





## Innovating Avocado

## From Lab to Orchard and Beyond



QAAFI, The University of Queensland, Australia

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Mitter Lab: Innovating Avocado We love Challenges.... We love Avocado...









## Innovating Avocado – The list keeps on growing...



The Queensland Alliance for Agriculture and Food Innovation (QAAFI) is a research institute of The University of Queensland (UQ), supported by the Queensland Government.





### **Chromosomal level genome of Hass**



QAAF







### Reference genome sequencing of >45 avocado cultivars

	#86-Berwill	A10	A8	Ashdot	Barr Duke	Belle Prima	Bounty	Carmen Hass	Duke7	Dusa	Edranol	Esther	Ettinger	Fuerte-NQ	Gem	GEM	Gwen	Harvest	Hass	Hazzard	Kidd	Lamb Hass	Latas	Laurie	Maluma	Nabal	Parida	Pinkerton	Plowman	Reed	Sharwil	Shepard	SHSR-04	Simmonds	Sir Prize	Skhirate	SR1	Thomas	Topa Topa	Tora Canyon	Velvick	Whitsell	Wurtz	Zutano
M1 (T-G)																																												
M2 (T-C)																																												
M3 (A-G)																																												
M4 (G-A)																																												
M5 (C-T)																																												
M6 (G-A)																Γ																												
M7 (T-C)																	4									<b>.</b>														1 -				
M8 (C-T)																47 more avocado cultivars sequenced and compared to Hass																												
M9 (T-A-G)		Minimal marker set developed that distinguishes cultivars																																										







## Innovating Avocado – genomics

- Hass and Gwen Genome as a reference for future studies.
- Identification, characterization and functional analysis of features like genes, proteins, non-coding RNAs etc.
- Markers assisted identification of cultivars
- Further analysis of variants will assist in identifying reasons for differences between cultivars.

This genome provides a tool to support future advances in the development of elite avocado varieties with desired traits







### **Clonal Rootstock Propagation**

Clonal propagation of rootstocks. (Ernst 1999) (with adaptations)





Dr André Ernst at the Maluma Symposium in March 2020

Industry lost a Champion









#### New Australian technology can significantly cut avocado tree wait times

June 05 , 2021

(More News) (Today's Headline) (Top Series)

WE ARE AVOFAMOUS!





10TH WORLD AVOCADO CONGRESS New Zealand 2023







### **AVOCADO TISSUE CULTURE: LAB - NURSERY - ORCHARD**

**200** to **500** plants from < 1mm cutting from a mature tree

Andersons Horticulture – Fruitful collaboration



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## Nursery & Field Evaluated - 4 years of data







### Tissue culture plants in Bundaberg, 2019-2023









## **End User Perceptions**

## **International and Domestic Survey**

### **Domestic cohort:**

- 42% identified as growers/farmers
- 19% as breeders/researchers
- 11% Agronomists
  - 11% consultants
  - 8% as nursery/suppliers

Perceptual Factors Influencing the Adoption of Innovative Tissue Culture Technology by the Australian Avocado Industry. Thorne, H.B.; Axtens, J.; Best, T. Agriculture 2022, 12, 1288. <u>https://doi.org/10.3390/agriculture12091288</u>









## End User Perceptions- Major Survey Finding #1

- >72% cannot access enough plants
- >56% cannot access the quality of plants they want
- >53% cannot access the cultivars they want
- $\rightarrow$  Confirmed these problems are not just perceived by researchers



Figure 2. Industry ratings of access to avocado trees.



Figure 3. Industry rating of their access to the quality of avocado trees they want.







## Major Survey Finding #2

- 2/3 Domestic respondents believe TC could be beneficial
- Most desired characteristics:
  - Quality of TC root system
  - Disease tolerance/Disease-free status
  - Fruit quality
- $\gamma$  What would be the major influencer for TC adoption?
  - Field Observations (80%) and Professional Recommendations (83%)
- $\rightarrow$  Field-testing performance is essential for industry acceptance

A major financial benefit at the grower level will be on-demand supply of desired trees – earlier planting = earlier returns

### Speed to market - new rootstocks and scions









## **Frozen orchards – plants now in the field**



Developed the worlds first cryopreservation protocol using shoot tips for avocado germplasm conservation – NEED FOR GLOBAL CONSORTIUM – CRYOBANKS FOR AVOCADO

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Science

#### Cryopreserved avocado shoots could mean guacamole on Mars

Researchers are working to head off any chance of an "avocado apocalypse" on Earth.







University of Queensland's Neena Mitter and Chris O'Brien inspect avocado tissue cultures in the lab. Peter Geale/University of Queensland

Humans are intent on going to Mars, but what will martian cuisine be like? Thanks to the work of researchers at the University of Queensland in Australia, there could be a side of guacamole to go with all the Mars notatoes





## Cell Fusion/ Gene Editing (CRISPR):

### **Cell Fusion Technology – Non GM**

Ability to hybridise diverse germplasm without the need for flowering

Duboisia The Queensland Alliance for Agriculture and Food Innovation (QAAFI) is a research institute of The University of Queensland (UQ), supported by the Queensland Government.

Callus proliferation

























## Unleashing the Power of Avocado Indoor

**Speed Breeding** 

Aeroponics/hydroponics/light/temperature

#### Synchronous flowering Reduced time to flowering and fruit set Rapid Screening



**Prof Paul Gauthier** 



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CKICOS code 00025E





## Avocado in Iceland! 6.8 Avocado in greenhouse



#### Business case for large scale crop production in greenhouse facilities in Iceland for the global market

E. J. Baeza<sup>1</sup>, Y. Dijkxhoorn<sup>2</sup>, K. Logatcheva<sup>2</sup>, W. Hennen<sup>2</sup>, G. Splinter<sup>2</sup>,
C. Stanghellini<sup>1</sup> and S. Hemming<sup>1</sup>
1 Wageningen Plant Research, Business Unit Greenhouse Horticulture, 2 Wageningen Economic Research



In the greenhouse, in order to meet the same amount of light received in the indoor farm, a light intensity of 525 micromol/m<sup>2</sup>s. Since the temperature regime is 25/20 °C, we can simulate the tomato crop with the new temperature and light intensity settings. Results are summarized in Table 25 and Table 26.

#### Table 36

Summary of main resource uses and predicted avocado yield for Keflavik and Akureyri for HPS lamps.

	Energy used for heating (MJ m <sup>.2</sup> )*	Electricity used for artificial lighting(KWh/ m <sup>2</sup> )	Hours lamps are on (h)	Amount of CO <sub>2</sub> used (kg/m <sup>2</sup> )	Avocado yield (kg/m <sup>2</sup> )
Keflavik-reference scenario	976	1607	6213	46.3	4.4
Akureyri-reference scenario	1019	1623	6228	42.5	4.4

\* This number already includes the extra energy required on an average year to deal with the snow in Keflavik and Akureyri

Report WPR-1049







## **RNA-based Biopesticides - BioClay<sup>TM</sup>**

- RNA as the biological active ingredient
- Clay particles as carriers of the active
- > Non-GM
- > NO RESIDUE
- > SPECIFIC
- > STABLE
- > SUSTAINABLE
- > SAFE

 Multiple Patents
Nufarm Australia Limited – industry partner
ARC Hub ~\$18M Inventors: Prof. Neena Mitter Prof. Gordon Xu Prof. Max Lu

BILL& MELINDA GATES foundation









Environmentally friendly?

No residue?

Stabilise? Stick to

Stick to the leaf?



- Unstable
- Easily washed off
- Short protection window

Can we deliver RNA sprays as a viable system for growers without genetic modification?

Non-toxic?

Protect from rain?

Easy to adopt?



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CRICOS code 00025B





## What is BioClay?

Inert biodegradable clay (Mg + Fe) to deliver RNAi

- Applied as a spray application without the need to alter the plant genome
- Clay layers degrade naturally leaving no residue
- Extended stability and slow release of dsRNA on plant surface



# Double stranded RNA of the pest or pathogen is used to kill the pathogen itself – Nature vs Nature



## **BioClay Platform**

- Australian Research Council Research Hub for Sustainable **Crop Protection -Targeting Fungal Diseases**
- ~\$18 million cash and in-kind
- Universities, multiple RDCs, State Governments and Nufarm Australia Limited

https://crophub.com.au/







Dueensland

Government













### The Research Hub process











## BioClay it works -



Tomato spotted wilt virus/capsicum



Water



Naked dsRNA Zucchini yellow mosaic virus



Live Whitefly

BioClay Dead Whitefly



**BioClay** Control



Botrytis on chickpea



dsRNA protects lemon myrtle trees from rust



School of Chemistry and Molecular Biosciences







## Targeting Phytophthora cinnamomi



No P. cinnamomi

+ P. cinnamomi

+ P. cinnamomi + dsRNA

Unpublished



School of Chemistry and Molecular Biosciences







Unpublished

## Testing different RNA application methods



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School of Chemistry and Molecular Biosciences







# dsRNA can be delivered to avocado leaves and roots via foliar sprays and trunk injections



Dr Anne Sawyer



			lea	ves	· ·			roots										
	in	jecte	d	S	oraye	d	ir	njecte	d	sprayed								
+ve	1	2	3	1	2	3	1	2	3	1	2	3						



Intact dsRNA is present in new leaves and roots 6 weeks post delivery







On 8<sup>th</sup> October 2019 the Australian Parliament formally agreed with the Office of the Gene Technology Regulator's proposal that topically-applied dsRNA be exempt from GMO regulations



Gene Technology Amendment (2019 Measures No. 1) Regulations 2019

#### 24 Schedule 1A (at the end of the table)

Add:

- 11 Introduction of RNA into an organism, if:
  - (a) the RNA cannot be translated into a polypeptide; and
  - (b) the introduction of the RNA cannot result in an alteration of the organism's genome sequence; and
  - (c) the introduction of the RNA cannot give rise to an infectious agent.







## Trade and Markets

- Naked dsRNA:
- Short or nil withholding periods (set to allow chemical residues in edible commodities for domestic markets)

• Short or nil export intervals (to satisfy the standards imposed by overseas trading partners)







Innovations aimed at contributing to the supermarket trolley

## **Endless** possibilities...

**Design** of regulation and public opinion are crucial







PROTECTED **CROPPING !!** 



#### PACKHOUSES!!



#### ANIMAL HEALTH !!





**BIOSECURITY!!** 



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g Jin, Nature Plants, 2016

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Hort Innovation





Wine Australia



Seen in the @UQ\_News car park this morning. Rumors are that it belongs to @neenamitter from @QAAFI . Looking forward to seeing what @QAAFI\_Animal drives to work *2* 



#### QAAFI



## Thank you FROM AVONDERFUL MITTER TEAM

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