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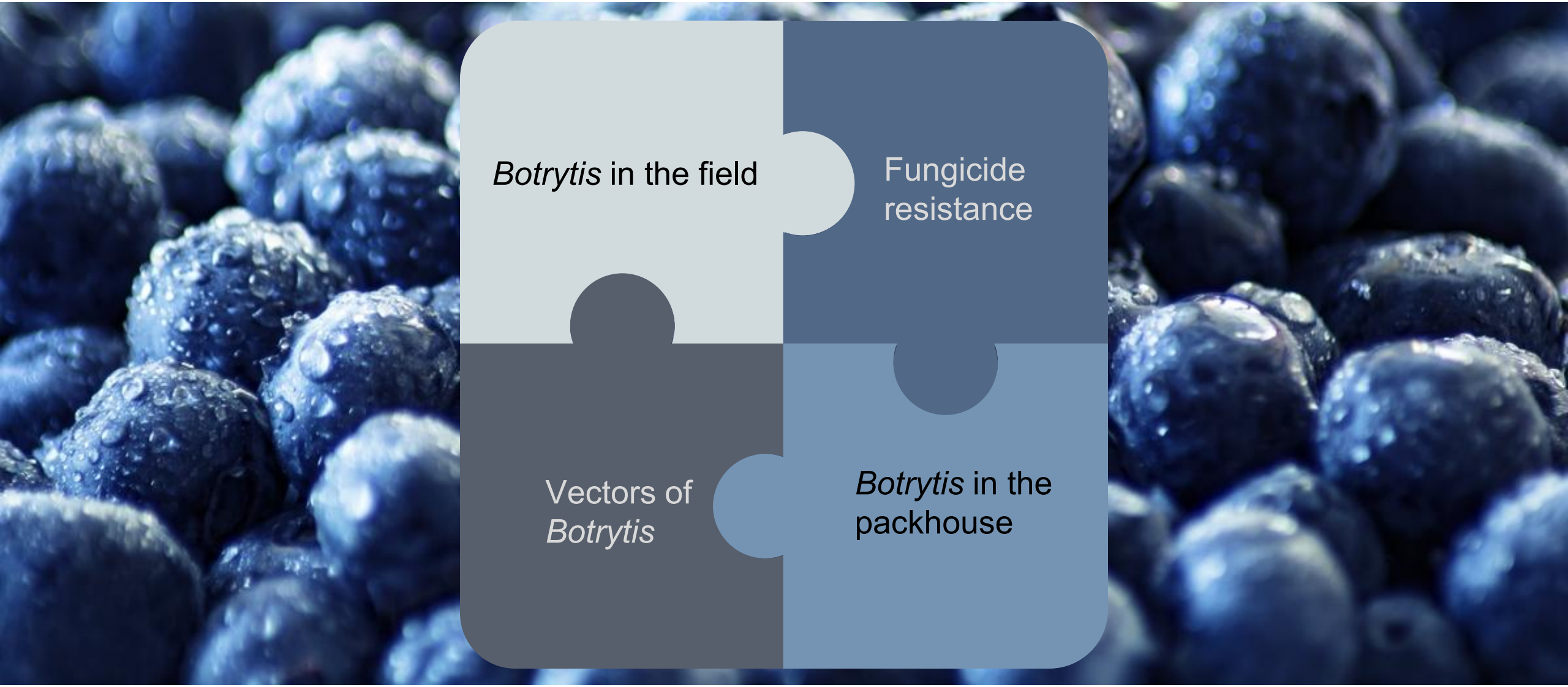
The relationship between blueberries and fungi: it is Still complicated!

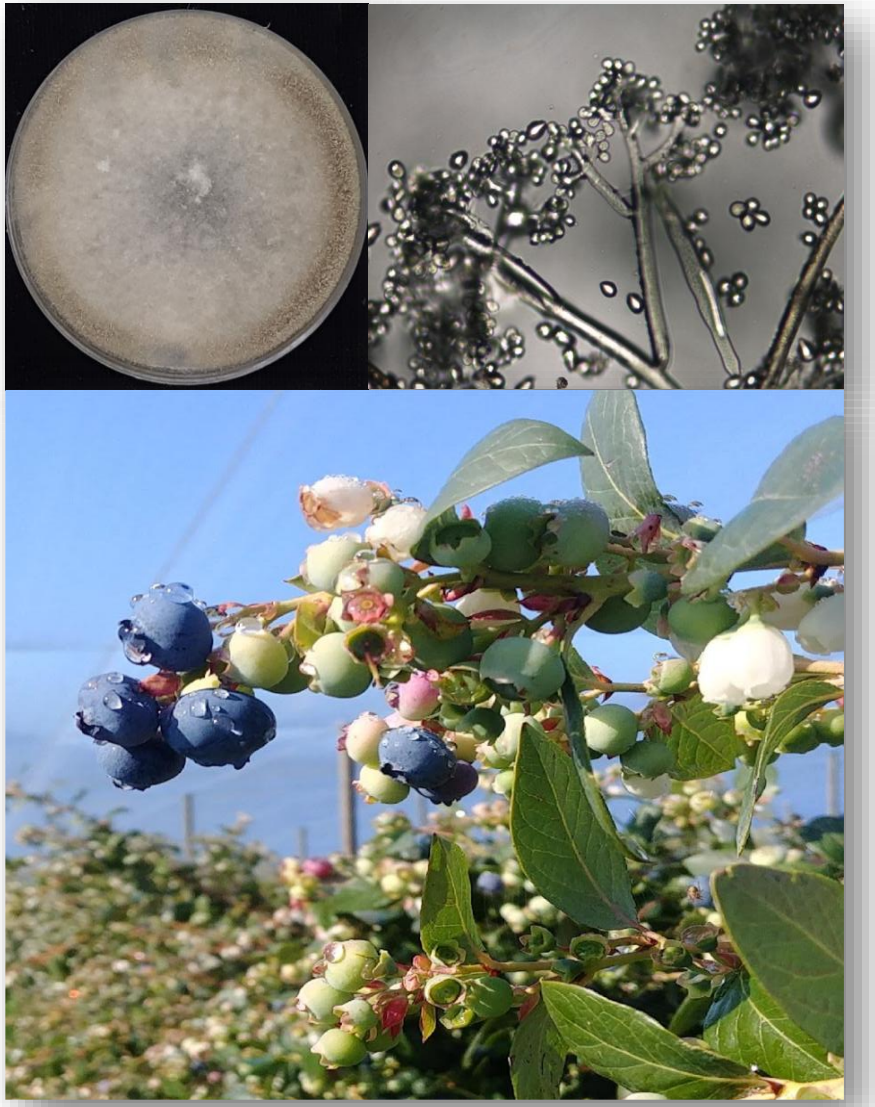
Karin Jacobs

Photo by Rawpixel

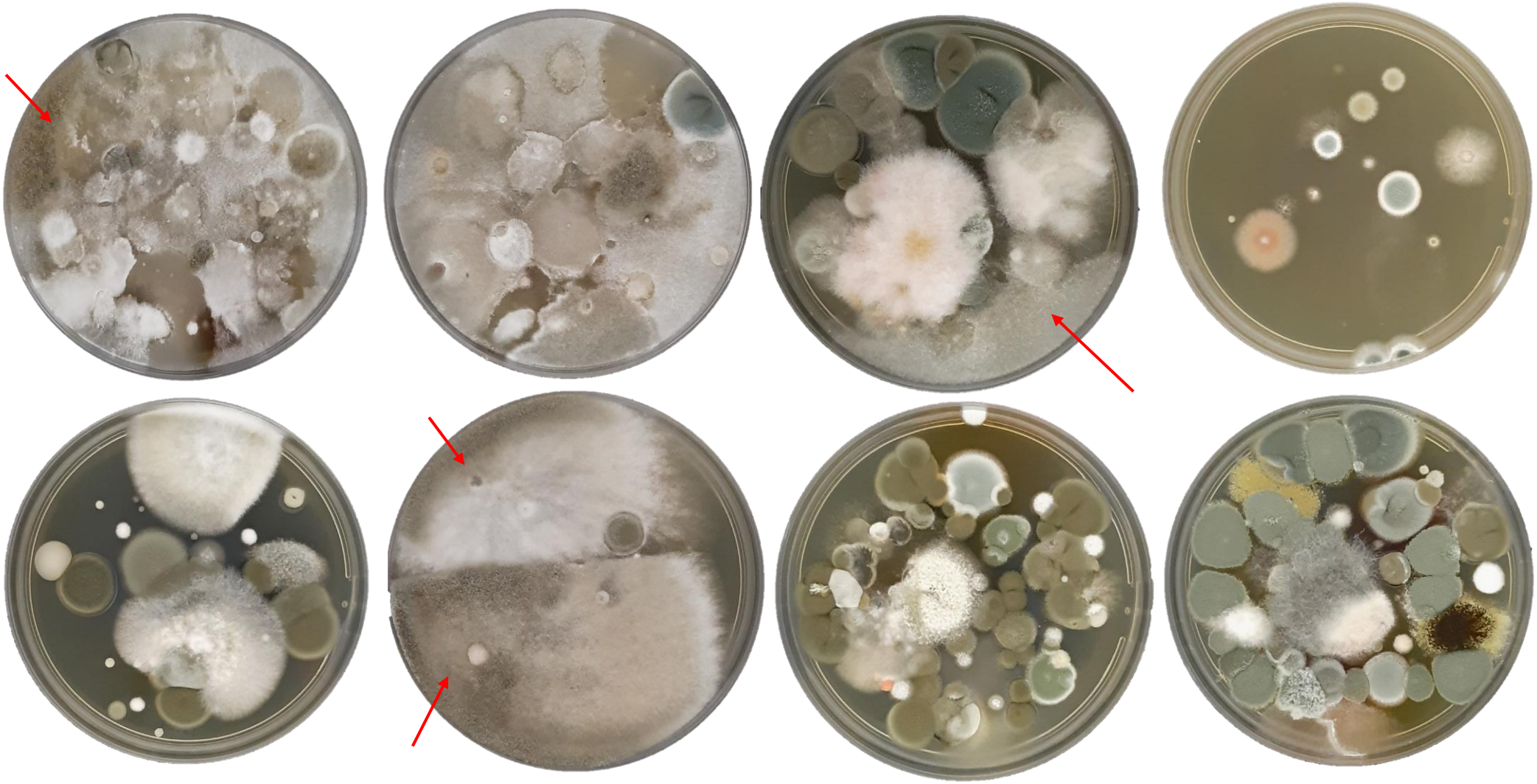

Botrytis in the field
Fungicide resistance
Vectors of *Botrytis*
Botrytis in the packhouse

From previous studies…

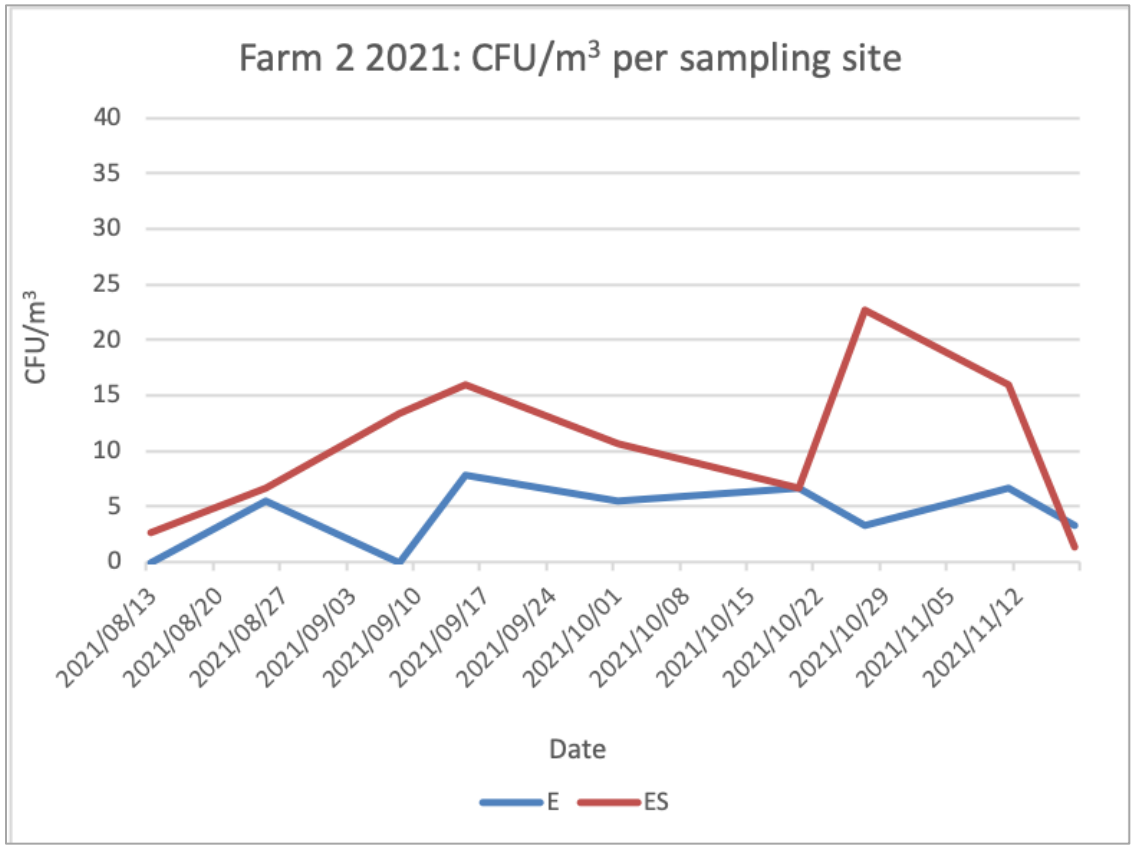
- *B. cinerea* is prevalent on blueberries
- No evidence that *B. cinerea* is an endophyte
- Most above-ground blueberry tissues are susceptible to infection by *B. cinerea*
- Majority of the strains are pathogenic

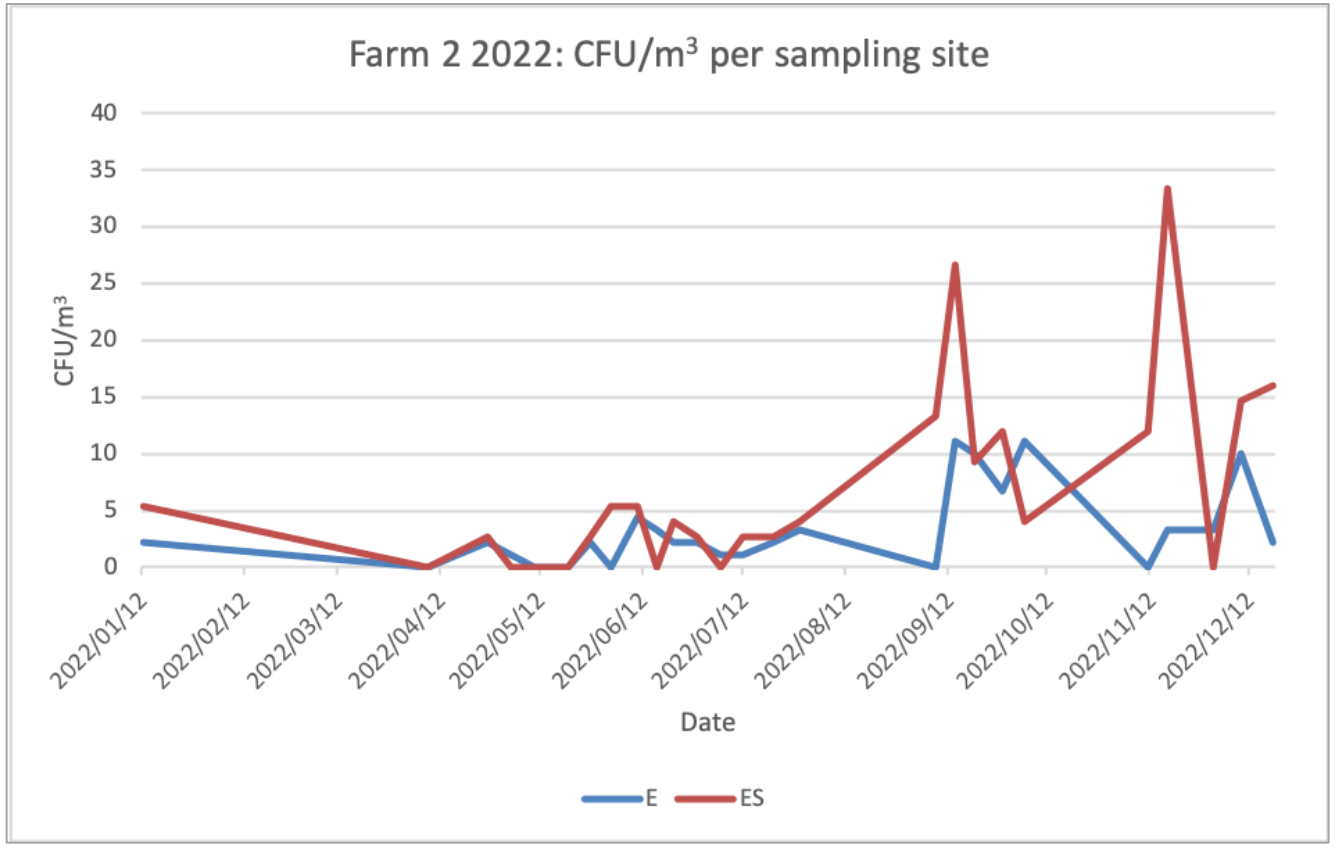


Airborne *Botrytis* spore load



Airborne *Botrytis* spore load





Take-home message

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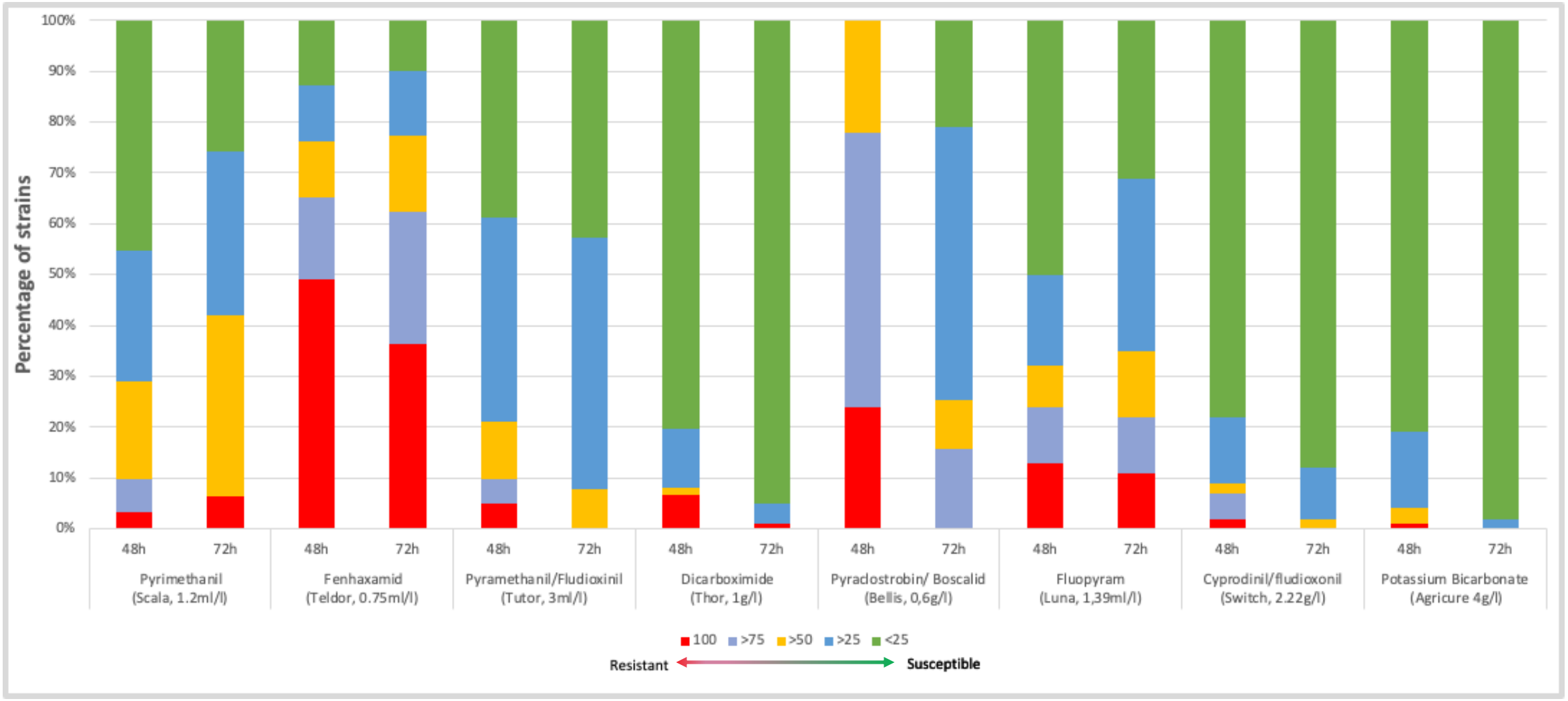
1. *Botrytis* in blueberry orchards vary over a season and increase as the berries ripen.

Fungicide resistance



Take-home message

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1. There are some fungicide resistance in orchards.
2. Cyprodinil/fludioxonil (Switch) and Potassium Bicarbonate (Agricure) still provide good protection.
3. Fungicide resistance should be monitored.

The mycobiome of blueberry-foraging Western Cape honeybees (*Apis mellifera capensis*)

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- Support biodiversity
- Food safety
- Economic impact
- Environmental indicators
- Face several challenges

Brief Report | Open access | Published: 09 January 2023

Beehives possess their own distinct microbiomes

Lorenzo A. Santorelli, Toby Wilkinson, Ronke Abdulmalik, Yuma Rai, Christopher J. Creevey, Sharon Huws & Jorge Gutierrez-Merino

Environmental Microbiome **18**, Article number: 1 (2023) | Cite this article

Review > Microorganisms. 2022 Nov 29;10(12):2359. doi: 10.3390/microorganisms10122359.

Micro"bee"ota: Honey Bee Normal Microbiota as a Part of Superorganism

Daniil Smutin [1 2], Egor Lebedev [1], Maxim Selitskiy [1], Nick Panyushev [1], Leonid Adonin [1 3]

Affiliations + expand

PMID: 36557612 PMCID: PMC9785237 DOI: 10.3390/microorganisms10122359

The Bee Microbiome: Impact on Bee Health and Model for Evolution and Ecology of Host-Microbe Interactions

Philipp Engel [a,✉], Waldan K Kwong [b,c], Quinn McFrederick [d], Kirk E Anderson [e], Seth Michael Barribeau [f], James Angus Chandler [g,*], R Scott Cornman [h], Jacques Dainat [i], Joachim R de Miranda [j], Vincent Doublet [k,l], Olivier Emery [a], Jay D Evans [m], Laurent Farinelli [n], Michelle L Flenniken [o], Fredrik Granberg [p], Juris A Grasis [q], Laurent Gauthier [a,b], Juliette Hayer [r], Hauke Koch [c,s], Sarah Kocher [t], Vincent G Martinson [u], Nancy Moran [c], Monica Munoz-Torres [v], Irene Newton [w], Robert J Paxton [k,l], Eli Powell [c], Ben M Sadd [x], Paul Schmid-Hempel [y], Regula Schmid-Hempel [y], Se Jin Song [z], Ryan S Schwarz [m], Dennis vanEngelsdorp [aa], Benjamin Dainat [ab,ac,✉]

▸ Author information ▸ Article notes ▸ Copyright and License information

PMCID: PMC4850275 PMID: 27118586

JOURNAL ARTICLE EDITOR'S CHOICE

Bees just wanna have fungi: a review of bee associations with nonpathogenic fungi

Danielle Rutkowski ✉, Makena Weston, Rachel L Vannette

FEMS Microbiology Ecology, Volume 99, Issue 8, August 2023, fiad077, https://doi.org/10.1093/femsec/fiad077

Published: 08 July 2023 **Article history** ▾

From literature

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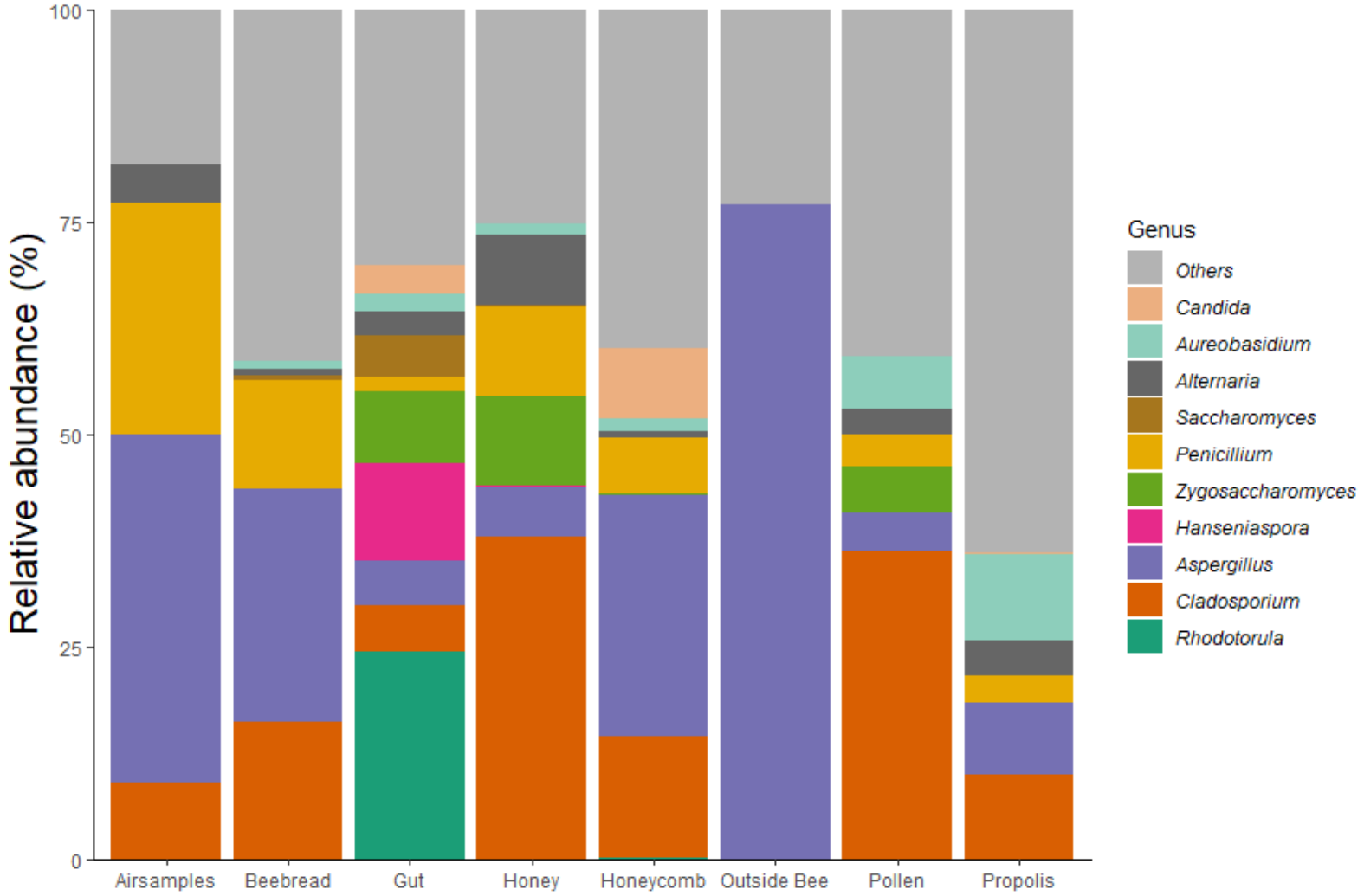
Relative abundance of the top 10 genera across different parts of the bee mycobiome from all reviewed studies.

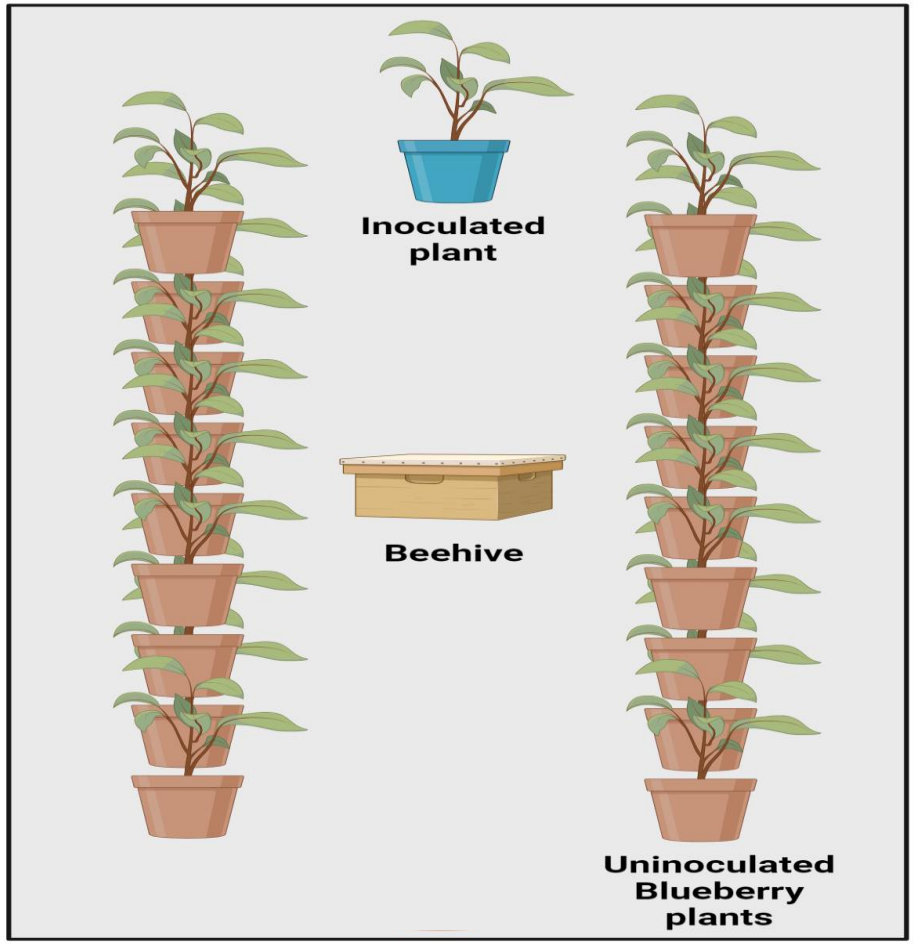


Fungi on bees

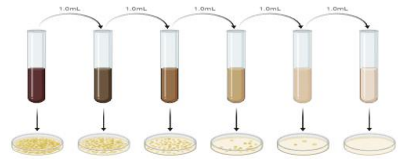
Gregory, 1941; de Vries, 1952; Staats *et al.*, 2005; Bench *et al.*, 2012

Can bees spread *Botrytis*?

Greenhouse experiment



UV search



Dilution series and plating out

Rose,1996; Edwards and Seddon, 2001; Bullinger *et al.*, 2023

Can bees spread *Botrytis*?

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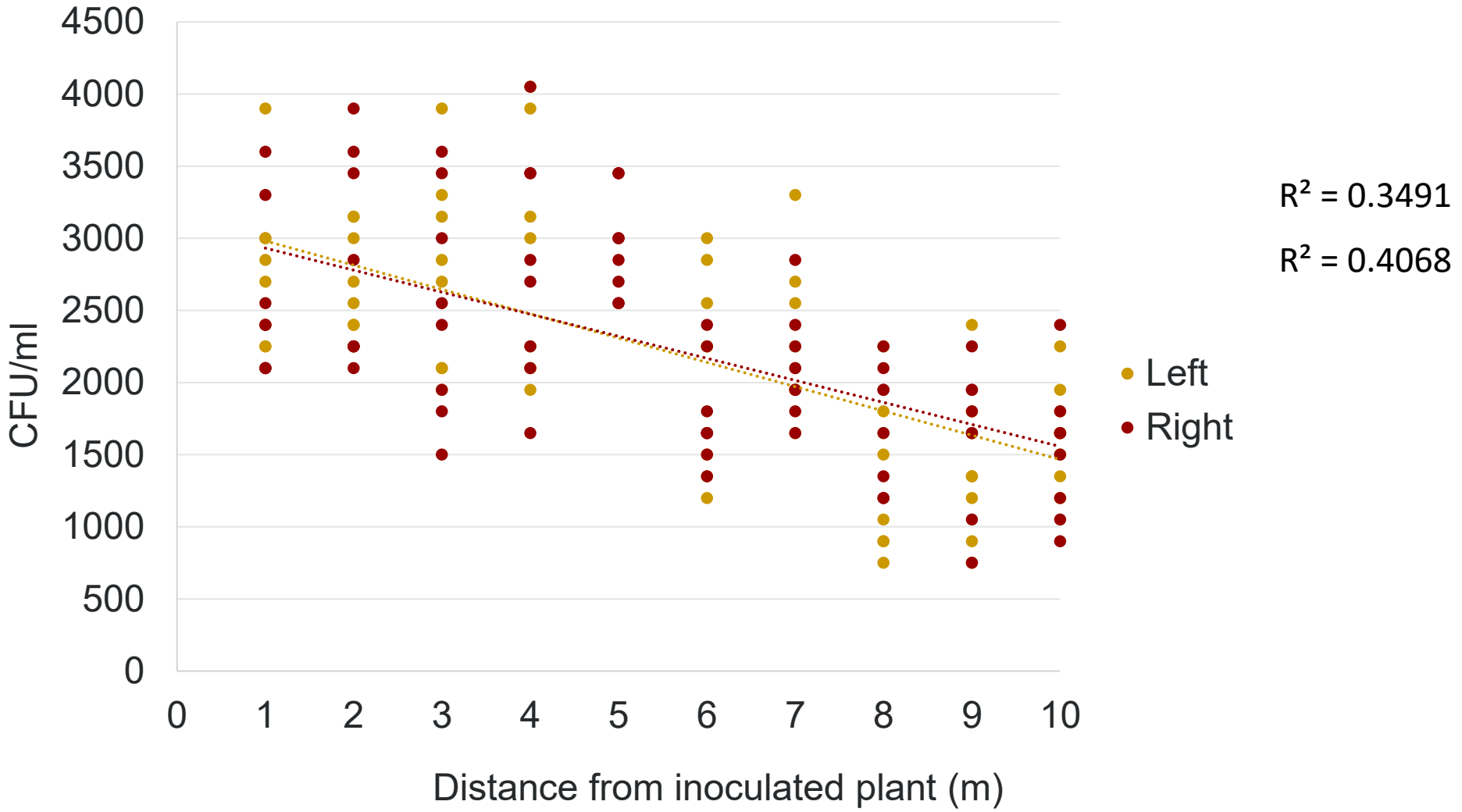
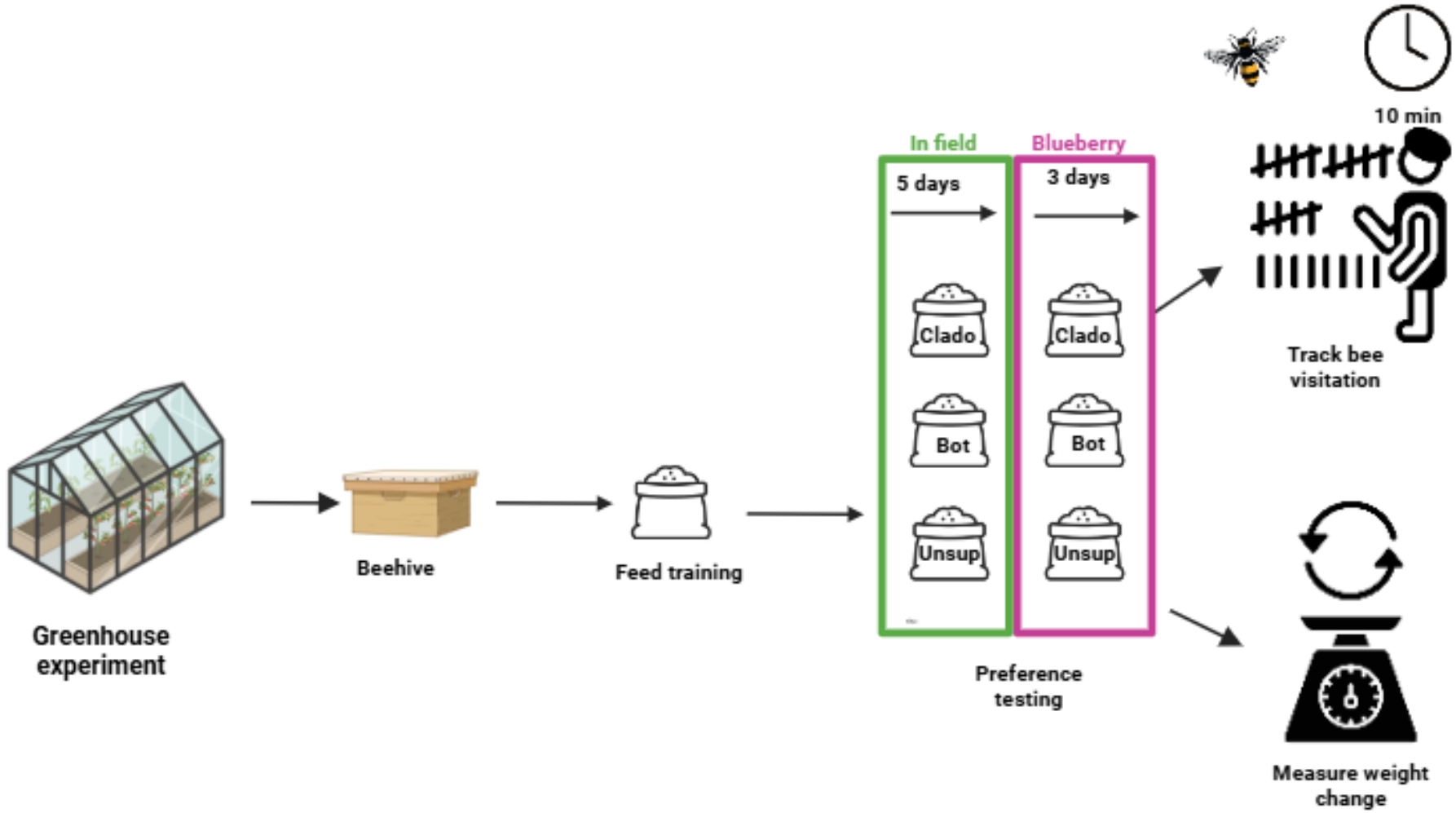
Can bees spread *Botrytis*?

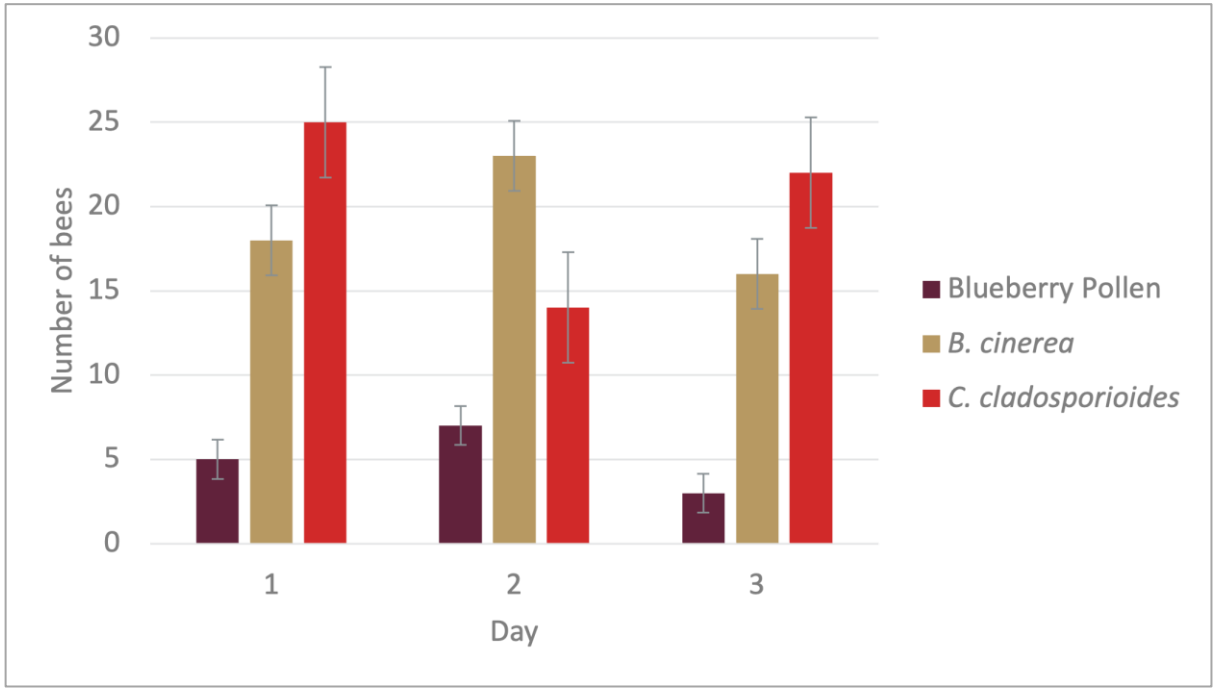


Feed preference

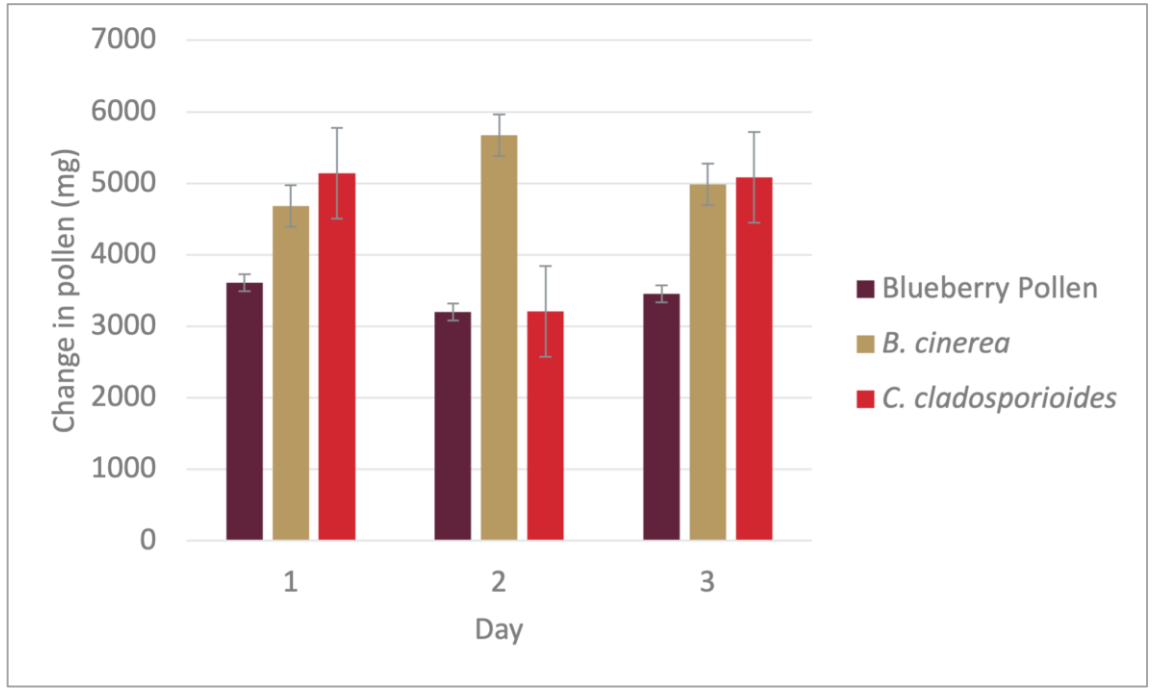


Arenas *et al.*, 2021; Kim *et al.*, 2024

Do bees like fungi?



Changes in the number of bees present at different food sources



Changes in pollen weight (mg) from different food sources

pollens can be one of their important source. The essential amino acids for honeybees are arginine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan and valine (De Groot, 1953). The concentrations of essential amino acids in bee-pollens, expressed as the percentage of the total amino acids ranged from 34.59%

The pollen harvested in June 2016 at the blueberry apiary (blueberry management strategy and/or double management strategy) had significant differences in five essential amino acids (arginine ($F(3,103) = 23.11$, $P < 0.001$), lysine ($F(3,102) = 9.14$, $P < 0.001$), tryptophan ($F(3,103) = 12.57$, $P < 0.001$), methionine ($F(3,103) = 8.57$, $P < 0.001$), and isoleucine ($F(3,103) = 4.75$, $P < 0.01$)) compared to pollen harvested at the farmland apiary (control management strategy

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Article | Open access | Published: 24 September 2020

Nutritional benefit of fungal spores for honey bee workers

Jorgiane B. Parish, Eileen S. Scott & Katja Hogendoorn

Scientific Reports **10**, Article number: 1

4926 Accesses | **22** Citations | **4** Al

Original Articles

The Incidental Collection of Fungal Spores by Bees and the Collection of Spores in Lieu of Pollen

Dorothy E Shaw

Pages 158-176 | Published online: 01 Apr 2015

Cite this article https://doi.org/10.1080/0005772X.1990.11099059

New insights into the regulation of plant immunity by amino acid metabolic pathways

Jürgen Zeier [1]

Affiliations + expand

PMID: 23611692 DOI: 10.1111/pce.12122

Free article

SOS – too many signals for systemic acquired resistance?

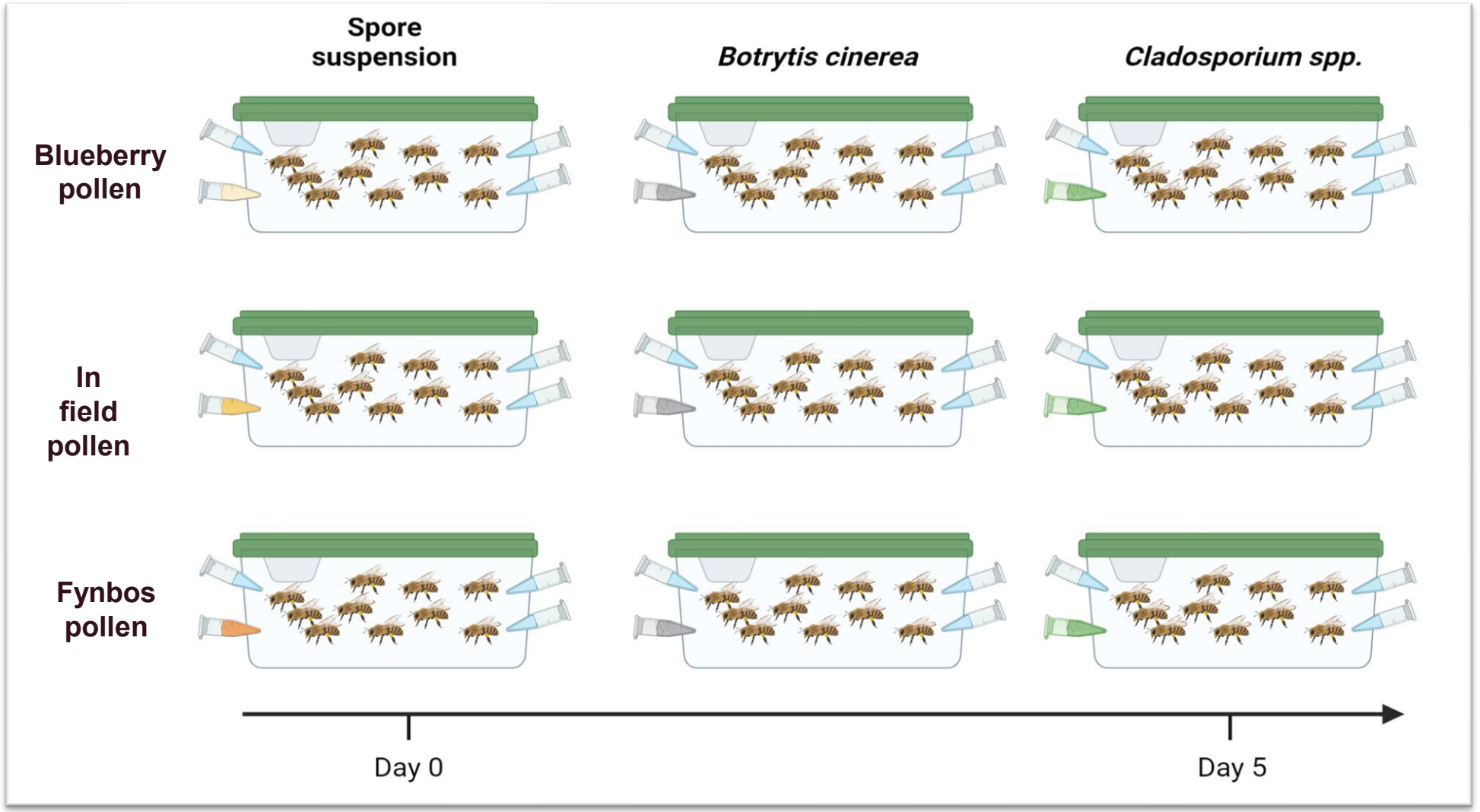
D'Maris Amick Dempsey [1], Daniel F Klessig

Affiliations + expand

PMID: 22749315 DOI: 10.1016/j.tplants.2012.05.011

Full text links Cite

The experiment



Huang *et al.*, 2014

How did we test this

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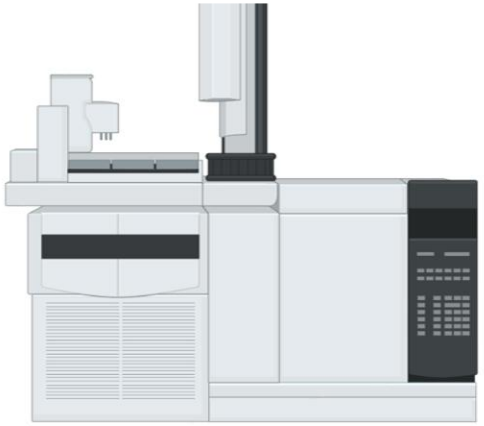
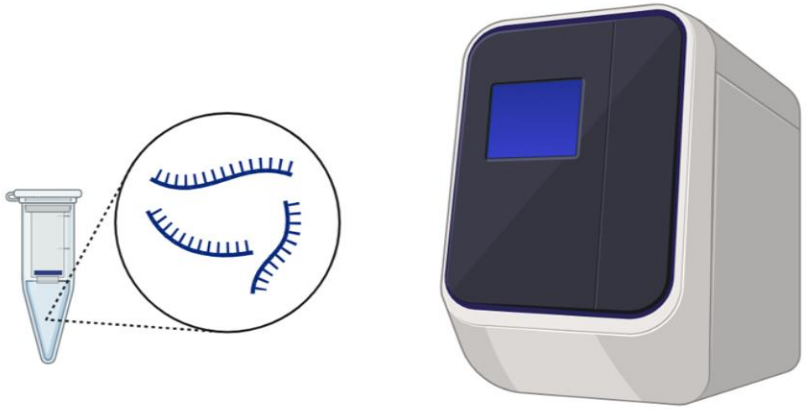
Pollen was subjected to GCMS

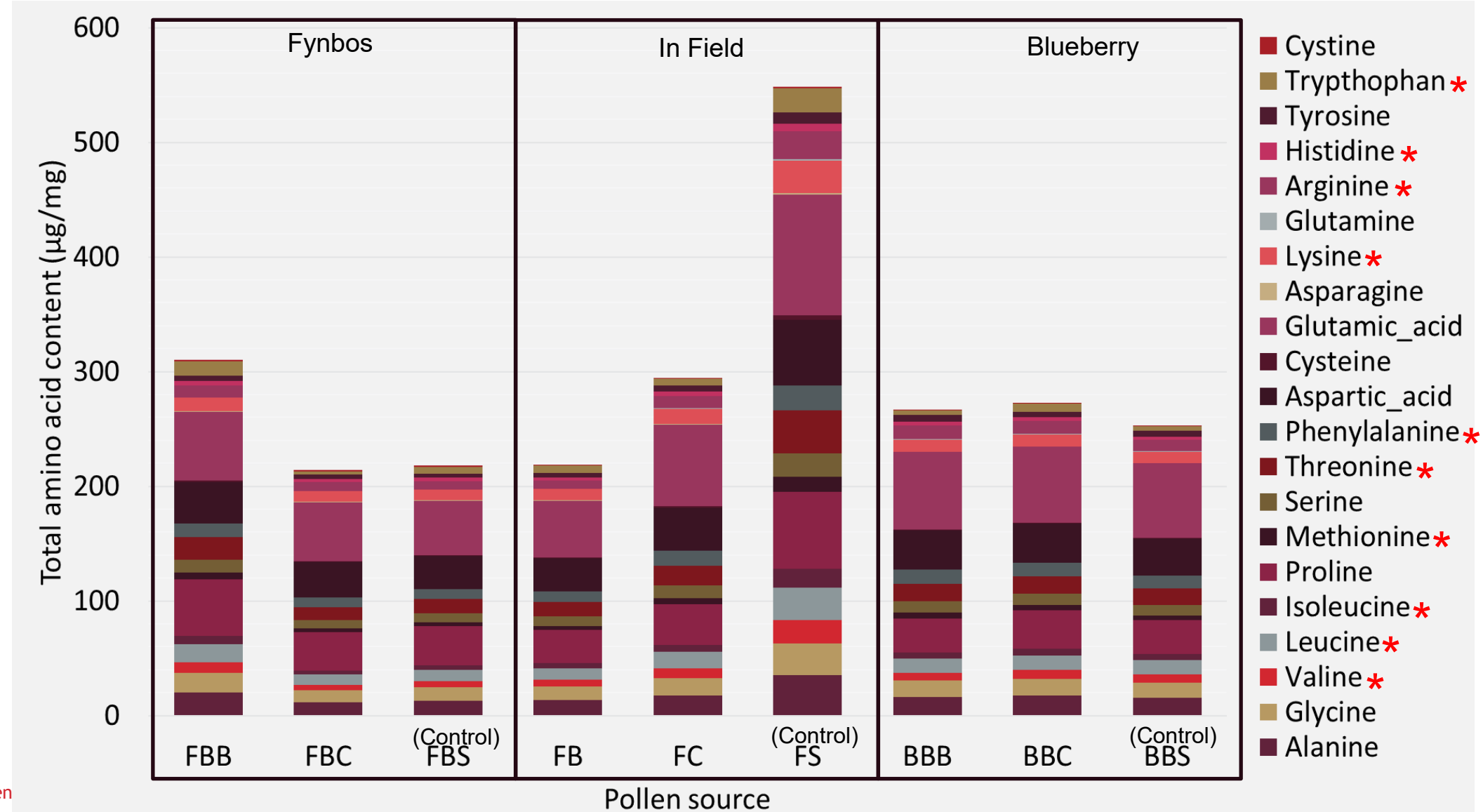


Honeybees were subjected to RNA extraction and qPCR



Kitson *et al*. 1996; Molnar-Perl and Katona, 2000; Sobolevsky *et al*.,2003; Matyash *et al*., 2008; Stenerson *et al*., 2011, Proestos *et al*., 2013; Conradie *et al*., 2024; Brutscher *et al*., 2015.

Amino acid analysis



Scien

Amino acid analysis

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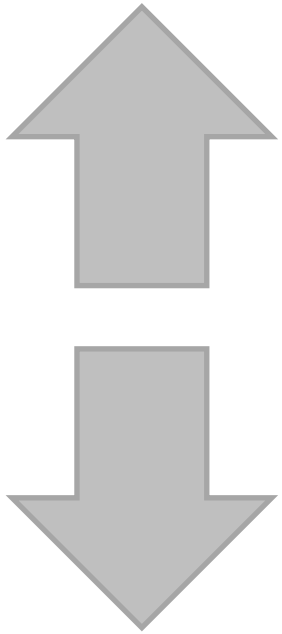
		Total amino acid concentration
Fynbos (Native)	*B. cinerea*	↑
	C. cladosporioides	↓
In field (Realised)	*B. cinerea*	↓
	C. cladosporioides	↓
Blueberry (Experimental)	*B. cinerea*	↑
	C. cladosporioides	↑

Increased amino acid availability → enhanced nutrition and physiological support



Reduced amino acid levels limit availability for honeybee use → potentially weakening bees

Gene regulation

↑ Significantly upregulated

↓ Significantly downregulated

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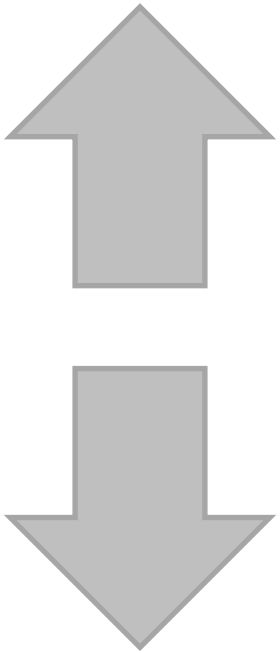
		Vitellogenin (vg)	*Defensin-2 (def2)*	*Prophenoloxidase (proPO)*	*Immune deficiency (Imd)*	*Spaetzle (spz)*
Fynbos (Native)	*B. cinerea*	↑	↑	↑	↑	↑
	C. cladosporioides	↑	↑	↑	↑	↑
In field (Realised)	*B. cinerea*	↑	↓	↑	↑	↑
	C. cladosporioides	↑	↑	↓	↑	↑
Blueberry (Experimental)	*B. cinerea*	↑	↑	↓	↑	↓
	C. cladosporioides	↓	↑	↑	↑	↓

Healthy bee → normal lifecycle, adequate nutrition



Stressed bees → pressure to forage, trade-off between immune activation and nutrition

Take-home message

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1. Bees are able to spread pathogens.
2. Fungal spores can increase the nutritional value of pollen.
3. The study challenges reductionist views and highlights interconnectedness of pollinators, pathogens, and agricultural systems.

Fungi within blueberry packhouses

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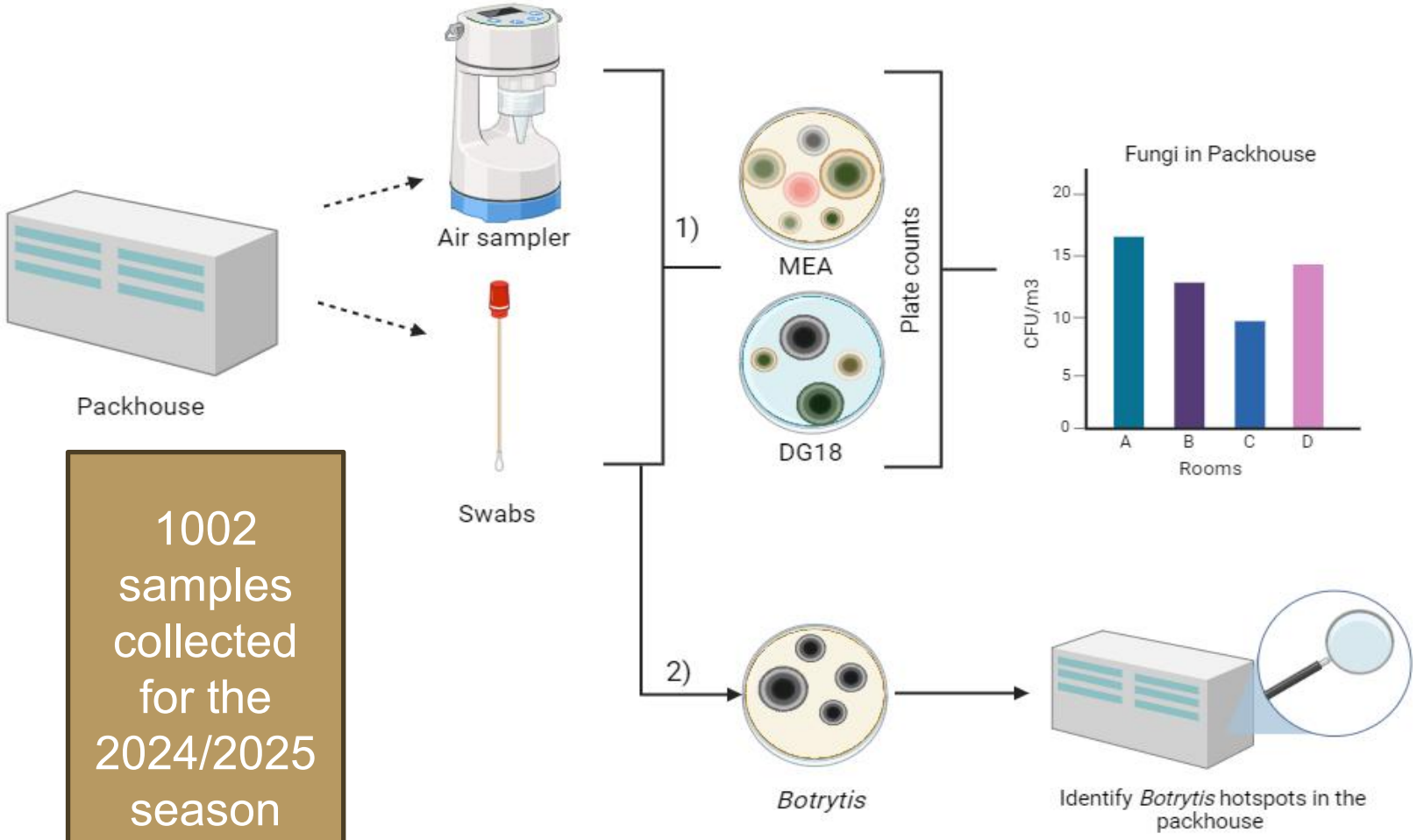
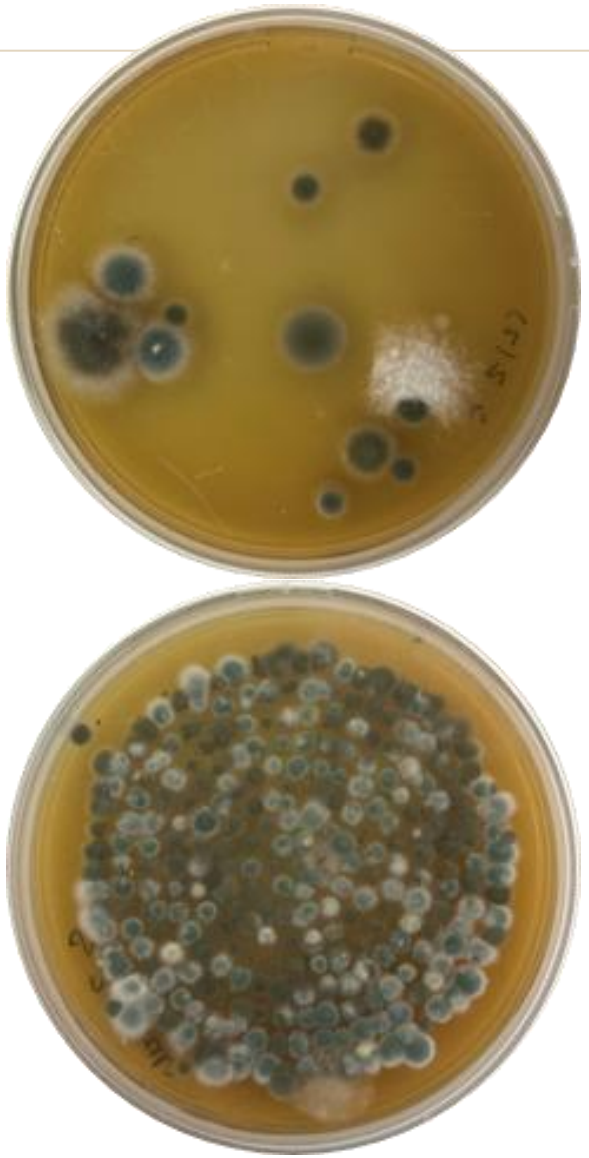


1002 samples collected for the 2024/2025 season



Fungi within blueberry packhouses

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- General threshold 1000 CFU / m^3
  - Global discrepancies
  - 200-300 CFU / m^3 often set for sterile environments

- Indoor to Outdoor (I/O) ratio
  - Indicative of favourable fungal survival conditions
    - *Acceptable*: > 1
    - *Poor*: < 2

Characterisation of the fungal population in citrus packing houses

Published: 23 October 2008

Volu

Identification of fungal population in the environment and on surfaces of stone fruit packinghouses

Published: 06 December 2016

Volume 148

ELSEVIER

Postharvest Biology and Technology

Volume 128, June 2017, Pages 153-160

Penicillium air mycoflora in postharvest fruit handling environments associated with the pear export chain

Ilonka Scholtz, Nazareth Siyoum, Lise Korsten

B. cinerea presence over time

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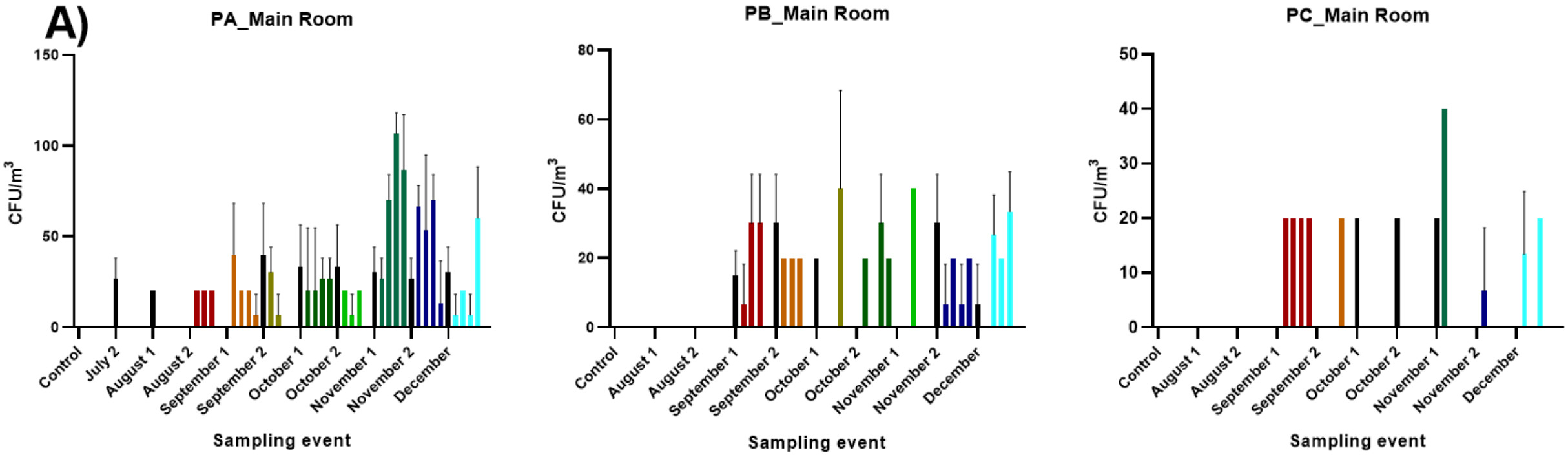

Fungi in the Main Packing Areas

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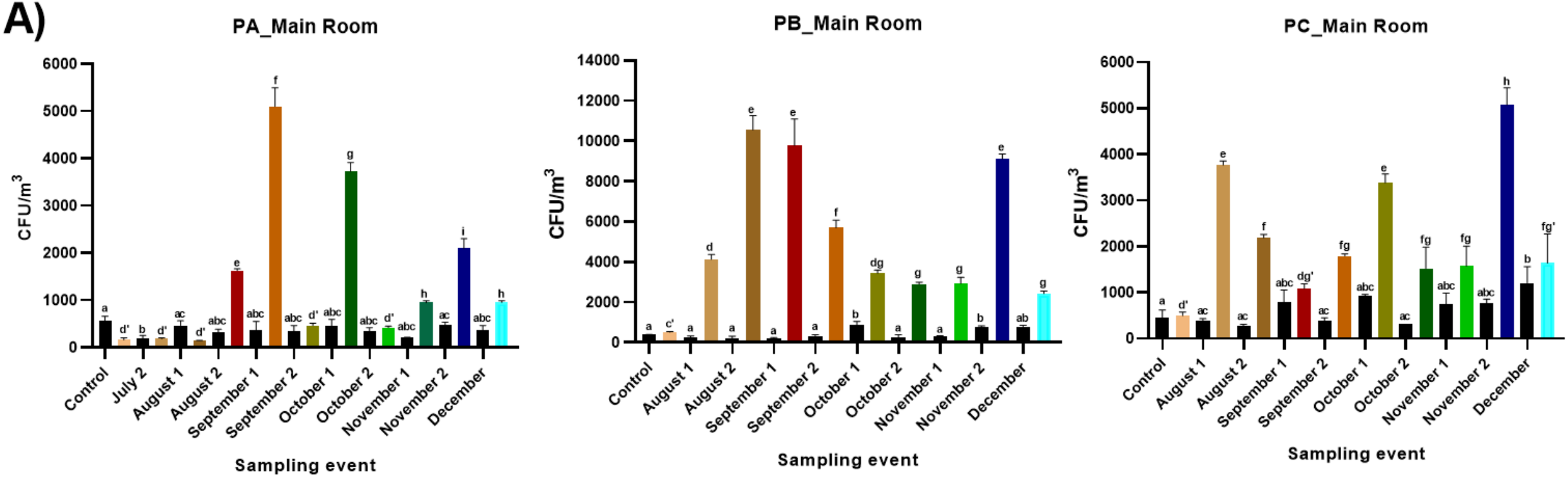
A)



Fungi in the Main Packing Areas

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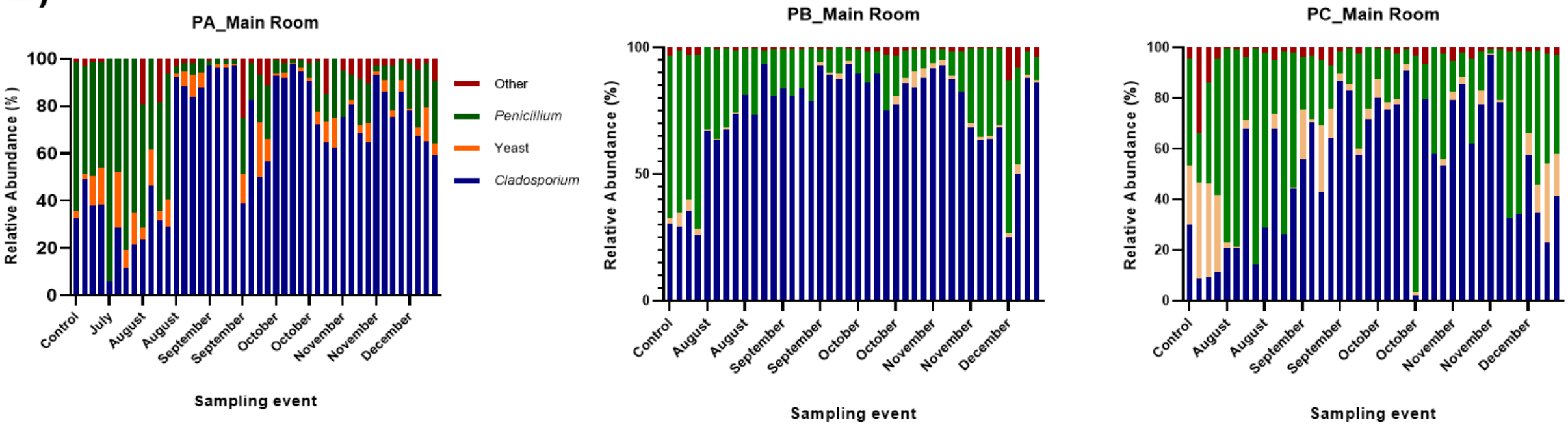
B)



Known postharvest pathogens + human allergen producers

I/O ratios

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	Intake Room	Cooling Room	Holding Room	Main Room	Final Cooling Area	Dispatch Room
July	1.2	1.2	1.6	1.4	1.8	1.6
August	0.9	5.4*	5.4*	17.7*	24.5*	16.7*
August	0.7	3.9*	2.5*	53.1*	48.2*	27.3*
September	0.4	7.6*	2.8*	55.1*	39.1*	3.1*
September	0.1	0.3	1.0	17.8*	16.3*	0.5
October	0.1	0.0	1.0	4.3*	5.3*	0.3
October	0.3	0.5	0.9	13.0*	17.3*	0.2
November	1.2	1.2	0.6	10.7*	17.0*	3.2*
November	0.0	0.0	0.1	12.3*	6.4*	1.5
December	0.2	0.0	0.1	4.5*	6.7*	0.1

Fungi within blueberry packhouses

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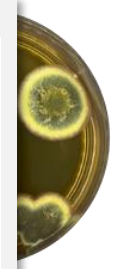
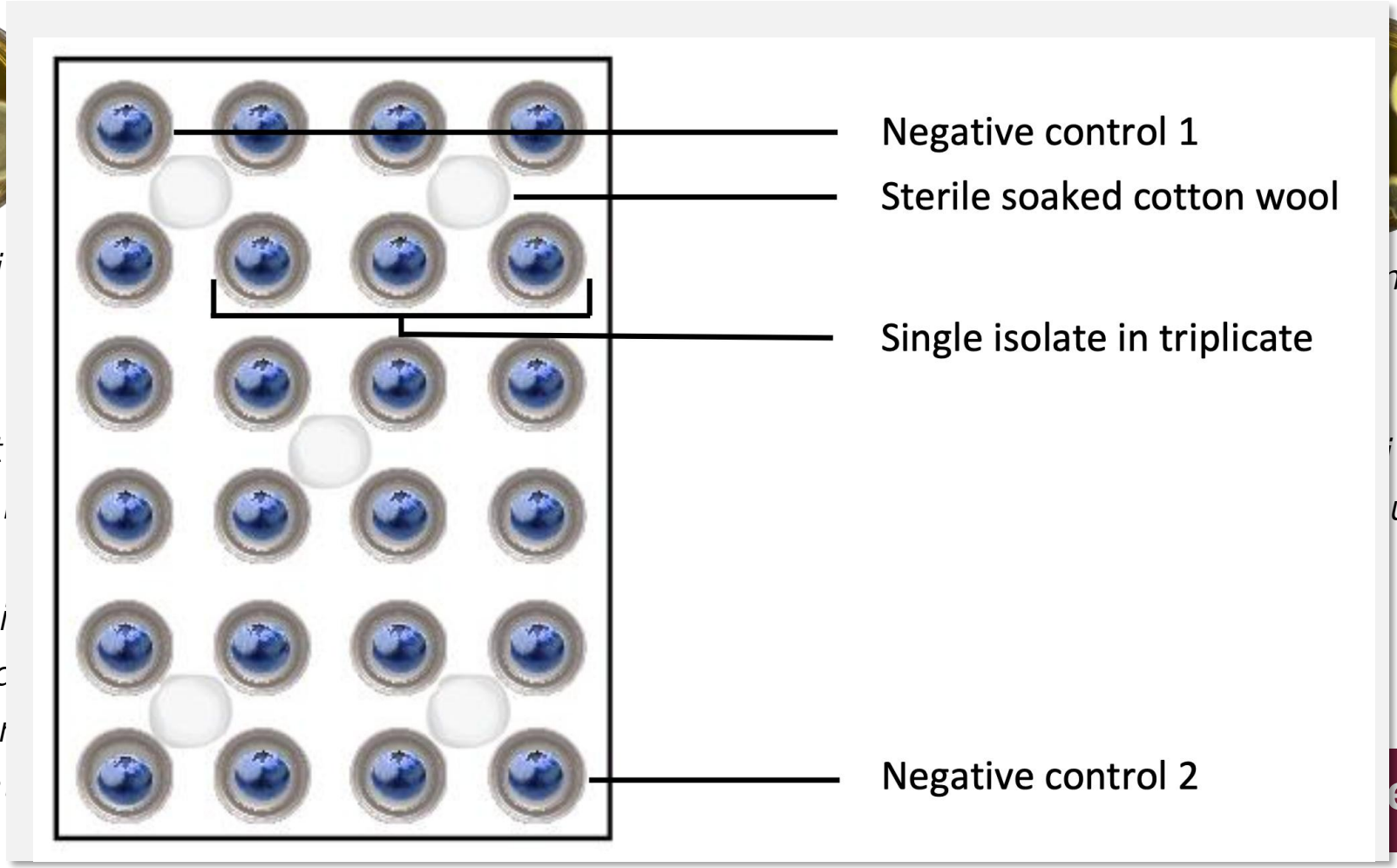
Cladospori



nyces

Trichoderma



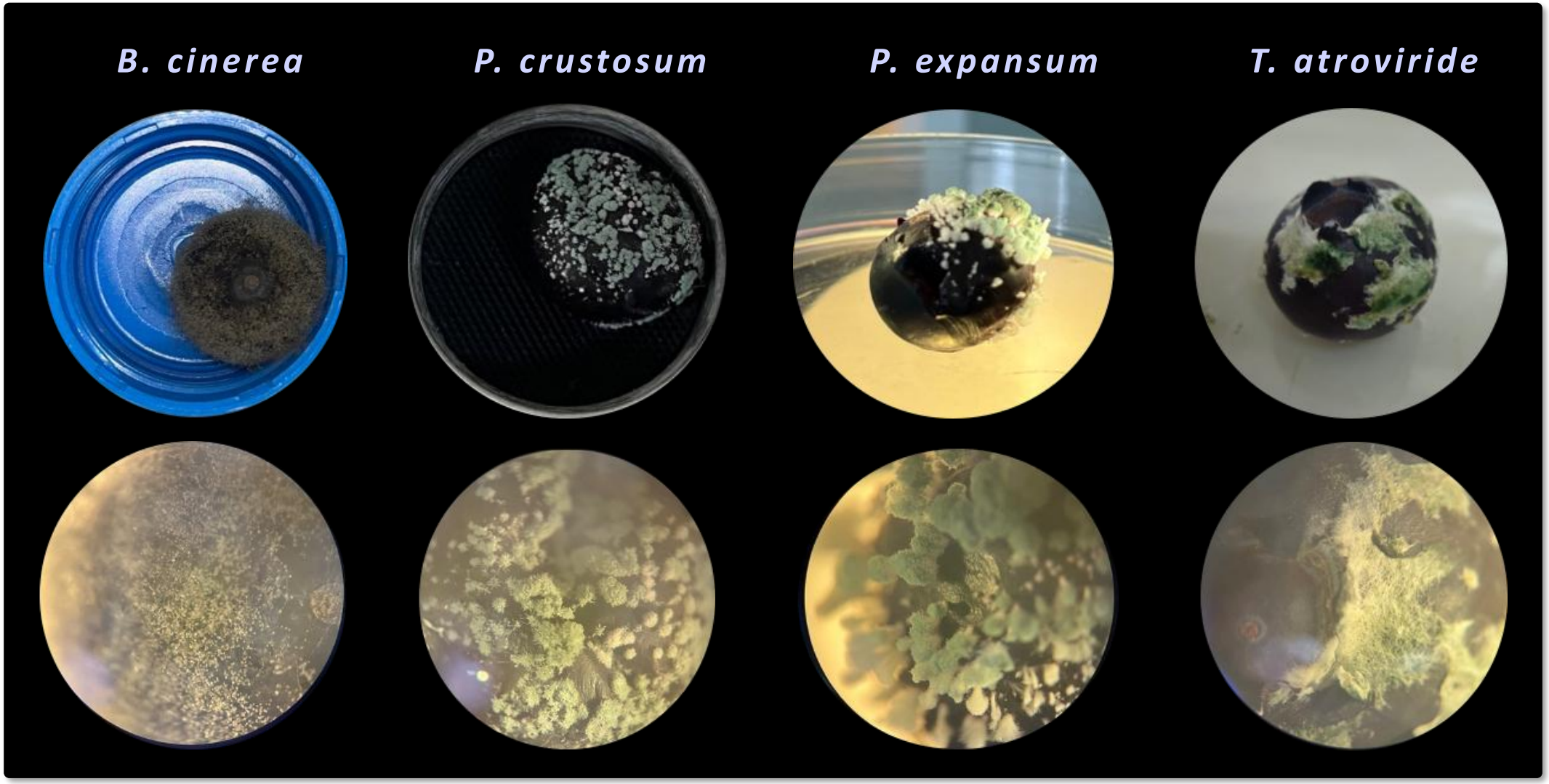
C. asperulat
C. europaeu
C. pseudo-
cladosporioi
C. proteaced
C. cladospor
C. ramotene
C. tenellum

iverticillius
uteus

T. atroviride

ecies, 6 genera

Pathogenicity Trials



Pathogenicity Trials

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Pathogenicity Status						
	Day 5				Day 16	
Isolate	4 °C	8 °C	26 °C	30 °C	4 °C	8 °C
B. cinerea B1	✗	✗	✗	✗	✗	✓
B. cinerea B4	✗	✗	✓	✗	✗	✗
B. cinerea B5	✗	✗	✓	✗	✗	✗
P. expansum P1	✗	✗	✓	✗	✗	✓
P. expansum N10	✗	✗	✓	✗	✗	✗
P. crustosum P12	✗	✗	✓	✗	✗	✗
P. crustosum P13	✗	✗	✓	✗	✗	✗
T. atroviride N8	✗	✗	✓	✗	✗	✗

Human Health Assessment

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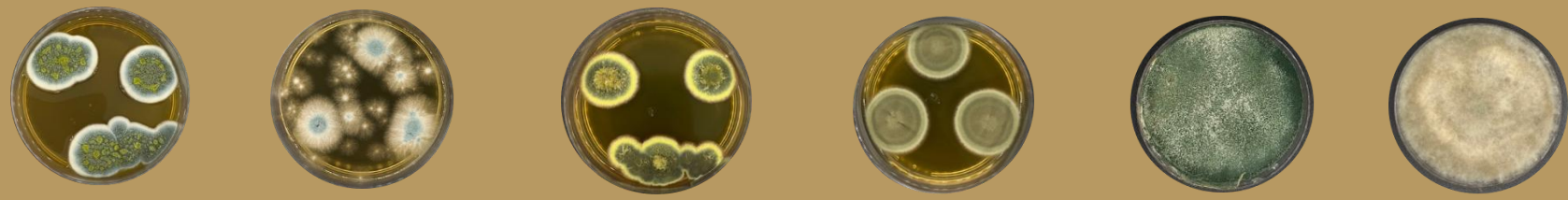
Genus level:



Penicillium *Aspergillus* *Talaromyces* *Cladosporium* *Trichoderma* *Botrytis*

Allergenic

Allergenic, toxigenic & opportunistically pathogenic

Species level:

P. expansum *C. cladosporioides* *A. fumigatus* *A. flavus*

Take-home message

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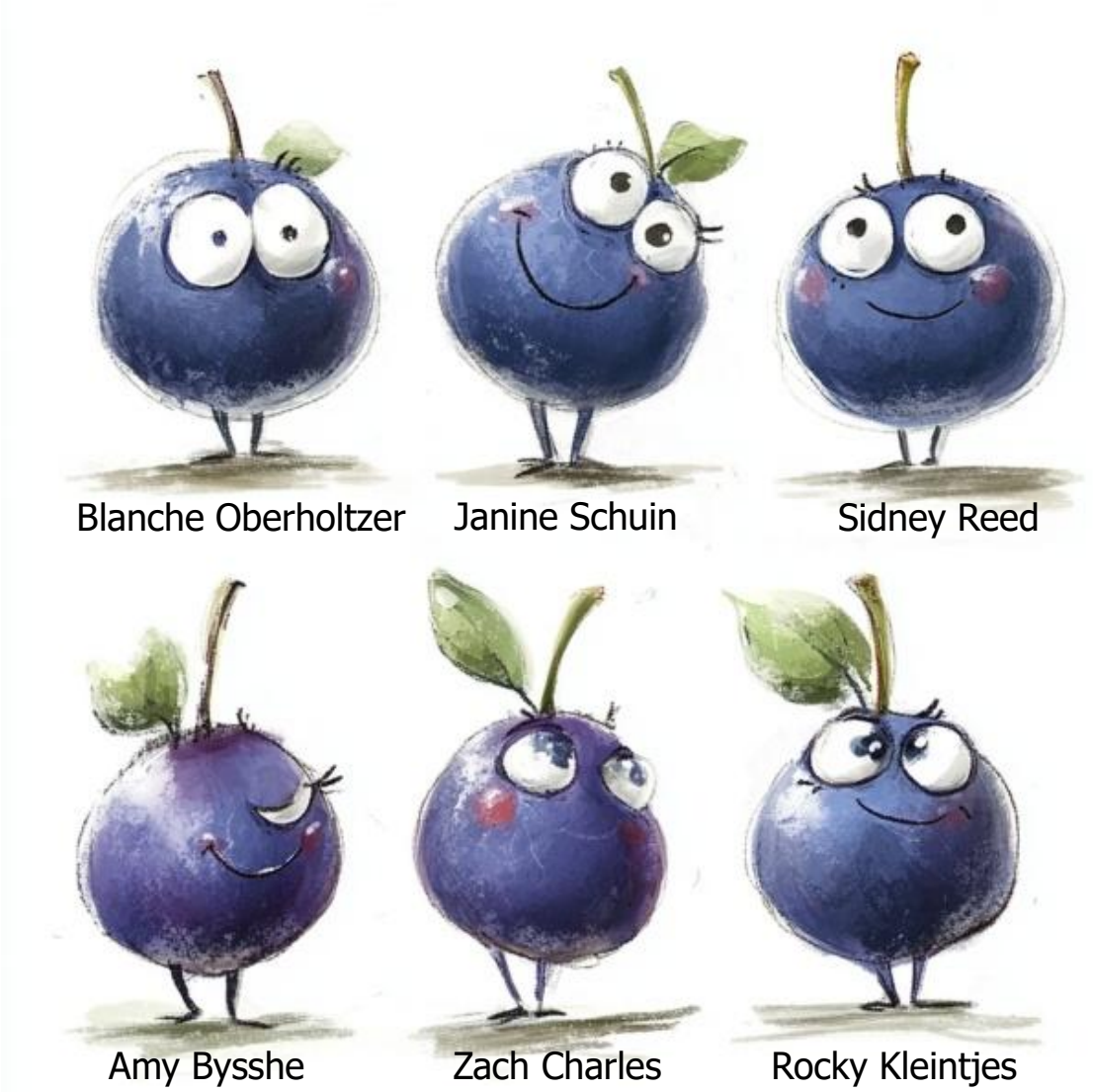
1. Fungal spores in packing facilities increase over the season.
2. *Botrytis* at low levels and can be controlled with cold treatment.
3. Different fungal species in facilities, some of which are potential pathogens.
4. Fungal loads should be monitored and managed.

Acknowledgements

Funding:

- BerryZA
- FEPF (Fresh Produce Exporters Forum)
- NRF
- Stellenbosch University

- Renate
- All the farmers and packhouse managers



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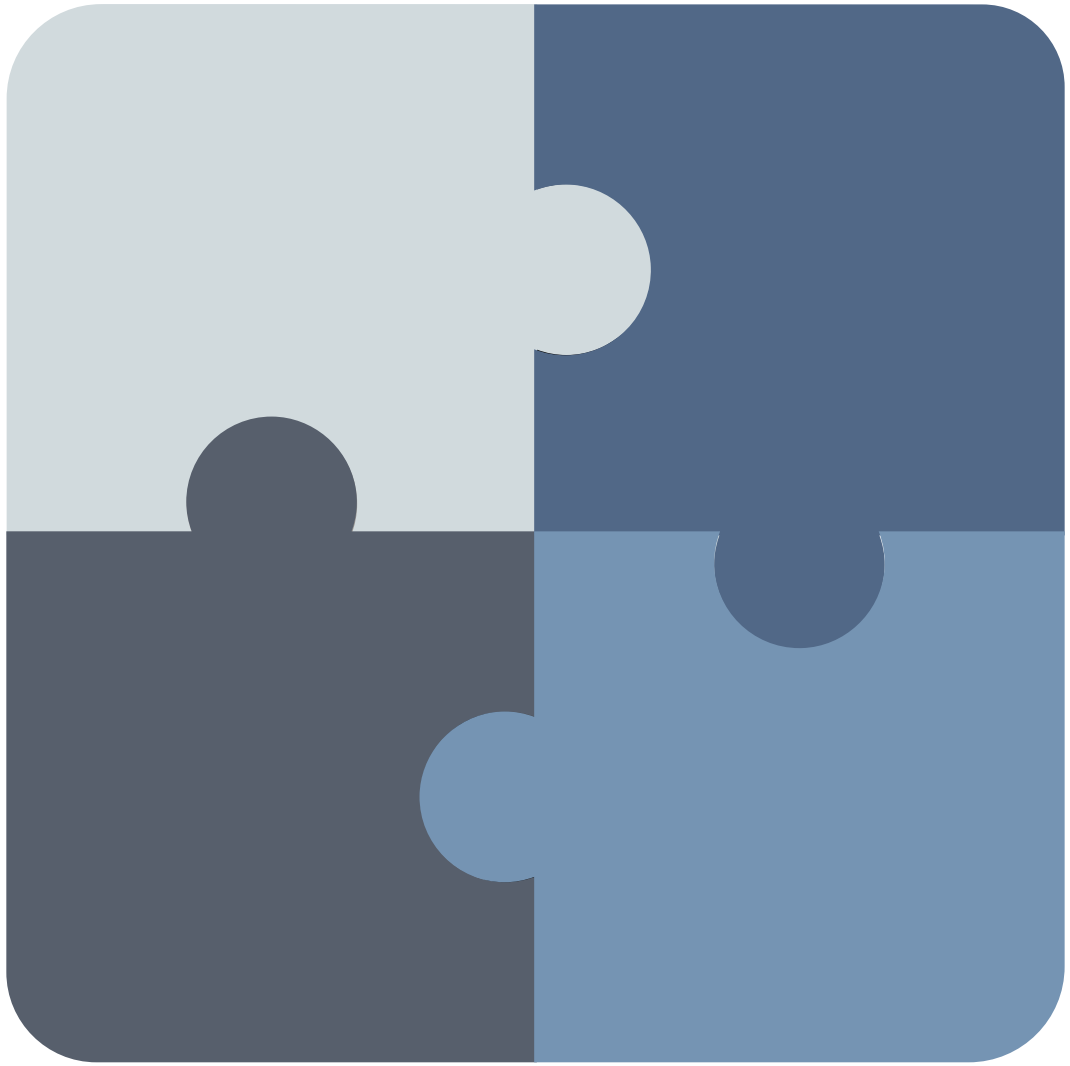
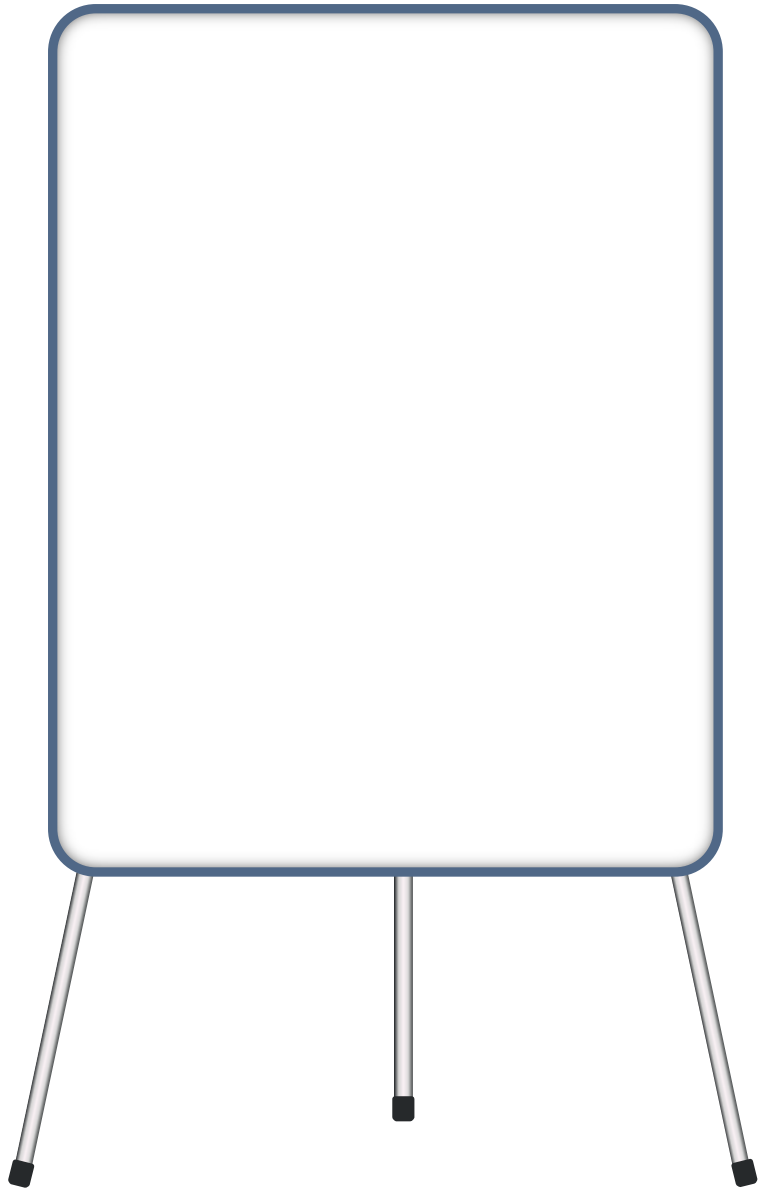
Thank you
Enkosi
Dankie





palet

7594B3

575F6D

516887

D1D9DC

0E0F0E

coolors