

Let's toast – Avocado tissue culture goes commercial

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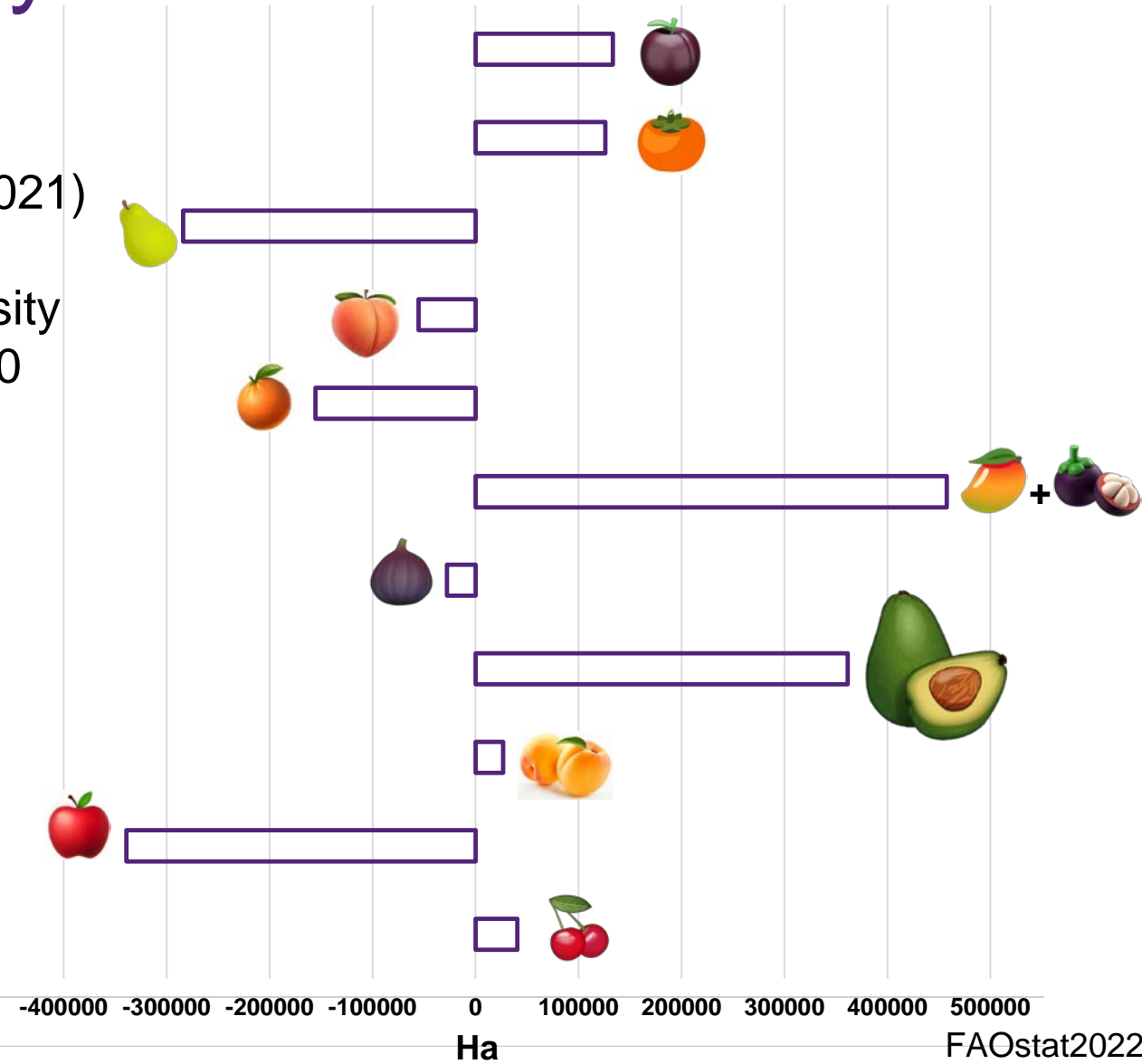


Trend of avocado industry

- Global production increasing with cultivation expansion
- Area harvested increased by 52,411 ha (2020-2021)
- Intensive orchard practices
- Conventional planting 400 plants/ha to high density planting 800 plants/ha to ultra high density >1200 plants/ha



Change in cultivated area 2011-2020



Avocado Propagation



Scion (Bears fruit)

Rootstock

Seedling rootstocks

Genetically different
Comparatively cheap
Comparatively fast
Depend on seeds/season



Clonal Rootstock

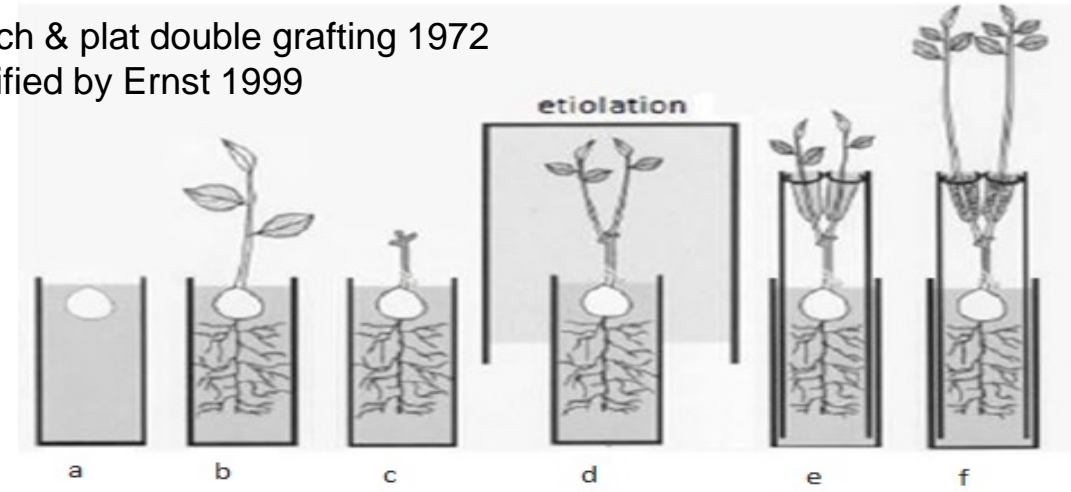
Genetically identical
Depend on seed/budwood/season
Time consuming
Resource intensive



- Large orchard maintenance to obtain rootstock/nurse seeds, budwood
- Pest and disease risks

Clonal propagation.. Way too cumbersome

Frolich & plat double grafting 1972
Modified by Ernst 1999



Pictures – Allesbeste, SA



Nurse seeds



Grafted with rootstock budwood



Etiolated buds of rootstock shoots



Rooted rootstocks



Clonal rootstocks ready for scion grafting

Tissue culture

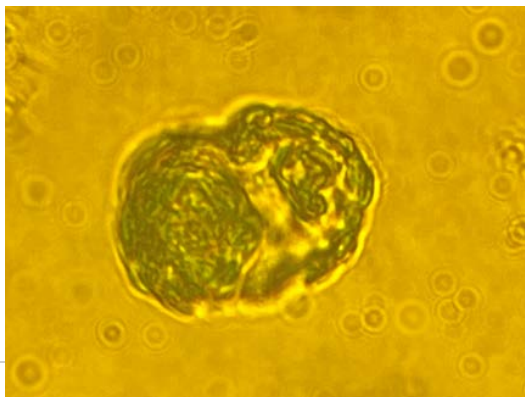
**Aseptic culture of cells/tissues/organs/components under defined
physical and chemical conditions in vitro.**

(Thrope., 2007)

Totipotency of cells

Total potential of a single cell to become a complete individual

- De-differentiation – Change from given differentiated state to a less differentiated or stem cell-like state
- Re-Differentiation – Change from less differentiated state to a more differentiated state



TC as a propagation tool

- True-to-type multiplication of elite genotypes
- Seed-free (no supply risk, germination + season free)
- Soil and field-free (pesticide/fertiliser/run-off/land-use)
- Disease-free
- Climate-independent (resilience)
- High efficiency
- Scalable production
- Year-round production



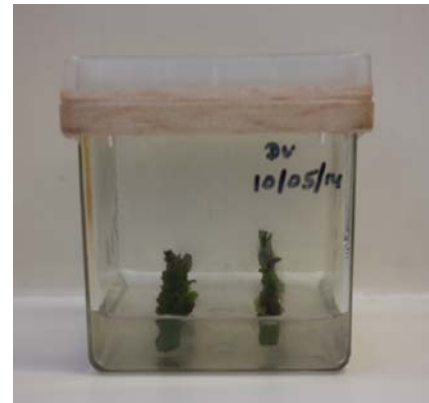
Avocado tissue culture

45 – 50 years of research in global sphere

Highly recalcitrant woody plant species, specially when mature

Problems

- Very low multiplication
- Loss of vigour in culture
- Failures for long term culture
- Poor plant quality
- Very difficult to root *in vitro*
- Low acclimatisation success
- Problems in practicality for large scale production



Micropropagation of Avocado (*Persea americana* Mill.)

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No industrially applicable tissue culture technology for avocado propagation

Key considerations

- Propagation of mature cuttings
- High multiplication rate
- Produce high quality plants (pest & disease free)
- Practical procedure applicable at industrial scale

Pathway for commercialisation



TC Proof-of-concept

TC Scale up

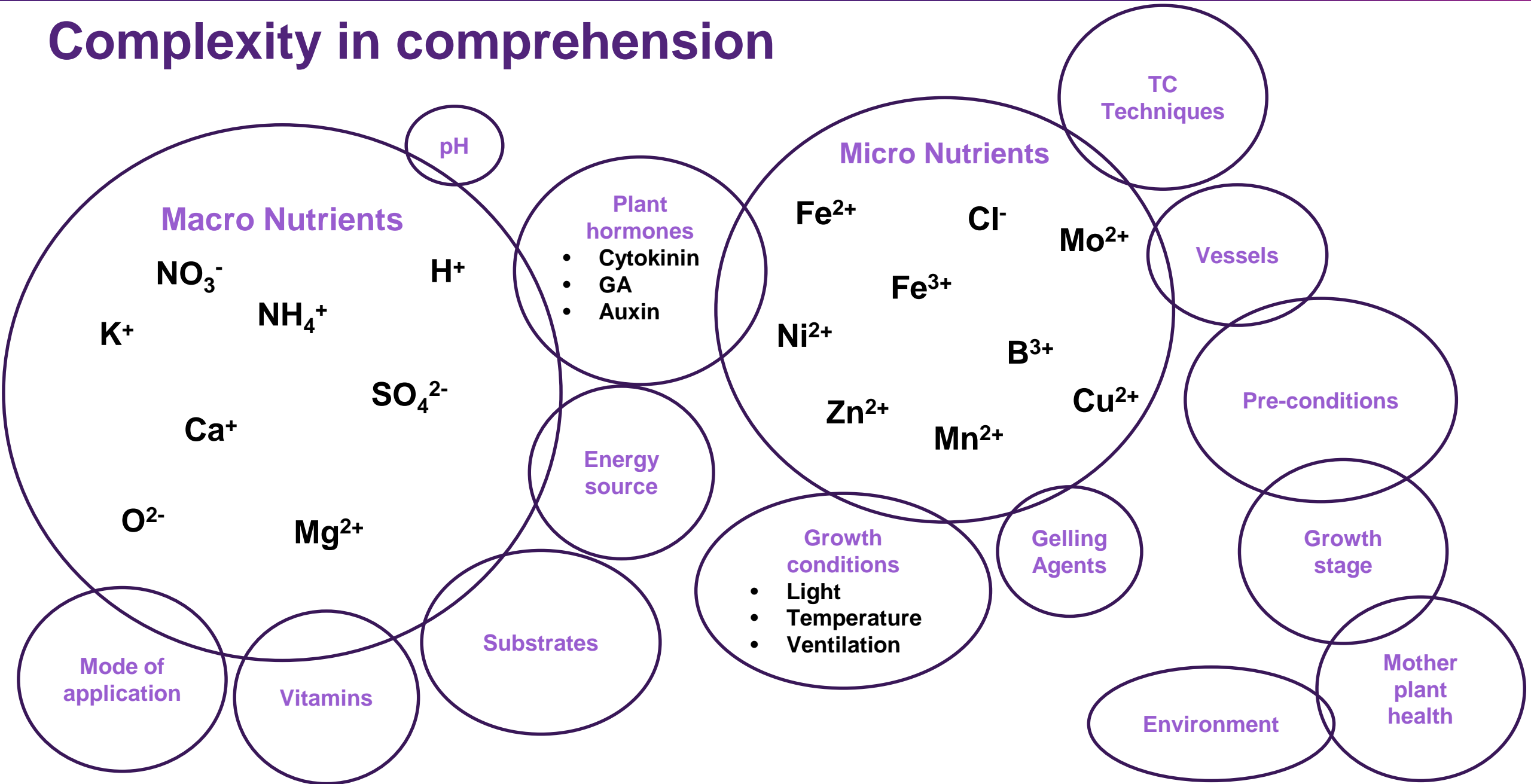
Acclimatisation

Nursery adoption

Morphology trials

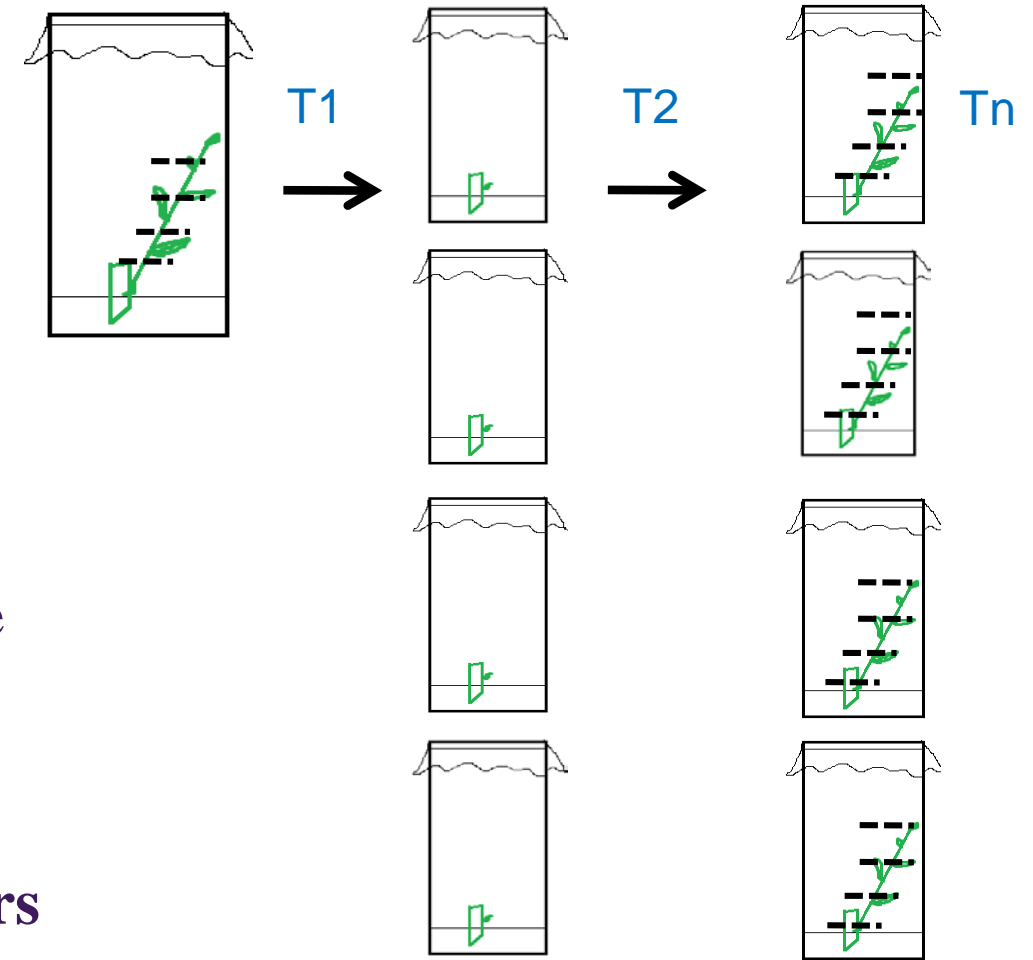
Productivity trials

Complexity in comprehension



Nodal culture

- **Numerous optimisation options**
 - **Basal nutrient media**
 - type and strength
 - Vitamins and micronutrients
 - **Hormone levels/combinations/exposures**
 - Combinations of auxins and exposure time
 - Additives to breakdown/inhibit hormones
 - **Gelling agents**
 - **Non-hormone modulators/rooting promoters**
 - **Culture vessels**
 - **Incubation conditions (light, temperature)**



J Hiti Bandaralage, 2019 (Thesis)

Nodal culture

- Optimised cultivar specific basal media and hormone conditions tested for long term culture

$$\text{Multiplication Rate} = \frac{\text{Produced no of nodes}}{\text{Initial no of nodes per tub}}$$



Cultivar	Total multiplication (culture period)	Average multiplication rate at subculture
Velvick	10.68 (12 months)	1.25
Reed	3.48 (6 months)	1.24
Kidd	21.78 (12months)	1.32
Hass	0.28 (3 months)	0.78



Actas • Proceedings | RECURSOS GENÉTICOS Y MANEJO DE VIVEROS • GENETIC RESOURCES AND NURSERY MANAGEMENT

Gibberellin and cytokinin in synergy for a rapid nodal multiplication system of avocado

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Inadequate multiplication for a commercial production system

Success in Nodal culture



Reed (80% - 8 wks)



Velvick (50% - 12 wks)



Kidd (90% - 4 wks)



Zutano (50% - 8 wks)



Reed



Velvick



Kidd



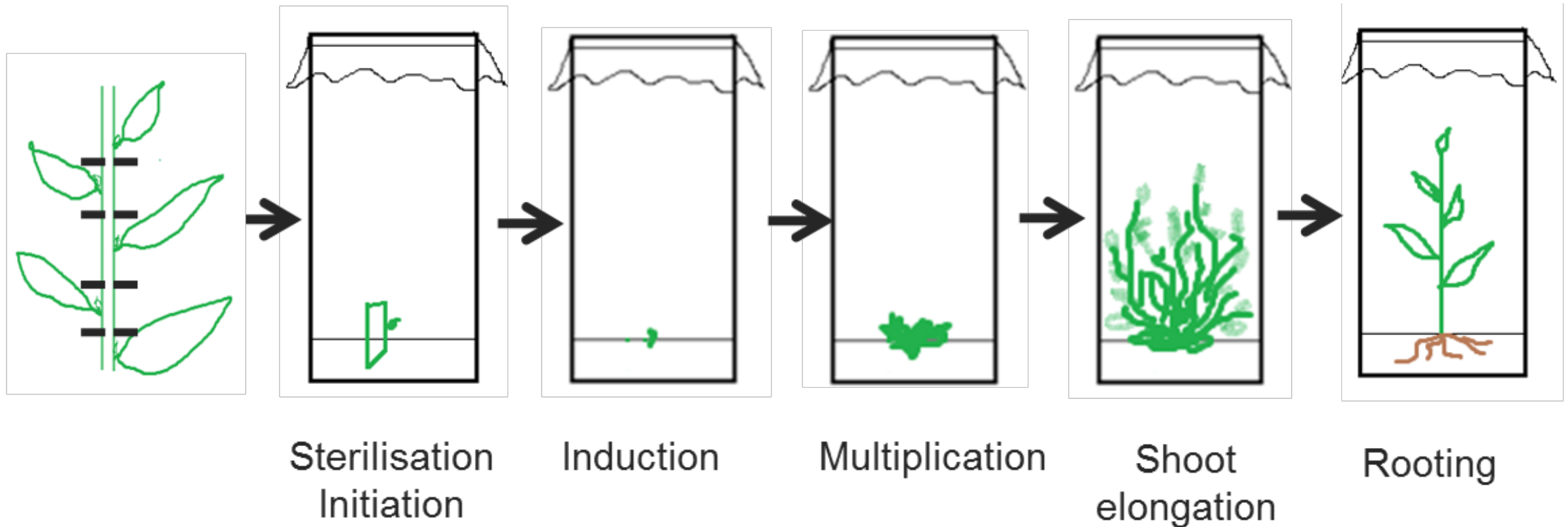
Zutano

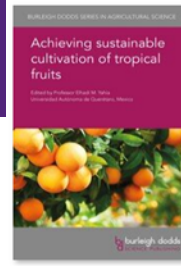
Inadequate multiplication for a commercial production system

APICAL AND OTHER RESPONSES OF TISSUES OF AVOCADO IN
ASEPTIC CULTUREC. A. Schroeder
Professor of Botany, University of California, Los Angeles

Meristem culture

Culturing of extreme apical tissues (ideally <0.1 mm) with no visible leaf primordia (Lane., 1978)





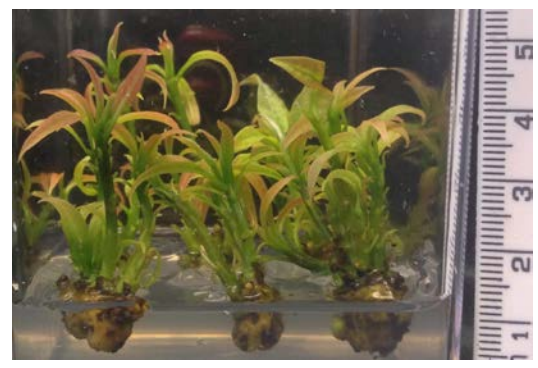
Reed



Meristem induction



Multiplication



Shoot elongation



Single shoots



Nursery maintained plants



<1 year old



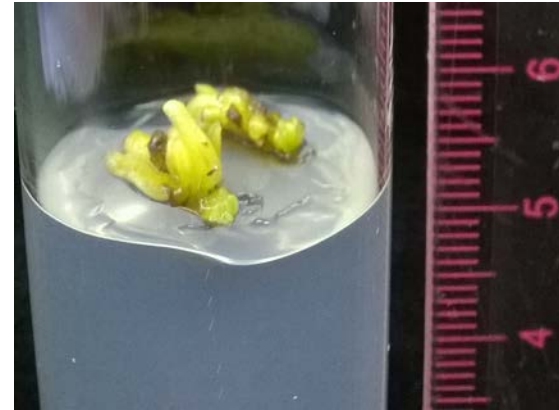
Acclimatised plant



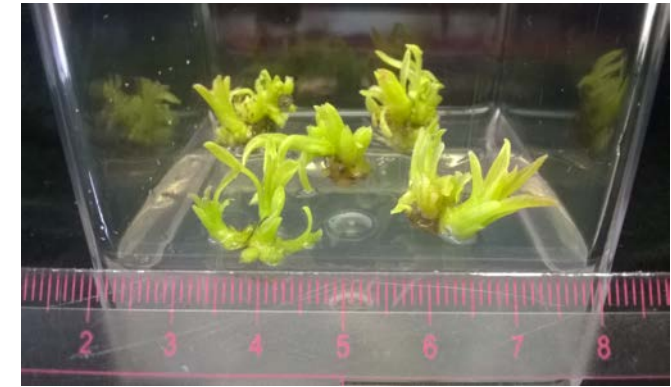
Rooting

Velvick

- Australian rootstock
- Highly productive, consistent high yielding
- Disease tolerant
- High in demand



Meristem induction



Multiplication



Shoot elongation



Single shoots



In vitro hardening



Rooting

Kidd

- Selection from Mount Tamborine
- Rootstock with farmer interest

J Hiti Bandaralage, 2019 (Thesis)



Meristem induction



Multiplication



Shoot elongation



Single shoots



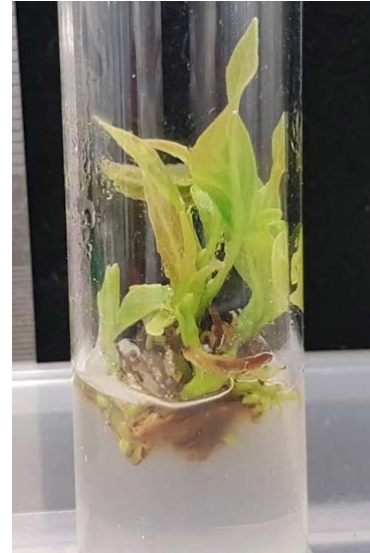
In vitro hardening



Rooting

Zutano

- Moderate to high yielding rootstock
- Considerable demand in Australia and internationally



Meristem induction



Multiplication



Shoot elongation



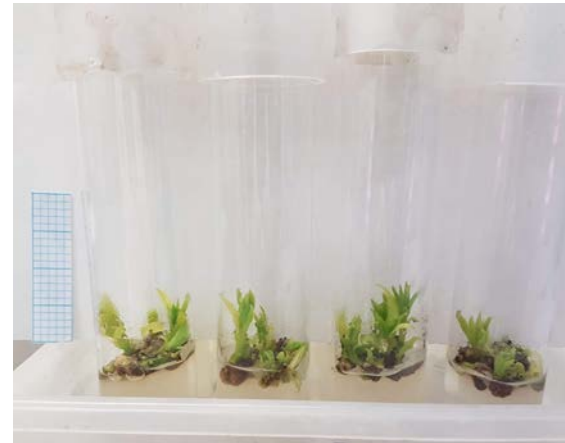
Single shoots



Rooting

Dusa

- Commercial level of *Phytophthora*-resistance
- Moderate salinity tolerant
- High international demand



Meristem induction



Multiplication



Shoot elongation



Single shoots

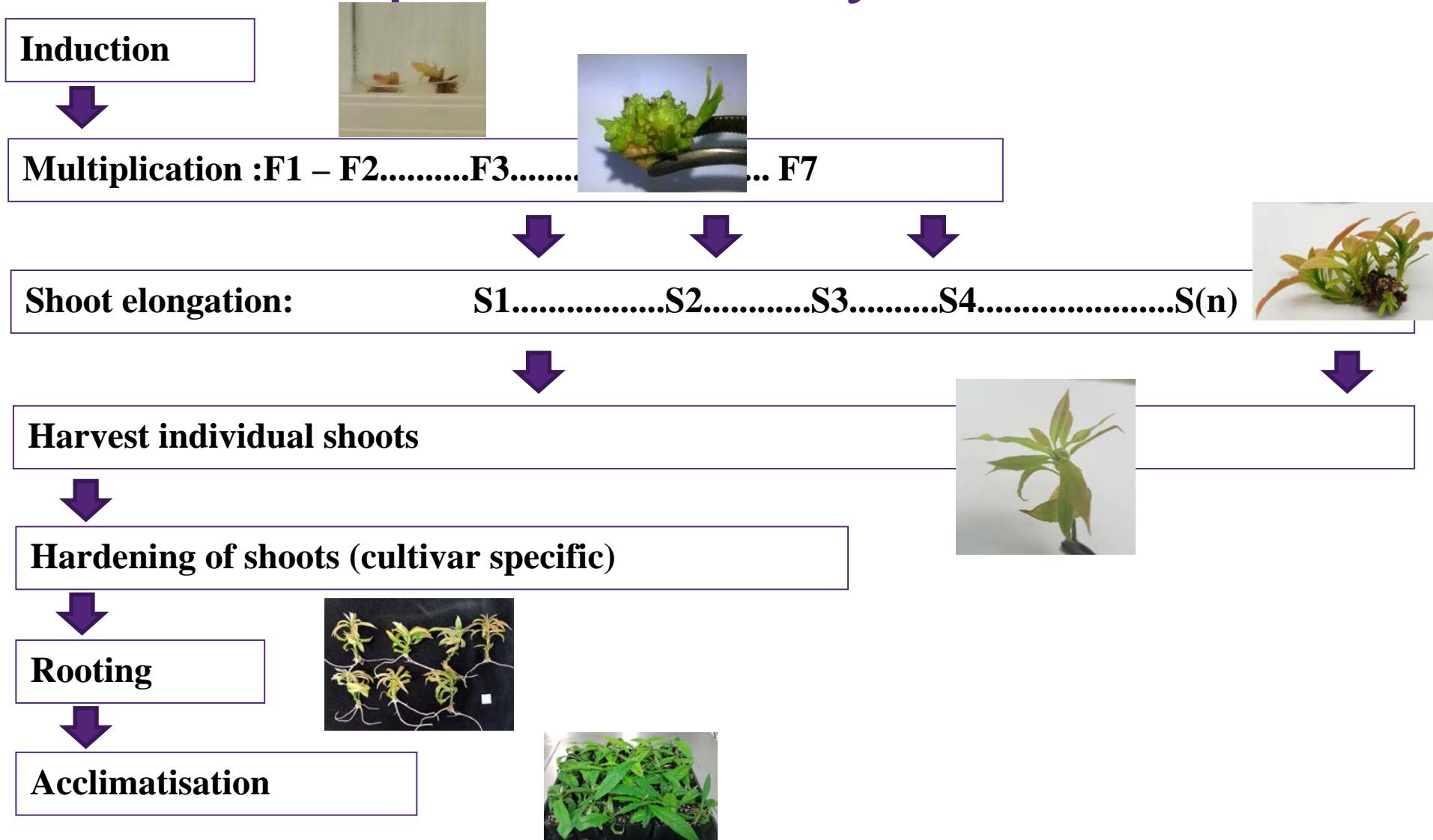


In vitro hardening



Rooting

Commercial production system



Acclimatisation: Jar to outside



Acta Hortic. 1224. ISHS 2018. DOI 10.17660/ActaHortic.2018.1224.3
Proc. VII Int. Symp. on Production and Establishment of Micropropagated Plants
Eds.: R. Paiva et al.



International Journal of Plant Biology






Acclimatization of micropropagated mature avocado

J.C.A. Hiti Bandaralage^a, A. Hayward, C. O'Brien, C. Beveridge and N. Mitter
The University of Queensland, Brisbane, Queensland, Australia.

Article

Structural Disparity of Avocado Rootstocks In Vitro for Rooting and Acclimation Success

Jayeni Hiti-Bandaralage^{*}, Alice Hayward^{} and Neena Mitter^{}



Reed

Velvick

Zutano

Kidd

Dusa

Lab to Nursery



Reed



Velvick



Zutano



Kidd



Dusa

Lab to Nursery – Anderson nursery, Duranbah, NSW



Lab to Nursery



Kidd



Dusa

Morphology trial - Reed

- Ungrafted TC Reed at Anderson Horticulture, Duranbah, NSW
- Planted – September 2017

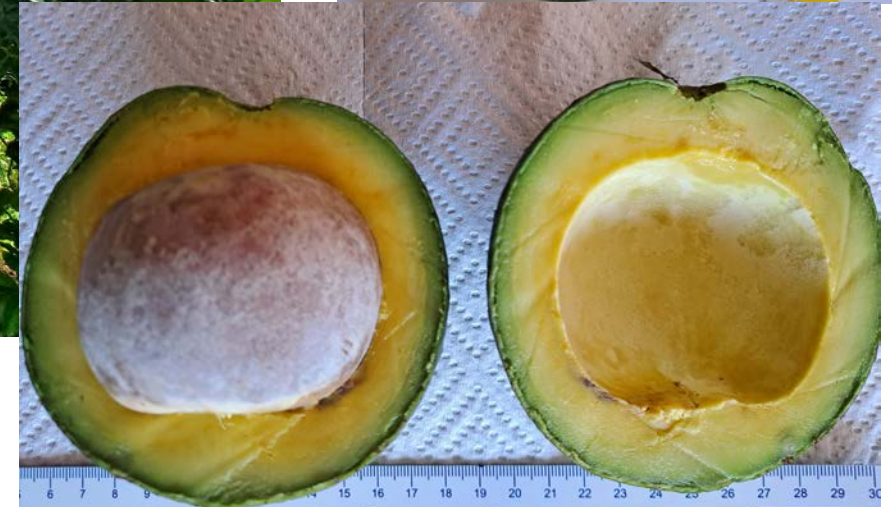


Morphology trial - Velvick

- Ungrafted Velvick at Anderson Horticulture, Duranbah, NSW
- Planted – April 2019



2 years old plants



Morphology trial – Kidd

- Ungrafted Kidd at Anderson Horticulture, Duranbah, NSW
- Planted – April 2019



2 years old plants



Initial Productivity trial - Reed

- Hass Grafted with Reed
- Seedling Reed
- Nursery clonal
- Tissue culture
- At Anderson Horticulture, Duranbah, NSW
- Planted – September 2017
- RCBD, N=10
- Planting density 5 m x 2.5 m

'Reed' seedling



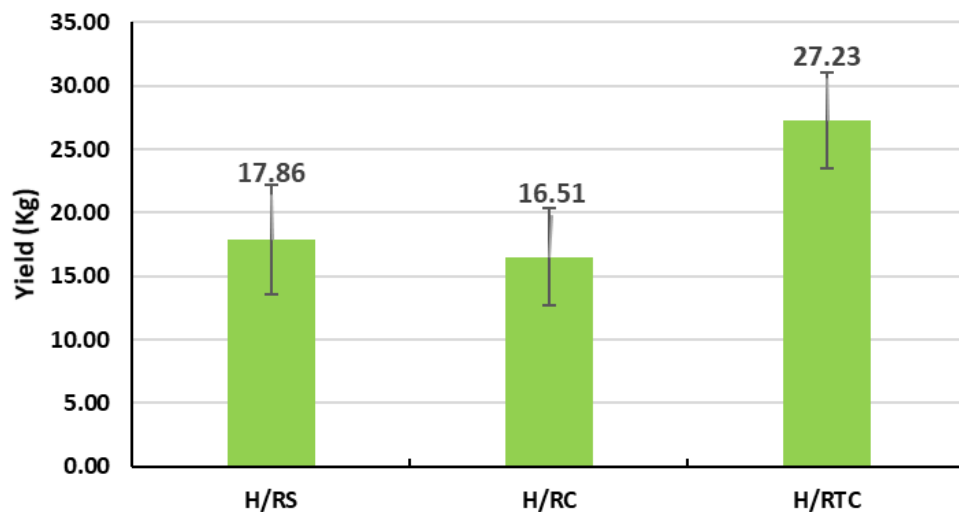
'Reed' clonal



'Reed' tissue cultured



Mean Yield - Year 3





J Hiti Bandaralage, 2019 (Thesis)

Large Productivity trials - Reed

- TC Reed grafted to Hass vs industry standard grafted seedlings

Location	Climate Range (°C)	Avg Annual Rainfall (mm)	Trial Planted date	Trial Design	Establishment Success
Childer's QLD (Donovan Family Investments)	Summer 20.0-30.8 Winter 9.9-21.7	1071	April 2018	43 replicates of Reed seedling and Reed TC Rootstocks. All grafted with Hass scion.	97.6% surviving Reed Seedling 100% surviving Reed TC
Lakeland QLD (Mac Farms)	Summer 24.3 - 32.0 Winter 18.1 - 26.3	1529	September 2018	50 replicates of Reed seedling and Reed TC Rootstocks. All grafted with Maluma scion. These trials are being trained onto a trellis with due central leaders under a shade cloth	96% surviving Reed Seedling 90% surviving Reed TC
Busselton WA (Jasper Farms)	Summer 24.3 - 32.0 Winter 7.5 - 16.3	807	October 2018	50 replicates of Reed seedling and Reed TC Rootstocks. All grafted with Hass scion.	96% surviving Reed Seedling 100% surviving Reed TC
Lakeland QLD (L&R Collins)	Summer 24.3 - 32.0 Winter 18.1 - 26.3	1529	November 2018	50 replicates of Reed seedling and Reed TC Rootstocks. All grafted with Maluma scion.	96% surviving Reed Seedling 90% surviving Reed TC
Pemberton WA	Summer 13.6 - 26.5 Winter 7.2 - 15.2	1185	November 2018	50 replicates of Reed seedling and Reed TC Rootstocks. All grafted with Hass scion.	98% surviving Reed Seedling 100% surviving Reed TC



Dr. Maddy Gleeson

mackays
bananas

Lakeland x2
L & R COLLINS PTY LTD
BANANA PLANTATIONS
ABN: 50 865 836 973
PO Box 498
TULLY QLD 4854

Childers

DFI
DONOVAN FAMILY INVESTMENTS
Quality, Consistency, Reliability

Busselton

- JASPER FARMS -

Pemberton

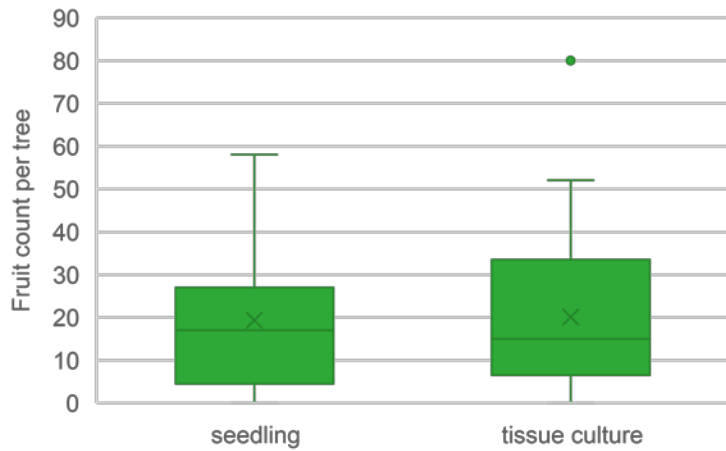
DELROY ORCHARDS
taste the deep south of Western Australia

Production Year 3 - Childers

<https://qaafi.uq.edu.au/files/72009/Tissue%20Culture%20for%20Australian%20Avocados%20-%20PIP.pdf>

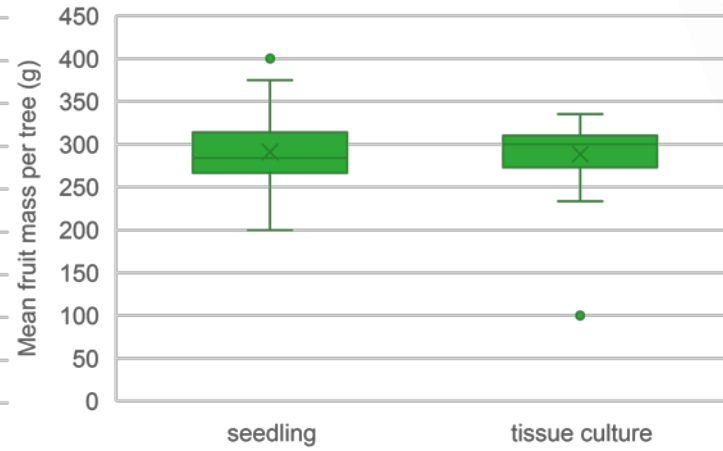
no difference

Fruit Count



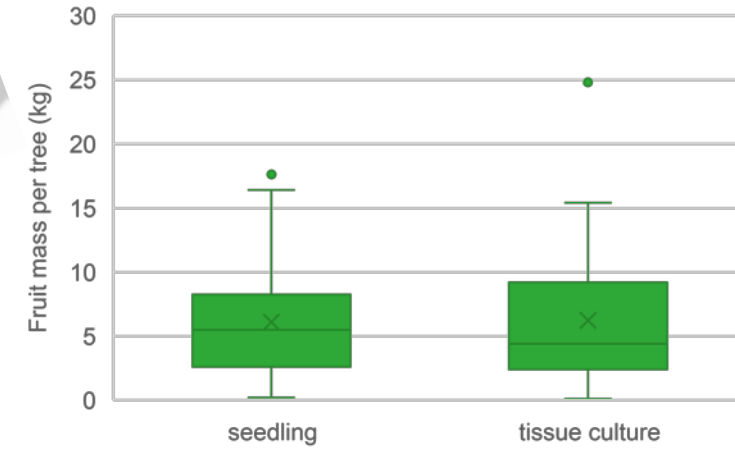
no difference

Mean Fruit Mass



no difference

Fruit Yield



Technology commercialisation

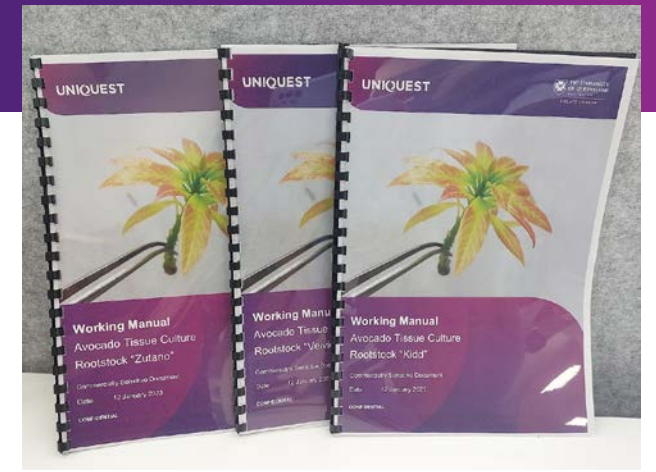
NEWS

Smashing avocado know-how

The biggest industry bottleneck for avocado production could soon be a thing of the past with the signing of a licence agreement between The University of Queensland's commercialisation company UniQuest and Anderson Horticulture Pty Ltd.

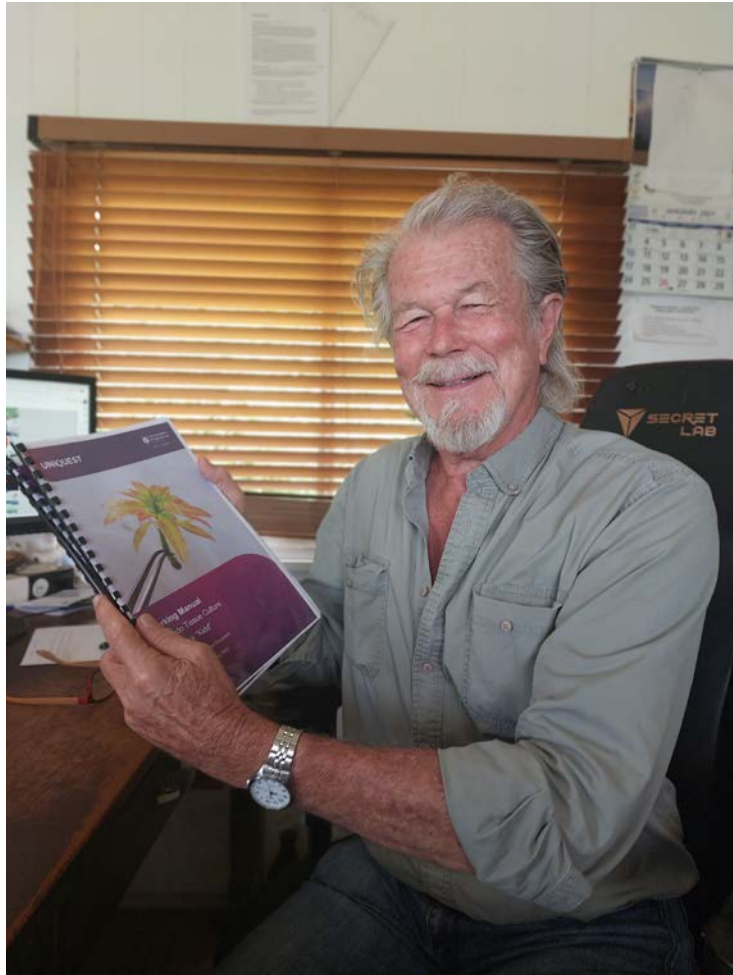
UQ researchers have developed a world-first commercial-scale tissue culture propagation technology for Reed avocado rootstock





Technology commercialisation

- Reed – Late 2020
- Velvick, Kidd and Zutano – Early 2023



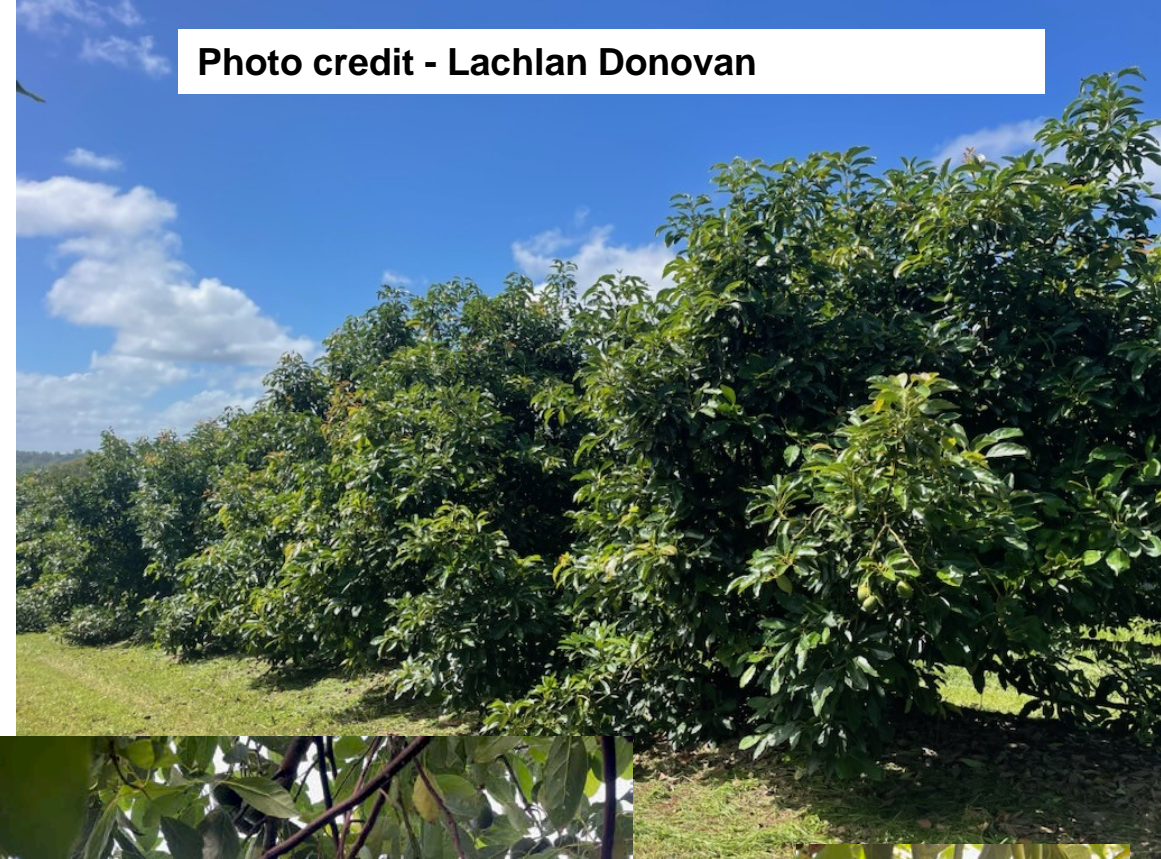
Commercial avocado TC is not a dream



TC rootstock graft success 90 - 98% →

Commercial orchard – Hass/TC Reed

Givelda farm, Donovan family investments
200 trees



Smart & sustainable Avocado propagation system Commercialised

- Production capacity of 500,000 plants using only 400 cuttings, 1 bud to 200-500 plants
- 10,000 plants in 10 sq meters



Another reason to call **Avocado** is



Let's toast ... Thank you...

