol.* 13(4): 338-349.
- Hilber-Bodmer, M., Schmid, M., Ahrens, C. H., and Freimoser, F. M. 2017. Competition assays and physiological experiments of soil and phyllosphere yeasts identify *Candida subhashii* as a novel antagonist of filamentous fungi. *BMC Microbiol.* 17: 1-15.
- Huang, R., Che, H. J., Zhang, J., Yang, L., Jiang, D. H., and Li, G.Q. 2012. Evaluation of *Sporidiobolus pararoseus* strain YCXT3 as biocontrol agent of *Botrytis cinerea* on post-harvest strawberry fruits. *Biol. Control.* 62(1): 53-63.

- Huang, R., Li, G. Q., Zhang, J., Yang, L., Che, H. J., Jiang, D. H., and Huang, H. C. 2011. Control of postharvest Botrytis fruit rot of strawberry by volatile organic compounds of *Candida intermedia*. *Phytopathology*, 101(7): 859-869.
- Into, P., Khunnamwong, P., Jindamoragot, S., Am-In, S., Intanoo, W., and Limtong, S. 2020. Yeast associated with rice phylloplane and their contribution to control of rice sheath blight disease. *Microorganisms*. 8(3): 362.
- Jiang, Y., Ma, J., Wei, Y., Liu, Y., Zhou, Z., Huang, Y., Wang, P., and Yan, X. 2022. De novo biosynthesis of sex pheromone components of *Helicoverpa armigera* through an artificial pathway in yeast. *Green Chem.* 24(2): 767-778.
- Junker, K., Chailyan, A., Hesselbart, A., Forster, J., and Wendland, J. 2019. Multi-omics characterization of the necrotrophic mycoparasite *Saccharomycopsis schoenii*. *PLoS Pathog.* 15(5): 1007692.
- Knight, A.L. and Witzgall, P. 2013. Combining mutualistic yeast and pathogenic virus—a novel method for codling moth control. *J. Chem. Ecol.* 39: 1019-1026.
- Konsue, W., Dethoup, T., and Limtong, S. 2020. Biological control of fruit rot and anthracnose of postharvest mango by antagonistic yeasts from economic crops leaves. *Microorganisms*. 8(3): 317-381
- Kowalska, J., Krzywińska, J., and Tyburski, J. 2022. Yeasts as a potential biological agent in plant disease protection and yield improvement—A short review. *Agriculture*. 12(9):1404
- Kowalska, J., Rożdżyński, D., Remlein-Starosta, D., Sas-Paszt, L., and Malusá, E. 2012. Use of *Cryptococcus albidus* for controlling grey mould in the production and storage of organically grown strawberries. *J. Plant Dis. Prot.* 119: 174-178.
- Kunz, S., Schmitt, A., and Haug, P. 2011. Field testing of strategies for fire blight control in organic fruit growing. *Acta Hortic.* 896: 431-436
- Langner, T. and Göhre, V. 2016. Fungal chitinases: function, regulation, and potential roles in plant/pathogen interactions. *Curr. Genet.* 62: 243-254.
- Lee, G., Lee, S. H., Kim, K. M., and Ryu, C. M. 2017. Foliar application of the leaf-colonizing yeast *Pseudozyma kashimaensis* elicits systemic defense of pepper against bacterial and viral pathogens. *Sci. Rep.* 7(1): 39432.

- Lopes, M.R., Klein, M.N., Ferraz, L.P., da Silva, A.C., and Kupper, K.C. 2015. *Saccharomyces cerevisiae*: a novel and efficient biological control agent for *Colletotrichum acutatum* during pre-harvest. *Microbiol. Res.* 175: 93-99.
- Lukša, J., Podoliankaitė, M., Vepškaitė, I., Strazdaitė-Žielienė, Ž., Urbonavičius, J., and Servienė, E. 2015. Yeast β -1, 6-glucan is a primary target for the *Saccharomyces cerevisiae* K2 toxin. *Eukaryot. Cell.* 14(4): 406-414
- Lutz, M. C., Lopes, C. A., Rodriguez, M. E., Sosa, M. C., and Sangorrín, M. P. 2013. Efficacy and putative mode of action of native and commercial antagonistic yeasts against postharvest pathogens of pear. *Int. J. food Microbiol.* 164(2-3): 166-172.
- Maserti, B., Podda, A., Giorgetti, L., Del Carratore, R., Chevret, D., and Migheli, Q. 2015. Proteome changes during yeast-like and pseudohyphal growth in the biofilm-forming yeast *Pichia fermentans*. *J. Amino Acids.* 47: 1091-1106.
- Morath, S. U., Hung, R., and Bennett, J. W. 2012. Fungal volatile organic compounds: a review with emphasis on their biotechnological potential. *Fungal Biol. Rev.* 26(2-3): 73-83.
- Natarajan, S., Balachandar, D., Senthil, N., Velazhahan, R., and Paranidharan, V. 2022. Volatiles of antagonistic soil yeasts inhibit growth and aflatoxin production of *Aspergillus flavus*. *Microbiol. Res.* 263:127150.
- Ortu, G., Demontis, M.A., Budroni, M., Goyard, S., d'Enfert, C., and Migheli, Q., 2005. Study of biofilm formation in *Candida albicans* may help understanding the biocontrol capability of a flor strain of *Saccharomyces cerevisiae* against the phytopathogenic fungus *Penicillium expansum*. *J. Plant. Pathol.* 87(4):300.
- Parafati, L., Vitale, A., Restuccia, C., and Cirvilleri, G. 2015. Biocontrol ability and action mechanism of food-isolated yeast strains against *Botrytis cinerea* causing post-harvest bunch rot of table grape. *Food microbiol.* 47: 85-92.
- Perez, M. F., Contreras, L., Garnica, N. M., Fernández-Zenoff, M. V., Farías, M. E., Sepulveda, M., Ramallo, J., and Dib, J. R. 2016. Native killer yeasts as biocontrol agents of post-harvest fungal diseases in lemons. *PloS one.* 11(10): 0165590.

- Prasongsuk, S., Lotrakul, P., Ali, I., Bankeeree, W., and Punnapayak, H. 2018. The current status of *Aureobasidium pullulans* in biotechnology. *Folia Microbiologica*. 63:129-140.
- Pretscher, J., Fischkal, T., Branscheidt, S., Jäger, L., Kahl, S., Schlander, M., Thines, E., and Claus, H. 2018. Yeasts from different habitats and their potential as biocontrol agents. *Ferment*. 4(2): 31-45
- Rossouw, D., Meiring, S. P., and Bauer, F. F. 2018. Modifying *Saccharomyces cerevisiae* adhesion properties regulates yeast ecosystem dynamics. *Mosphere*. 3(5): 383- 418.
- Shalaby, M. E. S. and El-Nady, M. F. 2008. Application of *Saccharomyces cerevisiae* as a biocontrol agent against Fusarium infection of sugar beet plants. *Acta Biologica Szegediensis*. 52(2): 271-275.
- Sommer, B., Overy, D. P., Haltli, B., and Kerr, R. G. 2016. Secreted lipases from *Malassezia globosa*: recombinant expression and determination of their substrate specificities. *Microbiol*. 162(7): 1069-1079.
- Spadaro, D. and Droby, S. 2016. Development of biocontrol products for postharvest diseases of fruit: The importance of elucidating the mechanisms of action of yeast antagonists. *Trends Food Sci. Technol*. 47: 39- 49.
- Teichmann, B., Labbé, C., Lefebvre, F., Bölker, M., Linne, U., and Bélanger, R.R. 2011. Identification of a biosynthesis gene cluster for flocculosin a cellobiose lipid produced by the biocontrol agent *Pseudozyma flocculosa*. *Mol. Microbiol*. 79(6): 1483-1495.
- Wang, W. L., Chi, Z. M., Chi, Z., Li, J., and Wang, X. H. 2009. Siderophore production by the marine-derived *Aureobasidium pullulans* and its antimicrobial activity. *Biores. Technol*. 100(9): 2639-2641.
- Weiss, A., Mögel, G., and Kunz, S. 2006. Development of " Boni-Protect"-a yeast preparation for use in the control of postharvest diseases of apples. In: *Ecofruit-12th International Conference on Cultivation Technique and Phytopathological Problems in Organic Fruit-Growing: Proceedings to the Conference*; 31 January to 2 February, 2006, Weinsberg/Germany, p.113-117.
- Weiss, A., Weißhaupt, S., Krawiec, P., and Kunz, S. 2012, June. Use of *Aureobasidium pullulans* for resistance management in chemical control of *Botrytis cinerea* in berries.

In: *X International Symposium on Vaccinium and Other Superfruits 1017*, 5-6 June 2012, Germany, p. 237-242.

Wisniewski, M., Biles, C., Droby, S., McLaughlin, R., Wilson, C., and Chalutz, E. 1991. Mode of action of the postharvest biocontrol yeast, *Pichia guilliermondii*. I. Characterization of attachment to *Botrytis cinerea*. *Physiol. Mol. Plant Patho.* 39(4): 245-258.

Wisniewski, M., Wilson, C., Droby, S. A., Chalutz, E., El-Ghaouth, A. H., and Stevens, C.L. 2007. Postharvest biocontrol: new concepts and applications. *Biol. control.* 6: 262-273.

Xu, H., Nobile, C. J., and Dongari-Bagtzoglou, A. 2013. Glucanase induces filamentation of the fungal pathogen *Candida albicans*. *PloS one.* 8(5): 63736.

Yan, F., Xu, S., Chen, Y., and Zheng, X. 2014. Effect of rhamnolipids on *Rhodotorulaglutinis* biocontrol of *Alternaria alternata* infection in cherry tomato fruit. *Postharvest Biol. Technol.* 97: 32-35.

Zajc, J., Gostinčar, C., Černoša, A., and Gunde-Cimerman, N. 2019. Stress-tolerant yeasts: opportunistic pathogenicity versus biocontrol potential. *Genes.* 10(1): 42- 60.

Zhang, H., Zheng, X., and Yu, T. 2007. Biological control of postharvest diseases of peach with *Cryptococcus laurentii*. *Food control.* 18(4): 287-291.

Zhimo, V.Y., Bhutia, D.D., and Saha, J. 2016. Biological control of post-harvest fruit diseases using antagonistic yeasts in India. *J. Plant Pathol.* 5: 275-283.