Green Chemistry

Green can also be mean

In the microbiological world, it is the job of 'good' microbes to keep the 'bad' ones under control. These miniature battles, which rage every day on the surfaces of the fruit we eat, have great potential for post-harvest disease control.

IN THE CITRUS and pome fruit industries, between 3% and 6% of fruit that is harvested in a typical season, decays mainly because of *Penicillium* rot. However, under favourable disease conditions, losses of up to 50% can occur in the post-harvest phase of fruit. These losses not only affect profit margins and initial input costs, they also eat away at retailer and consumer confidence in the marketplace.

The fresh fruit industry is therefore justifiably concerned about reducing the impact of blue and green mould. At the moment, producers rely on only a few post-harvest chemicals to protect fruit against the 'baddies'. Prof. Lise Korsten from the University of Pretoria's Department of Microbiology and Plant Pathology, however, is working on ways to expand the arsenal of weapons against the 'baddies' in a green way.

1 Prof. Lise Korsten.
2 Pears infested with green mould.

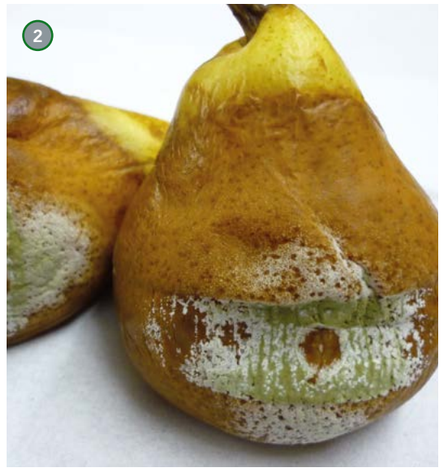
THE FIGHT-BACK

Like wily enemies, blue and green mould can defend themselves against fungicides. Under favourable conditions they proliferate rapidly to gain the upper hand. Producers often give the microbes a helping hand by following poor sanitation practices that result in a build-up of microorganisms that can infect the fruit later on, often at the end of the long export chain.

The mould's survival strategy also includes building up natural resistance against fungicides that are used regularly. Fungicide-resistant pathogens are a problem that has been detected in pack houses across the globe. To address it, the fruit industry has introduced hygiene management practices that include the use of a range of non-selective compounds and sanitisers. "But to be really effective, post-harvest disease management has to combine improved hygiene and sanitation practices with other disease control measures," says Prof. Korsten. "Our research is suggesting that pushing good microbes into the battleground through bio-control, green chemicals and best sanitation practices is the way to go. These greenies can also be combined or alternated with our existing arsenal of chemicals to manage and prevent the build-up of resistance."

Prof. Korsten adds that consumers across the world are increasingly concerned about human and environmental health and safety, hence the emphasis on going green. In response, researchers have intensified their efforts to develop new biocontrol products and low-toxicity compounds, also called soft chemicals.

Various bio-fungicides have already been developed and registered for the control of post-harvest diseases on fruit. "Although these agents show promise for commercial application, there are a variety of reasons, from questions about efficacy to customer regulations, why they have not been widely adopted," says Prof. Korsten. "The study my team and I undertook, evaluated the efficacy of different combinations of biocontrol agents and sanitisers against post-harvest fruit disease. Our aim was to develop an integrated disease management programme."



Two biocontrol agents were tested in the study. The first was a strain of yeast that occurs naturally on the surface of pears. The second was a product called Organisan II, which consists of a formulation of chitosan and yucca plant extracts. Chitosan, which consists of 75% deacetylated crab shells, has antimicrobial properties and prompts plants to deploy their own defence mechanisms.

IT'S NOT THAT SIMPLE

If a product is 'green' it has to be good, right? Not necessarily.

There are a variety of reasons for the slow uptake of green chemistry. In some instances, producers are reluctant to try something new when it comes to crop protection. Increased awareness and education can help to address this issue.

Another challenge is the difficulty to register bioproducts due to a lack of suitable regulations covering these novel products.

Market requirements can be a further potential barrier to trade. Sainsbury's in the UK, for instance, has a list of banned active ingredients that its producers are not allowed to use to spray crops.

As with inorganic chemicals, the green products need to be thoroughly researched and tested for all possible applications.

Penicillium was the pathogen targeted in the study. The project was done on-site in the Eastern Cape where the participating pack house made all facilities available and provided technical support. The research team used the microbiology laboratory at the University of Pretoria to identify the pathogens and to do *in vitro* trials.

DRAWING THE BATTLE LINES

The yeast test involved isolating dominant yeast colonies from the surface of a pear before and after it

PROJECT TITLE
The development and screening of alternative post-harvest disease control products and practices for citrus and pome fruit

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DURATION
One year

PHI-2 CONTRIBUTION
R131 513

LEAD INSTITUTION
University of Pretoria (Department of Microbiology and Plant Pathology)

BENEFICIARY
The citrus and deciduous fruit industries

FOCUS AREA
Green chemistry

HUMAN CAPITAL DEVELOPMENT
Three MSc and one BSc students

PRESENTATIONS AND PAPERS DELIVERED
Six

PUBLICATIONS
Two

Green Chemistry

> "We have proven the potential role that green chemistry can play in an integrated disease control approach."
> Prof. Lise Korsten

was placed in controlled atmosphere storage. Cultures of each of the isolates were prepared and propagated and then placed next to a spot of *Penicillium* spore suspension. The plates containing the isolates and the spores were incubated at 25°C and the researchers recorded measurements every day. The radius of the respective growths was used as a basis to determine whether and by how much the growth of the spores was being slowed down.

The Organisan II research was done on different citrus types both before and after the fruit has been harvested.

In the pre-harvest trials selected trees were sprayed with a solution of Organisan II. The fruit was then inoculated with a spore suspension of green mould and kept at 25°C for five to seven days to allow lesions to develop. Disease incidence and severity was noted. This experiment was done on 'Clementine' and 'Nova' mandarin oranges.

For the post-harvest investigation, the researchers selected and harvested unblemished and disease-free fruit that was export-ready. The fruit did not receive the usual post-harvest treatments, namely a warm fungicide bath and waxing. After being surface sterilised, the fruit was inoculated with a spore suspension of green mould (*P. digitatum*). The spore inoculation took place 12 hours before the Organisan II treatment was done as a preventative or 12 hours after inoculation for the curative approach. The trial was repeated under cold storage simulation circumstances where the fruit was kept for 21 days at 4,5°C

THE RESULTS

The researchers found that most of the natural yeast strains slowed down or prevented the growth of the *Penicillium* pathogen.

In terms of the Organisan II trials, the curative treatment of Organisan II on 'Midknight' Valencias and 'Clementine' and 'Nova' mandarins showed much less control of *P. digitatum* than the current commercial products. When it comes to preventative action, however, the treatment was more effective than the commercial fungicide application.

"I am satisfied that we have proven the potential role that green chemistry can play in an integrated disease control approach," says Prof. Korsten. "This project should be taken further to show the efficacy of new biocontrol agents and to ensure industry adoption of some of the new technologies."

Further studies will focus on integrated strategies for pome fruit and on biocontrol products in citrus pack house systems. The commercial value of this project is to develop new, integrated disease control strategies for the South African fruit industry to ensure product quality and safety.

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1 *Penicillium* up close and personal.
2 One affected fruit on an orange tree in the production chamber shows decay caused by *Penicillium*.
3 Pieter Louw, MSc student.
4 Prof. Lise Korsten and MSc student, SB Coetzee.

SPREAD THE WORD

The new knowledge of integrated disease control strategies that Prof. Korsten and her team have gained, has been transferred to the Eastern Cape citrus production region. New production practices have been shared with farmers of this region through pack house meetings. Additional knowledge transfer has been shared with the broader citrus community through a presentation at the Citrus Research International (CRI) conference. Research findings have also been shared with the international post-harvest community through an oral presentation at the International Post-harvest Conference in Stellenbosch.