## Next generation of strawberry disease control

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A team of researchers based at La Trobe University in Victoria, and at the Queensland Department of Agriculture and Fisheries (QDAF) in Nambour, is part of the Australian Research Council Sustainable Crop Protection Hub, funded by the Australian Government and industry partners, including Hort Innovation.

- BioClay<sup>TM</sup> is a completely new crop protection approach that is non-genetically modified, safe and environmentally sensitive
- Grey mould caused by Botrytis sp. is a serious pre- and post-harvest disease of berries, and other crops
- The team have developed sensitive in vitro assays to measure the effectiveness of BioClay against Botrytis on different tissues of strawberry plants

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The Sustainable Crop Protection (SCP) Hub launched in 2022, and aims to develop the novel BioClay platform and apply it to fungal disease management in a range of horticultural and crop species. The BioClay technology was developed by Professor Neena Mitter, the SCP Hub Director, and colleagues at the University of Queensland. BioClay is a completely new crop protection approach that is non-genetically modified, safe and environmentally sensitive. It is a biodegradable spray solution of clay particles that bind double stranded RNA (dsRNA), releasing it slowly once applied to the plant, to fight pests with longer protection periods.

Grey mould caused by Botrytis sp. (particularly B. *cinerea*), is a serious disease causing enormous economic losses around the world, including in berries, where it affects fruits pre- and post-harvest (Figure 1).

As part of the SCP Hub project, the team have developed sensitive in vitro assays to measure the effectiveness of BioClay against Botrytis on different tissues of strawberry plant that will underpin future work. Other research teams within the SCP Hub are working on Botrytis in grape and pulses. Initial experiments using commercial fruit from supermarkets, or fruit sourced directly from growers, often had residual and highly variable levels of background contamination, particularly Botrytis and *Rhizopus*. These latent infections greatly interfered with the disease assays that set out to measure Botrytis disease progression. At La Trobe, two early-career scientists, Ms Lorena Rodriguez Coy, a Ph.D. student, and Dr Donovan Garcia Ceron, a recent PhD graduate, have now developed a method to produce very clean fruit, flowers and leaves. Fruit and flowers are produced from plants grown in a research glasshouse with controlled temperature and lighting (Figure 2).

Sterile leaves can be obtained from sterilised seeds germinated and grown in sterile tissue culture in a controlled environment room (Figure 3).

At QDAF, Apollo Gomez has implemented disease assays that are conducted in dedicated plant pathology glasshouses, with different tissues of intact plants infected, and disease progression monitored.

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The strawberry team at a recent Council Sustainable Crop Protection Hub Meeting. (L to R) Dr Donovan Garcia Ceron, Dr Scott Mattner, Lorena Rodriguez Coy, Apollo Gomez, and Assoc. Prof. Tony Gendall. Photo credit: Ilaria Stefani



Figure 1. Strawberry fruit heavily infected with Botrytis. The fruit on the right has Botrytis that is sporulating. These spores can now spread and infect many more fruit and flowers. Photo credit: Scott Matther

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Figure 2. Disease-free Strawberry plants under lights in a glasshouse. Photo credit: Lorena Rodriguez Coy



Figure 3. Sterile Strawberry plants grown in tissue culture. Photo credit: Lorena Rodriguez Coy



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These clean or sterile tissues are then deliberately infected with a known inoculum of Botrytis spores. The tissues are maintained in conditions that promote the growth of the fungus, and the growth of Botrytis is then measured daily using a range of imaging systems (Figure 4).

Over the next two years, these types of assays will underpin our work on the validation of BioClay as an effective disease control option for Botrytis in strawberry, by testing and optimising the timing and rate of BioClay application and comparing it to existing fungicide chemistries.

Ultimately the research team aim to provide berry growers with a non-pesticide approach for managing *Botrytis* that integrates well with other control measures. It is a ground-breaking and ambitious task that will take many years of research but could minimise growers' reliance on pesticides and reduce the risk of fungicide resistance in populations of *Botrytis*.



Figure 4. An *in vitro* assay for monitoring deliberate Botrytis infections in fruits (top) and petals (bottom). d.p.i and h.p.i are days and hours post inoculation. Photo credit: Donovan Garcia Ceron





In a parallel study, Marlo Molinaro is conducting an Honours project to examine the diversity of possible beneficial and pathogenic fungi in different production systems in Victoria for strawberry and raspberry fruit.

Although the entire suite of microbes that live in strawberries, the microbiome, has been described in other countries, there is no data for fruit produced in Australian conditions. Marlo will use a Next-Generation DNA sequencing approach to describe the full spectrum of fungi present on the surface and within the plant. We can then compare these results to those reported in the literature from international studies.

Marlo's research is particularly important so that we can design BioClay technologies that target pathogenic fungi like *Botrytis*, but do not interfere with beneficials that promote the health of the crop. With this new work to investigate and control diseases in strawberries, which involves several young scientists, we hope the future of research in disease control in berries is promising – albeit with many challenges ahead!



Scott Mattner (L) and Marlo Molinaro (R) collect samples for fungal diversity assays from a farm in Wandin, Victoria. Photo credit: Tony Gendall

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