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RESEARCH PAPER

Application of chlorine dioxide (ClO_2) and marine yeasts to control postharvest anthracnose disease in mango (*Mangifera indica* L.)

Juan J. Reyes-Perez[1], Silvana Vero[2], Ernesto Diaz-Rivera[3], Liliana Lara-Capistran[4], Juan C. Noa-Carrazana[5], and Luis G. Hernandez-Montiel[3]

[1]Universidad Técnica Estatal de Quevedo. Av. Walter Andrade. Km 1.5 vía a Santo Domingo, Quevedo, Los Ríos, Ecuador.

[2]Universidad de la Republica. Gral. Flores 2124, Montevideo, Uruguay.

[3]Centro de Investigaciones Biológicas del Noroeste. Calle Instituto Politécnico Nacional 195, Playa Palo de Santa Rita Sur, La Paz, Baja California Sur, México.

[4]Universidad Veracruzana, Facultad de Ciencias Agrícolas. Circuito Universitario Gonzalo Aguirre Beltrán s/n, Zona Universitaria, Xalapa, Veracruz, México.

[5]Universidad Veracruzana, Instituto de Biotecnología y Ecología Aplicada. Zona Universitaria, Xalapa, Veracruz, México.

Abstract

J.J. Reyes-Perez, S. Vero, E. Diaz-Rivera, L. Lara-Capistran, J.C. Noa-Carrazana, and L.G. Hernandez-Montiel. 2019. Application of chlorine dioxide (ClO_2) and marine yeasts to control postharvest anthracnose disease in mango (*Mangifera indica* L.). Cien. Inv. Agr. 46(3): 266-275. Postharvest diseases in fruits cause serious losses of fresh produce worldwide. The application of synthetic fungicides for the control of postharvest diseases such as anthracnose in mangoes can cause adverse effects on the environment and on human and animal health and has generated phytopathogen resistance. Biological control with the application of marine yeasts and chlorine dioxide (ClO_2) to reduce the use of synthetic fungicides can be an alternative to prevent anthracnose in Ataulfo mango fruits. The results showed that different doses of ClO_2 inhibited the mycelium growth and spore germination of *Colletotrichum gloeosporioides in vitro.* When ClO_2 and the marine yeasts *Debaryomyces hansenii* and *Rhodotorula minuta* were applied to mango fruits, no signs of anthracnose disease incidence and lesion diameter were observed (LSD, $p<0.05$). Therefore, the application of ClO_2 plus antagonist yeasts provides excellent control of anthracnose disease in Ataulfo mango fruits.

Key words: *Colletotrichum gloeosporioides*, *Debaryomyces hansenii*, disinfectant, mango fruit, *Rhodotorula minuta*.

Introduction

The diseases caused by fungi in postharvest fruit cause economic losses during harvest, transport and commercialization (Mahunu *et al*., 2016; Tian *et al*., 2018). Mango (*Mangifera indica* L.) is an example of an agricultural product whose postharvest life is limited by the damage caused by plant pathogens, of which *Colletotrichum gloeosporioides*, the causal agent of anthracnose, is one of the main pathogens affecting production worldwide (Snowdon, 2010; Zhang *et al*., 2013). Losses are directly caused by this pathogen in the field and during processing, reducing quality and

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Corresponding author: lhernandez@cibnor.mx

commercialization (de Oliveira *et al*., 2017). The use of synthetic fungicides is among the main methods for controlling plant diseases, but their application has generated concerns about toxicity problems and environmental damage (Mailly *et al*., 2017). Moreover, there have been reports indicating that the control efficacy of fungicides has dwindled due to the appearance of resistant phytopathogen strains (Onyeani and Osunlaja, 2012; Esteriol *et al.,* 2017).

Among the different methods that have been used as alternatives to decrease synthetic fungicide applications, the use of antagonist microorganisms and the application of disinfectants such as chloride dioxide (ClO_2) stand out, as they have demonstrated a potential for plant disease control (Liu *et al*., 2013; Lee *et al.,* 2019).

Different microbial antagonists, including some yeast species isolated from fruits, plants or soil surfaces and/or lesions, have decreased several postharvest diseases (Sharma *et al*., 2009). Although the yeasts isolated from different land ecosystems have shown a capacity for phytopathogen control, the search for biological control agents continues. Different yeasts isolated from the marine ecosystem have been shown to have a high potential for phytopathogen antagonism (Rivas-Garcia *et al.,* 2019). The application of several marine yeast species has reduced disease presence in different crops (Di Francesco *et al*., 2016; Hernandez-Montiel *et al.,* 2017).

On the other hand, postharvest fruit disinfection with sanitizing agents plays an important role in conservation and shelf life. Traditionally, sodium hypochlorite (NaClO) has been applied as a fruit treatment; nonetheless, different studies have shown that trihalomethanes (THMs), which have carcinogenic effects, may be generated from the use of NaClO (Villanueva *et al*., 2003; Villanueva *et al*., 2017). Among the disinfectants that do not generate harmful subproducts, ClO_2, whose residual properties are limited, seems to be an alternative to NaClO (Arango *et al*., 2016) due to its oxidation capacity and wide antimicrobial spectrum (Wang *et al*., 2014; Sang-Hyun and Dong-Hyun, 2015). ClO_2 is capable of reducing phytopathogen populations that compromise food safety because of its high efficiency against the fungi and bacteria generally found on crops, such as pepper, tomato, grape, strawberry, among others (Trinetta *et al*., 2013; Sun *et al*., 2017a). Likewise, ClO_2 is a reliable and safe disinfectant for human and animal health (Calvo *et al.,* 2019).

The application of antagonistic microorganisms with other management strategies could be an alternative to achieve improved control of the diseases caused by fungi in fruits, minimizing the application of synthetic fungicides. Thus, the objective of this study was to determine the postharvest application efficiency of chlorine dioxide (ClO_2) and marine yeasts on Ataulfo mango fruits protection against anthracnose caused by *C. gloeosporioides.*

Materials and methods

C. gloeosporioides

The phytopathogenic strain used in this study belongs to the fungal culture collection of the Laboratory of Phytopathology of Centro de Investigaciones Biológicas del Noroeste (CIBNOR). This strain was previously isolated from Ataulfo mango fruits with anthracnose symptoms from a commercial orchard located in El Carrizal, Baja California Sur, México, and its pathogenicity was determined (Hernandez-Montiel *et al.,* 2017). The phytopathogenic fungus was activated in potato dextrose agar (PDA) and incubated at 28 °C for 7 days. Conidial suspensions were prepared from those cultures, and the concentration was adjusted to 1×10^5 conidia mL^{-1} using a hemocytometer.

Marine yeasts

The marine yeasts were obtained from the CIBNOR collection of microbial antagonists. Two

yeast strains identified as *Debaryomyces hansenii* (1R11CB) and *Rhodotorula minuta* (1R4CF) and evaluated as agents of biocontrol were used (Hernandez-Montiel *et al.,* 2017; Rivas-Garcia *et al.,* 2019). Yeast inocula were prepared from a one–day–old potato dextrose broth (PDB) culture that was in an orbital shaker at 28 °C at 130 rpm. The yeast concentration of each inoculum was adjusted to 1×10^4, 1×10^6 and 1×10^8 cells mL^{-1} with a hemocytometer.

Chlorine dioxide (ClO_2)

A biocide solution of the commercial product Dioxival® (manufactured by Suministros AZ, La Paz, Baja California Sur, México) that reports a concentration of 231.66 mg L^{-1} of ClO_2 was prepared following the manufacturer's instructions in concentrations of 1 mg L^{-1}, 3 mg L^{-1} and 5 mg L^{-1} with sterile distilled water.

In vitro inhibition of the mycelial growth of C. gloeosporioides by ClO_2

An aliquot (100 µL) from the solution of *C. gloeosporioides* was grown in Petri dishes containing PDA. Immediately afterwards, sterilized Whatman filter paper No. 1 discs (8 mm diameter) in which 30 µL of different ClO_2 solutions (1 mg L^{-1}, 3 mg L^{-1} and 5 mg L^{-1}) were previously absorbed were placed in the center of each Petri dish. The control was prepared in the same way, but with discs containing sterile distilled water (SDW). An additional treatment using discs inoculated with 30 µL of 2% NaClO was also included in the study. Petri dishes were incubated at 28 °C for 7 days. At the end of the experiment, *C. gloeosporioides* mycelium growth diameter was quantified (mm), and the reduction was calculated with the following equation (Soylu *et al.,* 2006): I (%) = DC-DT / DC × 100, where I% = mycelium growth inhibition in percentage, DC = mycelium measured in the SDW treatment and DT = mycelium diameter in the presence of disinfectant. Fifteen replicates were performed per treatment, and the experiment was carried out twice.

In vitro *inhibition of the spore germination of* C. gloeosporioides *by ClO_2*

A volume (500 µL) of a spore suspension of *C. gloeosporioides* in sterile distilled water adjusted to 1×10^5 spores mL^{-1} was placed in Eppendorf tubes. To each tube, 30 µL of a ClO_2 solution of different concentrations (1 mg L^{-1}, 3 mg L^{-1} and 5 mg L^{-1}) was added. In another set of tubes, the same volume of 2% NaClO was incorporated, and tubes without disinfectant were used as a control. All tubes were incubated at 28 °C. After 2, 4, 6, 8 and 10 h, 100 µL aliquots were collected to observe and quantify the germination of 100 spores. A spore was considered germinated when the germ tube size was equal to or greater than the spore diameter (Yao *et al.,* 2004). Five replicates were performed per treatment, and the experiment was carried out twice.

Application of ClO_2 and marine yeasts for the control of mango anthracnose

Mango fruits were washed with sterile distilled water. Three equidistant 3 mm in width wounds were established with a sterile scalpel in the equator zone of each fruit and were disinfected with 3 mg L^{-1} of ClO_2 (limit allowed by Food and Drug Administration, 2017). The fruits disinfected with ClO_2 were dried for 2 h, and each wound was inoculated with 15 µL of each marine yeast suspension at three different concentrations (1×10^4, 1×10^6 and 1×10^8 cells mL^{-1}) and subsequently with 20 µL of a suspension of *C. gloeosporioides* adjusted to 1×10^5 spores mL^{-1}. One batch of fruit was treated with a solution of ClO_2 (3 mg L^{-1}) or with NaClO (2%) and then inoculated with the phytopathogen as described above. Another batch was used as a control and was only inoculated with *C. gloeosporioides*. The

fruit was stored in sterile plastic containers at 28 °C for 7 days. Disease incidence (%) and lesion diameter (cm) were quantified. Ten replicates were performed with three mangoes per repetition, and the experiment was performed twice.

Wound site colonization

At the end of the experiment, tissue samples containing the whole wound on fruits were removed with a sterile scalpel and placed in falcon tubes with 5 mL of phosphate buffer and 0.06% (v/v) of Tween 20. The samples were agitated at 200 rpm for 20 min. Subsequently, serial dilutions were performed with 0.85% saline solution, and 1 mL of each dilution was inoculated in PDA medium containing 100 ppm chloramphenicol, 50 ppm ampicillin and 2 ppm fluconazole (Benbow and Sugar, 1999; Parafati *et al*., 2015). Petri dishes were incubated at 28 °C for 48 h. The quantified yeast population was expressed in colony forming units (CFU/per wound); ten repetitions per treatment were performed.

Scanning electron microscopy

Samples from wounds of mango treated with ClO_2, marine yeasts and *C. gloeosporioides* were collected and fixed by immersion in 2.5% glutaraldehyde dissolved in 0.1 M of phosphate buffer at pH 7.0 for 24 h. Afterwards, the samples were partially dehydrated by means of an ethanol gradient (30, 50, 70, 80, 95 and 100%) for 20 min. They were dried at a critical point with CO_2 and later coated with gold using a coating bath (Bozzola and Russell, 1999). The samples were examined with scanning electron microscope (Hitachi®, S-3000N).

Statistical analysis

Data were processed by a one-way variance analysis (ANOVA) using the statistical package STADISTICA (StatSoft, Tulsa, OK). Prior to ANOVA, percentages were arcsine–square–root transformed. For separation of means, Fisher's least significant difference (LSD) test was used with a significance level of 5% ($P < 0.05$).

Results and discussion

In vitro inhibition of C. gloeosporioides by ClO_2

The mycelium growth of *C. gloeosporioides* in PDA medium decreased significantly as ClO_2 doses increased in the *in vitro* assay. With 3 mg L^{-1} and 5 mg L^{-1} ClO_2, the phytopathogenic fungus was inhibited by 68% and 77%, respectively, and NaClO induced 78% inhibition (Fig. 1). When spores of the phytopathogenic fungus were suspended in water, 38% of them germinated after 6 h, while 87% germination was achieved after 10 h. In the presence of ClO_2 at all tested concentrations (1 mg L^{-1}, 3 mg L^{-1} and 5 mg L^{-1}), no germinated spores were observed at any time (2, 4, 6, 8 and 10 h). The results showed that *in vitro* ClO_2 reduced mycelium growth and inhibited the spore germination of *C. gloeosporioides*.

Our results align with many previous *in vitro* studies that have shown that ClO_2 decreases the growth rate and spore germination of fungi, such as *Alternaria alternata, Fusarium oxysporum, Dothiorella gregaria, Fusarium tricinctum, Phytophthora cinnamomi, Pythium aphanidermatum,* and *Fusarium sulphureum,* among others (Scarlett *et al*., 2016; Mei *et al*., 2017; Sun *et al*., 2017b). ClO_2 directly alters the cellular membrane of fungi, releasing sugar, proteins, DNA, and ions and causing irreversible damage to the fungal cellular membrane (Sun *et al*., 2017a; Wen *et al*., 2017). ClO_2 also behaves as a free radical that eliminates the hydrogen atom of the lipid molecules contained within the fungal cellular membrane (Sharma *et al*., 2012), increasing the damage to the membrane (Zhu *et al*., 2013).

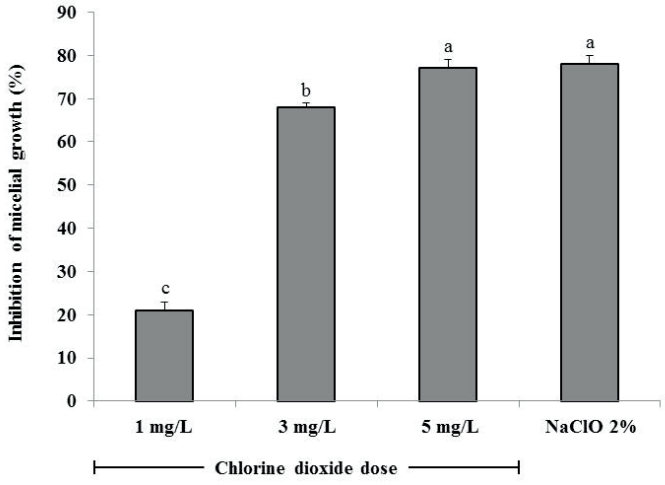

Figure 1. *In vitro* effect of chlorine dioxide (ClO_2) dose on the mycelial growth of *C. gloeosporioides*. Different letters indicate significant differences detected by ANOVA followed by LSD Fisher post hoc test ($p<0.05$).

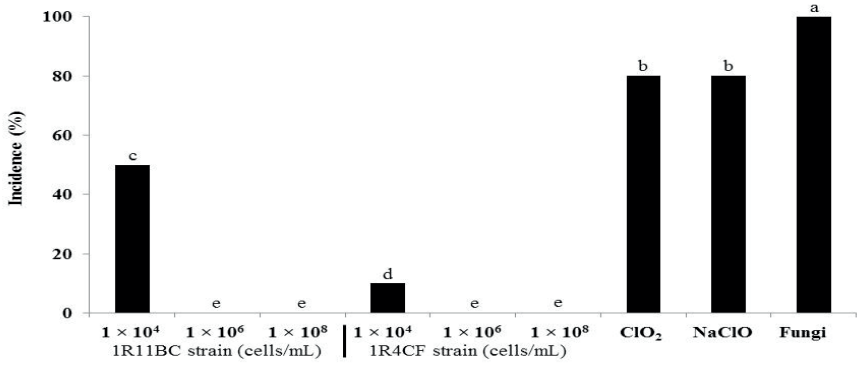

Figure 2. Application of ClO_2 and marine yeasts to control mango anthracnose caused by *C. gloeosporioides*. Before inoculation with marine yeasts, the fruits were disinfected with ClO_2 (3 mg/L). Fungi = *C. gloeosporioides*. Different letters indicate significant differences detected by ANOVA followed by the LSD Fisher post hoc test ($p<0.05$).

Disinfection and bioprotection of Ataulfo mango fruits

The incidence of anthracnose caused by *C. gloeosporioides* in mango fruits decreased significantly as the cellular dose of each of the yeasts applied to fruit increased (Fig. 2). The treatments where the disinfectant ClO_2 was applied with a dose of 1×10^6 and 1×10^8 cells mL^{-1} of both yeasts (*D. hansenii*, strain 1R11CB, and *R. minuta*, strain 1R4CF) did not show disease incidence. Compared with the control condition, fruit disinfection with ClO_2 or NaClO significantly decreased the presence of anthracnose. The application of marine yeasts after treatment with ClO_2 significantly reduced the presence of anthracnose on mangoes (Fig. 3). A large number of *D. hansenii* and *R. minuta* cells were observed to adhere to the mycelium of *C. gloeosporioides*, limiting their growth and decreasing anthracnose incidence and lesion diameter in Ataulfo mango fruits.

In the trial on mango fruits, the addition of marine yeasts after treatment with ClO_2 improved phytopathogen control principally through the biocide activity of ClO_2 (Meireles *et al*., 2017) and the diverse antagonist mechanisms exerted

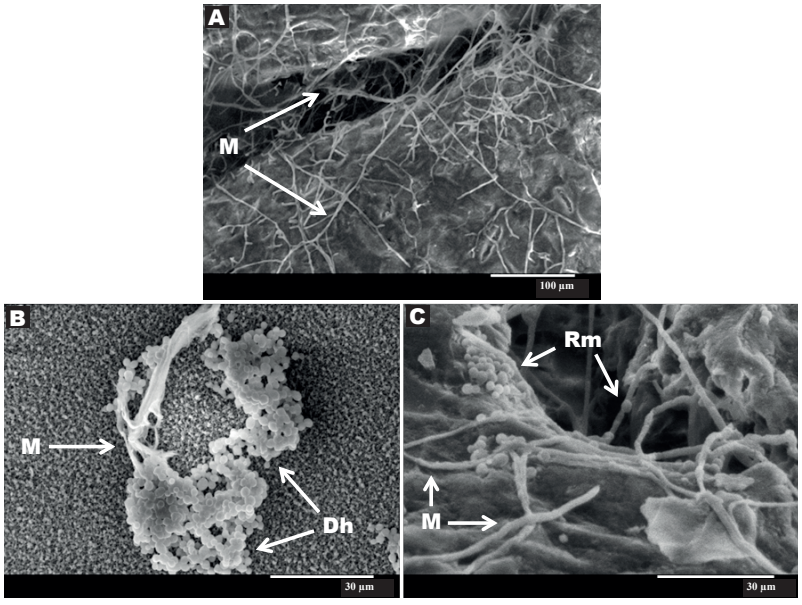

Figure 3. Micrographs of Ataulfo mango fruits disinfected with ClO_2 and inoculated with marine yeasts and *C. gloeosporioides*. [A] Mycelium (M) of *C. gloeosporioides* on a fruit wound disinfected with ClO_2. [B] Cells of *Debaryomyces hansenii* (Dh) and [C] *Rhodotorula minuta* (Rm) around the phytopathogen mycelium.

by yeasts against phytopathogens such as space and nutrient competence, lytic enzymes, parasitism, resistance induction, *killer* toxins, volatile organic compounds (VOCs), and biofilms (Di Francesco *et al*., 2016; Arrarte *et al*., 2017; Chen and Chou, 2017; Hernandez-Montiel *et al*., 2018). Marine yeasts have shown resistance to disinfecting agents, such as ClO_2, due to their ability to grow under conditions of abiotic stress (i.e., extreme pH, nutrient and oxygen limitation, and high saline concentration, among others) (Ochoa *et al.,* 1995; Ramírez-Orozco *et al.,* 2001). This study is the first report on the efficiency of the application of ClO_2 and marine yeasts *D. hansenii* and *R. minuta* on Ataulfo mango fruits to control the anthracnose caused by *C. gloeosporioides*.

On the other hand, ClO_2 and NaClO are widely used to disinfect equipment, tools and surfaces in packing plants, as well as for the postharvest disinfection of fruits and vegetables (Shinde *et al*., 2017; Calvo *et al*., 2019). In this study, Ataulfo mango fruits disinfected with ClO_2 or NaClO developed anthracnose caused by *C. gloeosporioides*. Kreske *et al*. (2006) and Zou and Wang (2017) reported that the greatest antimicrobial effect of chemical disinfectants is at the beginning of disinfection because they lose effectiveness due to the type of product and chemical composition, type of pathogen present in the host, type of cell, location of pathogens on the surface, among others.

Colonization of mango fruit

The data describing the colonization of *D. hansenii* and *R. minuta* in wounds on mangoes showed that the populations of both marine yeasts increased inside the fruit wounds (Fig. 4). The final colonization of *R. minuta* was significantly increased when inoculated initially with 1×10^6 and 1×10^8 cells mL^{-1} on fruit wounds. The results showed that both marine yeasts efficiently colonized host wounds. In this study, a positive relationship was observed between the application dose of each of the yeasts and the colonization of the host wounds and the antagonist activities of *D. hansenii* and *R. minuta* against *C. gloeosporioides* in mango fruit.

In our current study, a positive relationship between the concentration of the inoculum added to wound and antagonistic activity towards *C. gloeosporioides* was observed for the *in vivo* trials with marine yeasts. Previous studies have reported the existence of a positive relationship between high population density of antagonist microorganisms on fruit wounds and the efficiency of the biocontrol activity of microorganisms in postharvest diseases (Grzegorczyk *et al*., 2017). Moreover, a larger number of cells in the wound benefits antagonists when they increase their capacity to compete for space and nutrients, produce lytic enzymes, and inhibit other cells by VOCs and biofilms (Klein and Kupper, 2018).

The application dose of any antagonist against fruit phytopathogens could play an important role in disease development; therefore, knowing the minimum dose of an antagonist to protect the host would allow for improved control, efficiency and phytopathogen management (Mahunu *et al*., 2016).

Conclusions

Our study shows that the application of chlorine dioxide (ClO_2) and the marine yeasts *D. hansenii* and *R. minuta* on Ataulfo mango fruits enhanced the control of anthracnose caused by *C. gloeosporioides.* The mycelium growth of *C. gloeosporioides* was inhibited by ClO_2 treatment in the *in vitro* assay. Treatment with ClO_2 and medium- and high-density inocula of marine yeast significantly reduced the disease incidence and lesion diameter on mangoes fruits. Future studies will address the application of marine yeasts on Ataulfo mango fruits for the biocontrol of *C. gloeosporioides* in storage. The combination of ClO_2 and marine antagonists could be a viable alternative to the synthetic fungicides used for the postharvest management of anthracnose disease in Ataulfo mango fruits.

Disclosure statement

No potential conflict of interest was reported by the authors.

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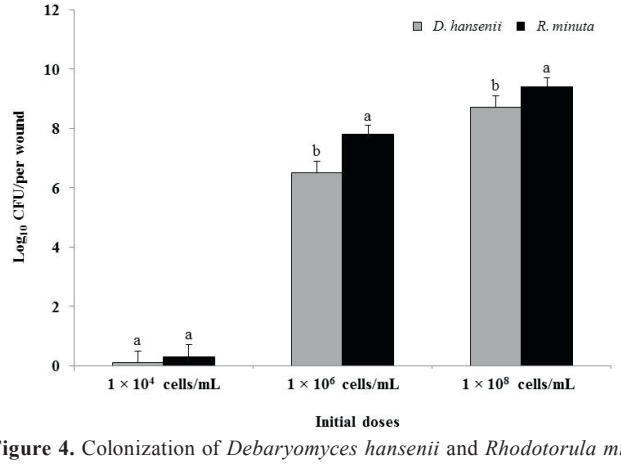

Figure 4. Colonization of *Debaryomyces hansenii* and *Rhodotorula minuta* yeasts on the wounds of Ataulfo mango fruits. Different letters indicate significant differences detected by ANOVA followed by the LSD Fisher post hoc test ($p<0.05$).

Resumen

J.J. Reyes-Perez, S. Vero, E. Diaz-Rivera, L. Lara-Capistran, J.C. Noa-Carrazana, y L.G. Hernandez-Montiel. 2019. Aplicación de dióxido de cloro (ClO_2) y levaduras marinas para el control poscosecha de la antracnosis en mango (*Mangifera indica* L.). Cien. Inv. Agr. 46(3): 266-275. Las enfermedades poscosecha de las frutas causan pérdidas económicas en todo el mundo. La aplicación de fungicidas sintéticos para el control de las enfermedades en poscosecha como la antracnosis en mango puede causar efectos adversos al medio ambiente, la salud humana y animal, además de generar resistencia en los fitopatógenos. El control biológico a través de levaduras marinas y la aplicación del dióxido de cloro (ClO_2) pueden ser una alternativa para el control de la antracnosis en frutos de mango cv. Ataulfo y, en la reducción de la aplicación de fungicidas sintéticos. Los resultados indican que diferentes dosis de ClO_2 inhibieron *in vitro* el crecimiento del micelio y la germinación de esporas de *C. gloeosporioides*. Cuando se aplicó ClO_2 y las levaduras marinas *Debaryomyces hansenii* y *Rhodotorula minuta* sobre frutos de mango, no se observaron signos de incidencia y lesión de antracnosis (LSD, $p<0.05$). Por lo tanto, la aplicación de levaduras antagonistas más ClO_2 proporciona un control eficiente sobre la antracnosis en frutos de mango cv. Ataulfo.

Palabras clave: *Colletotrichum gloeosporioides*, *Debaryomyces hansenii*, desinfectante, mango, *Rhodotorula minuta*.

References

Arango, J., M. Rubino, R. Auras, J. Gillett, A. Schilder, and A.L. Grzesiak. 2016. Evaluation of chlorine dioxide as an antimicrobial against *Botrytis cinerea* in California strawberries. Food Packaging Shelf Life 9:45–54.

Arrarte, E., G. Garmendia, C. Rossini, M. Wisniewski, and S. Vero. 2017. Volatile organic compounds produced by Antarctic strains of *Candida sake* play a role in the control of postharvest pathogens of apples. Biol. Control 109:14–20.

Benbow, J.M., and D. Sugar. 1999. Fruit surface colonization and biological control of postharvest diseases of pear by preharvest yeast applications. Plant Dis. 83:839–844.

Bozzola, J.J., and L.D. Russell. 1999. Electron microscopy principles and techniques for biologists. 2nd Ed. Jones and Bartlett Publishers, Sudbury, MA.

Calvo, H., D. Redondo, S. Remón, M.E, Venturini, and E. Arias. 2019. Efficacy of electrolyzed water, chlorine dioxide and photocatalysis for disinfection and removal of pesticide residues from stone fruit. Postharvest Biol. Technol. 148:22–31.

Chen, P.H., and J.Y. Chou. 2017. Screening and identification of yeasts antagonistic to pathogenic fungi show a narrow optimal pH range for antagonistic activity. Pol. J. Microbiol. 66:101–106.

de Oliveira, K.Á.R., L.R.R. Berger, S.A. de Araújo, M.P.S. Câmara, and E.L. de Souza. 2017. Synergistic mixtures of chitosan and *Mentha piperita* L. essential oil to inhibit *Colletotrichum* species and anthracnose development in mango cultivar Tommy Atkins. Food Microbiol. 66:96–103.

Di Francesco, A., C. Martini, and M. Mari. 2016. Biological control of postharvest diseases by microbial antagonists: how many mechanisms of action?. Eur. J. Plant Pathol. 145:711–717.

Esteriol, M., C. Copier, A. Román, M.J. Araneda, M. Rubilar, I. Pérez, and J. Auger. 2017. Frequency of fungicide-resistant *Botrytis cinerea* popula-

tions isolated from 'Thompson Seedless' table grapes in the Central Valley of Chile. Cien. Inv. Agr. 44:295–306.

Grzegorczyk, M., B. Zarowska, C. Restuccia, and G. Cirvilleri. 2017. Postharvest biocontrol ability of killer yeasts against *Monilinia fructigena* and *Monilinia fructicola* on stone fruit. Food Microbiol. 61:93–101.

Hernandez-Montiel, L.G., R. Zulueta Rodriguez, C. Angulo, E.O. Rueda Puente, E.E. Quiñonez Aguilar, and R. Galicia. 2017. Marine yeasts and bacteria as biological control agents against anthracnose on mango. J. Phytopathol. 165:833–840.

Hernandez-Montiel, L.G., E.D. Gutierrez-Perez, B. Murillo-Amador, S. Vero, R.G. Chiquito-Contreras, and G. Rincon-Enriquez. 2018. Mechanisms employed by *Debaryomyces hansenii* in biological control of anthracnose disease on papaya fruit. Postharvest Biol. Technol. 139:31–37.

Klein, M.N., and K.C. Kupper. 2018. Biofilm production by *Aureobasidium pullulans* improves biocontrol against sour rot in citrus. Food Microbiol. 69:1–10.

Kreske, A.C., J.H. Ryu, and L.R. Beuchat. 2006. Evaluation of chlorine, chlorine dioxide, and a peroxyacetic acid–based sanitizer for effectiveness in killing *Bacillus cereus* and *Bacillus thuringiensis* spores in suspensions, on the surface of stainless steel, and on apples. J. Food Prot. 69:1892–1903.

Lee, S.H., H. Shin, J.H. Kim, K.Y. Ryu, H.T. Kim, B. Cha, and J.S. Cha. 2019. Effect on colony growth inhibition of soil-borne fungal pathogens by available chlorine content in sodium hypochlorite. Plant Pathol. J. 35:156–163.

Liu, J., Y. Sui, M. Wisniewski, S. Droby, and Y. Liu. 2013. Utilization of antagonistic yeasts to manage postharvest fungal diseases of fruit. Int. J. Food Microbiol. 167:153–160.

Mahunu, G.K., H. Zhang, Q. Yang, X. Zhang, D. Li, and Y. Zhou. 2016. Improving the biocontrol efficacy of *Pichia caribbica* with phytic acid against postharvest blue mold and natural decay in apples. Biol. Control. 92:172–180.

Mailly, F., L. Hossard, J.M. Barbier, M. Thiollet-Scholtus, and C. Gary. 2017. Quantifying the impact of crop protection practices on pesticide use in wine-growing systems. Eur. J. Agron. 84:23–34.

Mei, L., T. Shilong, S. Jin, W. Xizhuo, C. Jianxin, L. Shouqiang, G. Xia, and T. Jiachun. 2017. Effects of chlorine dioxide on morphology and ultrastructure of *Fusarium sulphureum* and its virulence to potato tubers. Int. J. Agric. Biol. Eng. 10:242–250.

Meireles, A., C. Ferreira, L. Melo, and M. Simões. 2017. Comparative stability and efficacy of selected chlorine-based biocides against *Escherichia coli* in planktonic and biofilm states. Food Res. Int. 102:511–518.

Ochoa, J.L., M. Ramirez-Orozco, N.Y. Hernández-Saavedra, D. Hernández-Saavedra, and A. Sánchez-Paz. 1995. Halotolerant yeast *Debaryomyces hansenii* as an alternative source of Cu/Zn superoxide dismutase (SOD). J. Marine. Biotech. 3:224–227.

Onyeani, C., and S. Osunlaja. 2012. Comparative effect of Nigerian indigenous plants in the control of anthracnose disease of mango fruits. Int. J. Sci. Technol. Res. 1:80–85.

Parafati, L., A. Vitale, C. Restuccia, and G. Cirvilleri. 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.

Ramírez-Orozco, M., N.Y. Hernández-Saavedra, and J.L. Ochoa. 2001. *Debaryomyces hansenii* growth in nonsterile seawater ClO_2-peptone-containing medium. Can. J. Microbiol. 47:676–679.

Rivas-Garcia, T., B. Murillo-Amador, A. Nieto-Garibay, G. Rincon-Enriquez, R.G. Chiquito-Contreras, and L.G. Hernandez-Montiel. 2019. Enhanced biocontrol of fruit rot on muskmelon by combination treatment with marine *Debaryomyces hansenii* and *Stenotrophomonas rhizophila* and their potential modes of action. Postharvest Biol. Technol. 151:61–67.

Sang-Hyun, P., and K. Dong-Hyun. 2015. Antimicrobial effect of chlorine dioxide gas against foodborne pathogens under differing conditions of relative humidity. LWT-Food Sci. Technol. 60:186–191.

Scarlett, K., D. Collins, L. Tesoriero, L. Jewell, F. van Ogtrop, and R. Daniel. 2016. Efficacy of chlorine, chlorine dioxide and ultraviolet radiation as disinfectants against plant pathogens in irrigation water. Eur. J. Plant Pathol. 145:27–38.

Sharma, P., A.B. Jha, R.S. Dubey, and M. Pessarakli. 2012. Reactive oxygen species, oxidative damage, and antioxidative defense mechanism in plants under stressful conditions. J. Bot. 1–26.

Sharma, R.R., D. Singh, and R. Singh. 2009. Biological control of postharvest diseases of fruits and vegetables by microbial antagonists: a review. Biol. Control. 50:205–221.

Shinde, U.S., K.H. Pujari, and M.S. Shedge. 2017. Effect of sodium hypochlorite on chemical composition of graded mango (*Mangifera indica* L.) fruits cv. Alphonso. Adv. Agric Res. Technol. J. 1:41–43.

Snowdon, A.L. 2010. A colour atlas of post-harvest diseases and disorders of fruit and vegetables: General Introduction and Fruits. CRC Press: Boca Raton, Florida, USA.

Soylu, E.M., S. Soylu, and S. Kurt. 2006. Antimicrobial activities of the essential oils of various plants against tomato late blight disease agent *Phytophthora infestans*. Mycopathologia. 161:119–128.

Sun, X., B. Zhou, Y. Luo, C. Ference, E. Baldwin, K. Harrison, and J. Bai. 2017a. Effect of controlled-release chlorine dioxide on the quality and safety of cherry/grape tomatoes. Food Control. 82:26–30.

Sun, X., E. Baldwin, A. Plotto, J. Narciso, C. Ference, M. Ritenour, K. Harrison, J. Gangemi, and J. Bai. 2017b. Controlled-release of chlorine dioxide in a perforated packaging system to extend the storage life and improve the safety of grape tomatoes. J. Vis. Exp. 122:1–6.

Tian, Y.Q., W. Li, Z.T. Jiang, M.M. Jing, and Y.Z. Shao. 2018. The preservation effect of *Metschnikowia pulcherrima* yeast on anthracnose of postharvest mango fruits and the possible mechanism. Food Sci. Biotechnol. 27:95–105.

Trinetta, V., R.H. Linton, and M.T. Morgan. 2013. Use of chlorine dioxide gas for thepostharvest control of *Alternaria alternata* and *Stemphylium vesicarium* on Roma tomatoes. J. Sci. Food Agric. 93:3330–3333.

Villanueva, C.M., E. Gracia-Lavedan, C. Bosetti, E. Righi, A.J. Molina, V. Martín, E. Boldo, N. Aragonés, B. Perez-Gomez, M. Pollan, I.G. Acebo, J.M. Altzibar, A.J. Zabala, E. Ardanaz, R. Peiró, A. Tardón, M.D. Chirlaque, A. Tavani, J. Polesel, D. Serraino, F. Pisa, G. Castaño-Vinyals, A. Espinosa, N. Espejo-Herrera, M. Palau, V. Moreno, C. La Vecchia, G. Aggazzotti, M.J. Nieuwenhuijsen, and M. Kogevinas. 2017. Colorectal cancer and long-term exposure to trihalomethanes in drinking water: a multicenter case-control study in Spain and Italy. Environ. Health Perspect. 125:56–95.

Villanueva, C.M., M. Kogevinas, and J.O. Grimalt. 2003. Haloacetic acids and trihalomethanes in finished drinking waters from heterogeneous sources. Water Res. 37:953–958.

Wang, Z., J. Narciso, A. Biotteau, A. Plotto, E. Baldwin, and J. Bai. 2014. Improving storability of fresh strawberries with controlled release chlorine dioxide in perforated clamshell packaging. Food Bioprocess Tech. 7:3516–3524.

Wen, G., X. Xu, T. Huang, H. Zhu, and J. Ma. 2017. Inactivation of three genera of dominant fungal spores in groundwater using chlorine dioxide: Effectiveness, influencing factors, and mechanisms. Water Res. 125:132–140.

Yao, H., S. Tiana, and Y. Wanga. 2004. Sodium bicarbonate enhances biocontrol efficacy of yeasts on fungal spoilage of pears. Int. J. Food Microbiol. 93:297–304.

Zhang, Z., D. Yang, B. Yang, Z. Gao, M. Li, Y. Jiang, and M. Hu. 2013. β-Aminobutyric acid induces resistance of mango fruit to postharvest anthracnose caused by *Colletotrichum gloeosporioides* and enhances activity of fruit defense mechanisms. Sci. Hort. 160:78–84.

Zhu, C., Z. Chen, and G. Yu. 2013. Fungicidal mechanism of chlorine dioxide on *Saccharomyces cerevisiae.* Ann. Microbiol. 63:495–502.

Zou, H., and L. Wang. 2017. The disinfection effect of a novel continuous-flow water sterilizing system coupling dual-frequency ultrasound with sodium hypochlorite in pilot scale. Ultrason. Sonochem. 36:246–252.