



## Editorial

# Towards a novel bioreactor technology for treating fungicide-rich wastewaters from the fruit packaging industry

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### Abstract

The fruit processing industry is an important economic sector worldwide, whereas a wide range of plant pathogens affects the shelf life of fruits, resulting in great economic losses. Post-harvest fungicides are often applied in the fruit processing industry to address post-harvest infections. However, various fungicides have been reported to exhibit toxicity to aquatic and terrestrial non-target organisms. Thus, effective technological solutions are needed to treat fungicide-containing wastewaters generated by the fruit packaging industry. Regarding biological methods, few approaches have been explored to deplete high-strength fungicide-based wastewaters derived from the post-harvest treatment of fruits. Despite the application of post-harvest fungicides for several decades, we are still missing the implementation of a full-scale biobased system to treat fungicide-rich wastewater generated by the fruit-packaging industry. On the other hand, fixed and fluidized bed bioreactors compared to suspended solids systems result in higher removal efficiencies. Based on the advantages of such bioreactor technology and the recent findings regarding the depuration of post-harvest fungicides in these biosystems, immobilized cell bioreactors appear to be a promising biological approach to treat persistent fungicides present in fruit packaging wastewater. However, bioreactors technology scaling up for the biotreatment of fungicide-rich wastewaters from the fruit packaging industry is challenging. Towards this direction, “Minotaur”, a 3-year research project financed by EU and Greek funds, is on the way to develop a full-scale bioreactor technology for treating fungicide-based wastewaters.

**Keywords:** fruit processing industry, post-harvest fungicides, imazalil, fludioxonil, immobilized cell bioreactor, microbial inocula

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## Editorial

The fruit processing industry is an important economic sector worldwide since the annual production of fruits in 2019 exceeded 900 million tons globally. However, several plant diseases affect the shelf life of fruits, resulting in significant economic losses. Processing fruits as a canning product can be an alternative, leading to lower profit. Post-harvest losses are mainly attributed to fungal pathogens, which deteriorate fruits quality and reduce their market value. Common fungal pathogens causing fruit infections are *Alternaria*, *Botrytis*, *Fusarium*, *Phoma* and *Rhizoctonia*, while crops are also highly affected by molds like

*Penicillium*. An issue of concern raised from the growth of fungi during storage is the secretion of mycotoxins, which increases the potential health risk to humans and animals. Despite the low pH of the fruits, bacterial contamination caused by plant pathogens like *Erwinia* and *Pseudomonas* should not be excluded.

Preharvest factors like moisture, temperature, cultivar, and cultivation practices influence the degree of fruit infection. To address the post-harvest infection, various prevention approaches have been adopted by the fruit processing industry, including pesticides, control of post-

harvest environmental parameters, increased hygiene status during handling, packaging and storage, prevention of injuries and application of thermal and radiation treatments. Post-harvest fungicide application constitutes the most common method used in the fruit processing industry to prevent crop decay. Post-harvest fungicides exhibit various modes of action, including enzyme inactivation, metabolic pathway interference, cell membrane, and respiratory chain deterioration. Other recently developed fungicides can elicit plant response by introducing systemic acquired resistance (**Schirra et al., 2011**).

Commonly used post-harvest fungicides, like imazalil, thiabendazole and ortho-phenylphenol, and non-systemic fungicides, like fludioxonil, are applied to prevent post-harvest infection of fruits caused by a wide range of phytopathogenic fungi (**Kanetis et al., 2008**). Imazalil and thiabendazole are widely used to prevent post-harvest decay of apples, pears, pineapples, bananas, citrus fruits, and potato seeds, while ortho-phenylphenol is mostly used to control fungal infestations in citrus fruits. Fludioxonil is an effective alternative to control post-harvest diseases of pomes, cherries, kiwis, mangoes, citrus, nectarines, and peaches. These fungicides are known to exhibit toxicity to aquatic and terrestrial non-target organisms. **Pennati et al. 2006**, reported that imazalil substantially impacted the early development of ascidian *Phallusia mammillata*. **Velki et al. 2019**, recently reported that fungicide mixtures containing thiabendazole or fludioxonil, significantly affected the earthworm *Eisenia andrei*. Thus, effective technological solutions are needed to treat fungicide-containing wastewaters extensively generated by the fruit packaging industry.

Physicochemical approaches have been tested at a laboratory scale to treat wastewaters containing post-harvest fungicides. Chemical oxidation methods, including advanced oxidation processes, have been employed to dissipate post-harvest fungicides. For instance, Fenton approaches have been applied for imazalil treatment, while photocatalysts, such as TiO<sub>2</sub>, have gained ground in the chemical oxidation of the non-biodegradable fungicides (**Santiago et al., 2018**). Moreover, adsorbent materials like (organo) montmorillonites have been recently used to remove thiophanate-methyl from wastewaters produced by the fruit packaging industry (**Flores et al., 2020**).

A few approaches have been explored regarding biological methods for depressing high-strength fungicide-based wastewaters derived from the post-harvest treatment of fruits. Biobed systems have been employed to treat post-harvest fungicides imazalil and thiabendazole (**Karas et al., 2016**). **Vischetti et al. 2012**, employed a biobed-based approach to treat a mixture of ten fungicides, including fludioxonil, reporting dissipation of fungicides within three months. **López-Loveira et al. 2017**, used a combined chemical-biological approach,

where photo-Fenton was applied as pretreatment, followed by the biological treatment of the Fenton-derived effluent using an imazalil-resistant consortium. **Sánchez Pérez et al. 2014**, treated simulated thiabendazole-based wastewater in a membrane bioreactor (MBR), followed by oxidation of MBR's effluent discharge through Fenton oxidation. Moreover, biodegradation of the fungicide pentachlorophenol by the basidiomycetous fungus *Phanerochaete chrysosporium* was comparatively examined in suspended, fixed-biofilm and fluidized bed bioreactors (**Kang and Stevens, 1994**). Recently **Mavriou et al. 2021** effectively treated fludioxonil in an up-flow immobilized cell bioreactor, resulting in fludioxonil removal efficiency greater than 95%.

Despite the application of post-harvest fungicides for several decades, we are still missing the implementation of a full-scale biobased system to treat fungicide-rich wastewater generated by the fruit-packaging industry. On the other hand, fixed and fluidized bed bioreactors compared to suspended solids systems result in higher cell densities and pollutant removal rates while retaining microbial cells in the immobilization carriers, avoiding biomass washout. Based on the advantages of such bioreactor technology and the recent findings regarding the depuration of post-harvest fungicides in these biosystems, immobilized cell bioreactors appear to be a promising biological approach to treat persistent fungicides present in fruit packaging wastewater.

However, bioreactors technology scale-up considering the depuration of fungicide-based wastewaters from the fruit packaging industry is challenging since no previous knowledge exists in the international literature. Towards this direction, "Minotaur", a 3-year research project financed by EU and Greek funds, aims to develop a full-scale bioreactor technology for treating fungicide-based wastewaters. The implementation of this project will optimize the operation of a full-scale fixed-bed reactor installed in a fruit packaging industry located in central Greece to provide a reliable solution for the depuration of high-strength fungicide-containing wastewaters.

Considering the recalcitrance of most of the fungicides contained in effluents and the limited dissipation capacity of generic biological treatment systems like urban wastewater treatment plants against these pesticides (**Masia et al., 2013**), the bioreactor system proposed will be inoculated with tailored-made inocula specializing in the degradation of thiabendazole, imazalil, fludioxonil and ortho-phenylphenol. Such inocula are already available through previous studies by the Minotaur research network (**Perruchon et al., 2016 ; 2017**). At this stage, the transfer of "Minotaur" technology developed on the laboratory and pilot-scale experiments to an integrated full-scale technological solution is ongoing for providing an effective method for the depuration of fungicide-based wastewaters.

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