Hort Innovation

Final Report

Sowing success through transformational technologies

Project leader:

Dr Jitka Kochanek

Delivery partner:

The University of Queensland

Project code:

VG15021

Project:

Sowing success through transformational technologies (VG15021)

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Level 7
141 Walker Street
North Sydney NSW 2060

Telephone: (02) 8295 2300

www.horticulture.com.au

Content

Summary

VG15021 aimed to produce new products with new uses for the Australian Vegetable Industry via transformational precision technologies which included a novel plant growth regulator (PGR1) previously shown to enhance crop resilience and technologies to enable the cost-effective delivery of PGR1 (costed at *c.* \$20,000 g^{-1}). The target audience was innovators and leaders in the Australian Vegetable Industry. A benefit cost analysis at project onset confirmed that the net present value and BC ratios were positive and high and adoptability analysis predicted time to peak technology adoption was 7.8 to 12.9 years.

The project combined and refined technologies via on-the-ground development with growers on-farm.

Project outcomes were successful development of cost-saving new products that fit operational requirements and were deemed as having high potential benefit and impact for the vegetable sector by leading growers. Key specific outcomes:

1) The development of a novel plant micro-dosing prototype:

The outcome with potentially global impact was the development and validation of a novel precision application prototype to micro-dose crops with novel and classic PGRs. The prototype was validated within commercial systems whereby the novel PGR1 and a commercial PGR (PGR2) were successfully delivered to lettuce var. Blackbelt, broccoli var. Aurora, capsicum var. Katana and tomato var. Rebel and PGR effects lasted until trial termination (up to 10 weeks). Importantly, this innovation cut PGR costs by 100-1000-fold relative to currently utilised delivery mechanisms due to precise and targeted delivery. For example, economic analyses with Solanaceae crops showed that the micro-dosing prototype delivered PGR1 at \$110-1100 ha^{-1} versus conventional application which cost \$110,000 ha^{-1} (i.e. via a root drench), while spraying was ineffective. The clever design also minimises user error and can be automated, thus further cutting labour and other input costs. Significantly, the prototype delivered PGRs as or more effectively than all commercially available PGR delivery mechanisms trialed, including drenches, sprays and priming.

2) Validation of PGR1 as a tool to enhance crop resilience to stress and disease:

Also highly significant was validation that PGR1 was effective at enhancing crop resilience to environmental stress and disease and effective across a broad array of vegetable crops; in fact >90% of levied vegetable lines tested, including lettuce, carrot, capsicum, pumpkin, broccoli, cauliflower, sweet corn and baby spinach, were responsive to PGR1. Crop stress studies determined that PGR1 was a crop multi-protectant, maintaining normal growth under suboptimal conditions that harmed crops not treated with PGR1. Example PGR1 effects were:

a. Delayed bolting and improved yield quality for broccoli grown under suboptimal conditions. Specifically, PGR1 delayed flowering and 16-30% heavier heads were formed in glasshouse and field studies.
b. Stronger plants with better structure and faster fruit formation were validated for capsicum treated with PGR1 when grown on-farm under extreme heat wave/drought conditions. Growers confirmed that the more rapid quality fruit development was highly desirable to minimize grower risk.
c. In a parallel study the team made the world-first discovery that PGR1 significantly suppressed disease in Solanaceae crops via an upregulation of crop defense genes. Specifically, PGR1 halted disease progress and allowed normal fruit development, while the untreated crops were highly diseased and produced few fruit.

Technology transfer and extension activities during the program included 23 on-farm grower interviews, continuous consultative meetings with 5+ leading growers, four industry articles published, 6 presentations at field days/events/conferences, 5 scientific publications, 1 parallel report published and 2 PhD Theses at pre-publication stage. Program Logic, Monitoring & Evaluation and Technology Exploitation Plans were submitted to Hort Innovation. In 2019 the team won the CSIRO ON Prime accelerator program for best innovation, team pitch and team commitment before an audience of 150+ industry innovators. 130+ industry interviews created a technology ecosystem and a pitch video was provided to Hort Innovation. VG15021 funded one PhD scholarship. Labour and expertise was provided without cost to VG15021 by three PhD, four Masters, five Honours and 13 undergraduate UQ research students and additional R&D Advance Queensland, ACIAR and UQ funding bolstered VG15021 outcomes.

Keywords

Vegetable; transformational; disease suppression; resilience; heatwave; plant growth regulator; PGR; propagation; yield; bioengineering; priming; seed; seedling; harvest; abiotic stress; on-farm; grower trials.

Introduction

At project onset VG15021 brought together transformational precision technologies that had been developed with growers and researchers during 5 years of Hort Innovation-funded research (HG10025, MT13058, MT13042) and 10+ years of non-Hort Innovation funded research through multiple UQ and UWA R&D programs. The target audience was innovators and leaders in the Australian Vegetable Industry.

Technology mechanisms to be explored were (a) a plant growth regulator (PGR1) that can enhance crop resilience to environmental variability, especially at establishment, and (b) precision delivery technologies that deliver compounds to crops exactly when and where they are needed and at the right dose, in particular to enable the cost-effective delivery of PGR1, costed at *c.* \$20,000 g^{-1}.

Linkages to the Vegetable Levy Strategic Investment Plan were via:
Pillar 2: The novel plant growth regulator (PGR1) previously shown to enhance crop resilience and innovative precision delivery technologies were linked to Market and Value Chain Development – **New products, new uses, new markets** (Strategy 2.4) to develop promising novel products and technologies (2.4.2) and give Australian products a **competitive advantage** over imported products (2.3.4).
Pillar 3: Cost-effective delivery of PGR1 via innovative precision delivery technologies was linked to Farm Productivity, Resource Use & Management – **Transformational R&D** to enhance the productivity of the Australian vegetable industry (Strategy 3.1), **Manage risks** and improve industry environmental/natural resource management (3.3), including those risks associated with increased future climatic variability (3.4), **Reduce the costs of inputs** such as labour, fuel, energy, fertiliser and other costs (3.5).

Specific VG15021 project objectives were to significantly boost profitability of the Australian vegetable industry through (a) higher profits via enhanced crop establishment, growth and/or yield, (b) reduced costs via enhanced resource efficiency and (c) reduced risk via enhanced crop resilience to environmental variability.

The strategy was to combine and refine technologies via on-the-ground development within grower systems to create new products that fit operational requirements and are cost-effective.

Exhaustive on-farm visits and surveys of leading vegetable growers and industry affiliates within the southern Qld and central NSW vegetable growing areas throughout the project identified real opportunities for VG15021 technologies. In-field stress alleviation and enhanced harvest outcomes were of key importance to growers, as were uniform and reliable crop establishment, with growth outcomes superior to current transplant and direct seeding techniques.

At project onset, it was known that an experimental plant growth regulator (PGR) developed by UWA scientists, here named PGR1, showed promise to enhance crop establishment, growth and resilience to environmental variability (Soos *et al.*, 2010; Nelson *et al.*, 2012; Flematti *et al.*, 2013; Kochanek *et al.*, 2016). In the model plant *Arabidopsis thaliana*, the PGR1 family is proposed to bind a putative α/ß-hydrolase receptor protein, KAI2, transducing a signal to the F-box protein MAX2 that degrades various growth repressing proteins, thus stimulating seed germination, seedling growth and, as determined recently, extending the duration of healthy plant growth under suboptimal conditions (Kochanek *et al.*, 2016). Studies that had explored PGR1 for horticulture were promising and levied vegetable crops known to be PGR1 responsive (Dixon *et al.* 2009; Kulkarni *et al.* 2011) had a gross annual value of \$559m in 2013-14, representing 30% of the \$1.85b p.a. levied Australian vegetable industry, as indicated by benefit cost analysis undertaken at VG15021 onset. Importantly, up to 95% of levied vegetable growers were identified as potentially having direct benefit since they rely on seeds or transplants for production (worth \$1.8b p.a.).

PGR1 promised applicability across grower production systems, being active at minute concentrations, from 1 ppm to < 1 ppb (i.e. 1 nM), water soluble, stable at high temperatures (melting point is *c.* 119°C; Flematti *et al.*, 2004) and non-toxic at plant enhancing concentrations (Verschaeve *et al.*, 2006).

A steering committee, comprised of industry and research leaders, met biannually and oversaw the program to ensure outcome benefits for growers. Potential outcomes promised new products with new markets, soil and environmental quality enhancement and new linkages and opportunities across a cross-disciplinary team of industry and research leaders.

Methodology

At project onset a benefit cost analysis confirmed that the net present value and BC ratios were positive and high ($5.6-63m *per annum* and 1.1-2.2 for worst to best case scenarios, respectively) and adoptability analysis predicted time to peak technology adoption for crop establishment versus crop growth and yield enhancing technologies was 7.8 and 12.9 years, respectively.

1. Grower/industry technology exchange

1a) On-farm grower interviews in 2016 used surveys of eight leading SE Qld vegetable growers and 3 industry affiliates. **Key pain points** were environmental stress at crop establishment, particularly heat waves, and the need for better plant establishment technologies. Technologies were deemed to have high potential benefit for the vegetable sector and markets were confirmed to exist.

1b) On-farm grower interviews in 2017 were across SE Qld and central NSW. Growers exhibited unanimous support for potential VG15021 outcomes. Information collection was via on-farm grower consultation and a Sydney basin field day and non-confidential aspects of project aims and technology potential were communicated.

1c) Grower consultation and feedback from grower trials was collected and used to inform study 4c (i.e. phase III technology development conducted during 2019/20). Drought conditions in 2018-19 pivoted VG15021 to nursery transplant production as growers were unable to supply adequate irrigation for in-field *EmergUniform* prototype activation.

1d) Outcomes from VG15021 were deemed useful by growers at Kalfresh in the Fassifern Valley and Bowen, AustChilli, Flavorite Tomatoes, Gibb Brothers, Story Fresh, Sutton Farms, VegeFresh and others. In late 2019 the team won the CSIRO ON Prime accelerator program for best innovation, team pitch and overall commitment before an audience of 150+ industry innovators. 130+ industry interviews created a technology ecosystem and a pitch video was provided to Hort Innovation.

2. On-farm grower trials - crop establishment, resilience and yield

Grower trials tested and tailored VG15021 technologies for specific industry needs.

2a) Grower trials testing Phase I technologies (developed during study 4a) were completed at a commercial farm in the Lockyer Valley. **Results:** The first study demonstrated on-farm that PGR1 delivery via the seed outperforms root drenching for lettuce. PGR1 seed priming significantly accelerated recovery after a heatwave and resulted in significantly larger plants than control or PGR1 root-drenched plants at 5 weeks after transplant. The second study tested the Phase I prototype with broccoli seeds which determined that modification was required due to poor in-ground seedling outcomes.

2b) Grower trials testing Phase II technologies (developed during study 4b) were completed at a commercial farm in the Lockyer Valley and at UQ St Lucia field plots. **Results:** The first grower study demonstrated on-farm that PGR1 delivery via the seed resulted in 16% heavier broccoli heads relative to untreated plants under conditions that initiate bolting and that PGR1 outperforms a commercial GA suppressant, here named PGR2, which has previously been trialed for bolting suppression. The second study used capsicum var. Katana to demonstrate that a) media in *EmergUniform* prototypes drastically outperformed premium commercial peat media; b) priming drastically outperformed direct sowing and c) dose-optimised priming with PGR1 gave the highest number of premium seedlings (42%) versus 10% for the control. The third study used capsicum var. Katana to demonstrate that *EmergUniform* prototypes with PGR1 doubled the number of seedlings emerged at one week after sowing and resulted in 26% taller plants at two weeks post emergence than the direct sow control. Thus the second and third study demonstrated real merit to introducing *EmergUniform* prototypes into grower systems.

For the fourth and fifth on-farm study drought devastated *EmergUniform* prototype grower trials as growers were unable to provide the irrigation quantity and frequency agreed to at project onset. This was a major pivot point for the project: *EmergUniform* prototypes were put on hold while the micro-dosing prototype was tested extensively within a commercial nursery production system.

2c) During 2019-2020 grower trials tested Phase III technologies (developed during study 4c) at a commercial farm in the Lockyer Valley. **Results:** The first study demonstrated under extreme heat wave and drought conditions for capsicum var. Katana that *EmergUniform* prototype components resulted in stronger plants with better structure and faster fruit formation. Seed priming with very low doses of PGR1 was the most important contributor to positive effects and these plants produced almost three times the number of quality fruit from the first two harvests relative to treatments without PGR1. Leading growers confirmed that more rapid fruit development is highly desirable for industry as it minimizes grower risk by ensuring a more rapid fruit harvest. The second study made the world-first discovery that PGR1 significantly suppressed disease via an upregulation of internal plant defense genes. Specifically, PGR1 halted disease progress and allowed normal fruit development while the untreated crop was highly diseased and produced few fruit. PGR1 seed priming was more effective than root drenching or foliar spraying in suppressing disease, and was cost-effective at \$110-1100 ha^{-1} via the novel micro-dosing prototype, while root drenching was cost prohibitive (\$110,000 ha^{-1}) and foliar sprays ineffective.

3. Crop screening to validate compound effectiveness

3a) Crop screening was completed in Nov 2016. PGR1 plant promoting or retarding attributes were determined from innovative UQ plant growth bioassays, using a series of dosages (very low to overdose) against a control without PGR1. **Results:** All levied vegetable lines tested were responsive to PGR1 including lettuce, carrot, capsicum, pumpkin, broccoli and baby spinach. Thus the stop-go milestone scheduled for Nov 2017 was met one year early since this required >25% of crops to be responsive. Extreme dosage precision for PGR1 was demonstrated, whereby a variety-specific PGR1 dose within several crops was discovered. Importantly, innovative UQ plant bioassays successfully predicted within 7 days the active dosage required to elicit growth effects for each variety tested, predicting long-term effects for weeks to months, and until harvest for capsicum, broccoli and tomato (parallel project).

3b) Crop resilience against stress studies commenced 12 months early and continued throughout project life. PGR1 was demonstrated to be an important crop multiprotectant whereby seed priming with PGR1 maintained normal plant growth under suboptimal conditions while untreated crops were harmed. **Results:** In controlled temperature studies PGR1 seed priming was shown to significantly reduce lettuce bolting and reduce seedling abnormalities for carrot across three repeat heat stress experiments for lettuce and two for carrot. Also a longer duration of exposure to PGR1 was shown to elicit the strongest effects. Across multiple glasshouse and field abiotic stress studies, stressed broccoli plants produced 16-30% heavier heads from PGR1-seed-primed treatments relative to control treatments and PGR1 significantly outperformed a commercial GA suppressant, here named PGR2. Importantly, floral initiation was significantly slowed under stressful conditions for both glasshouse studies confirming that PGR1 compounds likely behave as important multiprotectants that allow crops to maintain normal growth under stressful environments and overcome unwanted consequences such as bolting, which reduce harvest quality for growers.

4. Technology development

4a) Phase I technology development designed prototypes based on grower needs, including for uniformity of emergence, maintenance of high seed viability, enhanced ability to survive transplant shock, especially after heatwaves, and rapid root formation with a taproot. Prototypes needed to fit into existing machinery and growing systems to save infrastructure costs. **Results:** Optimisation determined that (1) Innovative UQ seed priming techniques were superior to commercial seed priming protocol whereby the UQ protocol resulted in a strong PGR1 effect and maintained high seed and seedling viability, while commercial priming techniques did not; (2) Root application was an effective means for PGR1 delivery while shoot application had no effect on plant growth; (3) PGR1 and a commercial germination stimulant, here named PGR3, were delivered more effectively by the Phase I prototype than the UQ seed priming technique, hence also outperforming commercial seed priming protocols. Despite the successes, various design issues that impeded seedling growth in-field demonstrated the need for prototype redesign in study 4b.

4b) For Phase II technology development prototypes were tentatively named *EmergUniform* since uniform emergence and growth are highly sought for Australian vegetable growers. **Results:** A UQ technology used previously to deliver a germination stimulant was utilised with immense success with PGR1 and other novel and commercially available PGRs. The technology was then validated by releasing PGRs as or more effectively than innovative UQ priming protocols to slow or accelerate germination for thermo-inhibited broccoli and lettuce. A more complex prototype was also designed to maximize root growth and named the *EmergUniform* prototype. Since this prototype utilized biochar as a carbon matrix for enhanced root formation, studies focused on optimized biochar development. In the first biochar study a new biochar cleaning technique was validated by drastically improving plant growth outcomes relative to uncleaned biochar for two crops and PGR1 was successfully delivered via the biochar matrix. In the second biochar study dosage response glasshouse trials validated that 30% biochar replacement of peat was optimal for plant growth and this rate was used in subsequent *EmergUniform* prototype studies. In the third study pyrolytic products were analysed for PGR1 content from three Australian pyrolysis companies and biochars from one USA company. However, the dosage precision required for PGR1 to initiate a biologically significant plant response (study 3a) negated the use of such pyrolytic extracts for the horticulture industry, where precision and uniformity are paramount to productivity. Hence no further research was conducted on pyrolytic products.

4c) Phase III technology development attained the 'holy grail' by delivering PGRs using the novel plant micro-dosing prototype within grower systems more effectively than cutting-edge UQ priming methods, which in turn outperformed commercial industry priming techniques. **Results:** Grower demonstration trials validated the novel plant micro-dosing prototype by successfully delivering the novel PGR1 and commercially available PGR2 in a commercial nursery in the Lockyer Valley to lettuce var. Blackbelt, broccoli var. Aurora, capsicum var. Katana and tomato var. Rebel. Significantly, PGR effects lasted until trial termination (i.e. for up to 10 weeks), including after transplanting on-farm, hence confirmed an epigenetic 'plant memory' triggered via the UQ-developed technologies.

Six week old lettuce, broccoli and tomato seedlings were transplanted to a commercial farm in the Lockyer Valley for harvest index determination however all three trials were destroyed, being either accidentally ploughed in just prior to harvest, eaten by hares due to inappropriate location of the test site or destroyed by immense herbicide spray drift, respectively. Thankfully, excellent interim data was collected for lettuce at 10 weeks after sowing (4 weeks after transplanting on-farm) which confirmed that PGR1 and PGR2 delivered via the novel plant micro-dosing prototype continued to display significant differences relative to the controls (P = 0.005), while delivery via conventional priming was not effective (i.e. not different from the controls). Specifically, lettuce head diameter was *c.* 18% wider (*c.* 14.6 cm) with PGR1 and PGR2 delivered via the plant micro-dosing prototype relative to the untreated controls (*c.* 13.2 cm). Additionally, the PGR2 overdose behaved as expected, shrinking plants and resulting in heads being 11% smaller than the control (11.7 cm). Thus the novel plant micro-dosing prototype was demonstrated to work effectively on-farm for at least 1 month post-transplanting (i.e. prior to trial destruction).

No in-field data was collected for broccoli or tomato because plants were destroyed very early in their growth cycle. Had the trials run until harvest it is anticipated that positive harvest indices would be observed, as in earlier trials with PGR1.

5. Industry steering committee, M&E and Exploitation Plans

5a) The 5a industry committee meeting was completed in April 2016 and documents **a**pproved by Hort Innovation included **Program Logic, impediments to technology commercialisation and a Monitoring and Evaluation (M&E) Plan.**

5b) The 5b committee meeting was completed in Nov 2016 and an updated **M&E Plan** was approved by Hort Innovation.

5c) The 5c committee meeting was completed in June 2017 and VG15021 was reviewed and approved as being well on target since the stop-go milestone was met 1 year early. A commercialisation report for the Phase II *EmergUniform* prototype was prepared by UniQuest with UQ Business Masters students.

5d-e) The 5d committee meeting was completed in June 2018 and a new **Program Logic and Monitoring and Evaluation Plan were approved by Hort Innovation.** Version 1 of a 'living' exploitation plan document, updated as the project progressed, was constructed for commercialisation planning with input from Hort Innovation, vegetable industry, UniQuest and researcher stakeholders.

5f) An independent midterm review was completed and the project continued on the basis that project management was successful, the project leader well respected, internal communication between research personnel and growers was excellent and the project progressing as contracted or with activities completed earlier than scheduled. Review feedback was incorporated into the next exploitation plan version. Due to high commercialisation potential Hort Innovation advised **no external communication**.

5g) A commercialisation strategy was reviewed by Hort Innovation Executive Dr Anthony Kachenko and Byron De Kock at a face-to-face meeting at UQ on 26th July 2019 with chief investigator Dr Jitka Kochanek, UQ Dean of Agriculture Prof Neal Menzies and UniQuest Commercialisation Directors Dr Deon Goosen and Dr Kasra Sabermanesh. UQ presented excellent data and patent attorney advice that technologies had better than average potential to be patentable in multiple countries once a broadened scope, via additional data collection, was attained. UQ are actively pursuing a Horticulture Frontiers Fund, as suggested by Hort Innovation, and are presently seeking external industry funds.

Outputs

Phase I project outputs

1. On-farm grower interviews (study 1a) with 11 industry leaders and affiliates demonstrated very high potential benefits and clear market opportunities for vegetable Levy Payers from VG15021 technologies. **Key potential benefits** were alleviation of environmental stress, particularly at crop establishment and from heat waves, and the opportunity for new technologies for plant establishment to improve in-field outcomes.

2. Crop screening (study 3a) was completed in Nov 2016. All levied vegetable lines tested were responsive to PGR1, including lettuce, carrot, capsicum, pumpkin, broccoli and baby spinach. Hence a stop-go milestone scheduled for Nov 2017 was met one year early since this required >25% of crops to be responsive. Additionally, innovative UQ plant bioassays successfully predicted within 7 days the active dosage required to elicit crop growth effects in the long-term (i.e. weeks to months and until harvest for most responsive crops). This was a major breakthrough since extreme dosage precision and variety-specific active PGR1 dosing was demonstrated.

3. Phase I technology prototypes (study 4a) and subsequent designs were based on grower needs, specifically to ensure uniformity of emergence, maintenance of high seed viability, enhanced ability to survive transplant shock, especially after heat waves, rapid root formation and that prototypes must fit into existing machinery and growing systems to save infrastructure costs. Subsequent phase I prototype optimisation determined that:
a) Innovative UQ seed priming techniques were superior to commercial seed priming protocol whereby the UQ protocol resulted in a strong PGR1 effect and maintained high viability, while commercial priming did not,
b) Root application was an effective means for PGR1 delivery while shoot application had no effect on plant growth,
c) PGR1 and a commercial germination stimulant (PGR3) were delivered more effectively by the Phase I prototype than the UQ seed priming technique.

4. Grower trials testing Phase I prototypes (study 2a) were completed at a commercial farm in the Lockyer Valley. Within grower trails PGR1 seed priming (using UQ protocol) (a) significantly accelerated lettuce recovery after a heatwave and (b) resulted in significantly larger plants than control or PGR1-root-drenched plants at 5 weeks after transplant.

5. A committee meeting (milestone 5a) was completed in April 2016. Documents **a**pproved by Hort Innovation included **Program Logic, impediments to technology commercialisation and a Monitoring and Evaluation (M&E) Plan.**

6. A second committee meeting (milestone 5b) was completed in Nov 2016. An updated **M&E Plan** was approved by Hort Innovation.

7. Technology transfer and extension activities were via:
a) Two papers published in peer reviewed scientific journals:
   Kochanek J, Long RL, Lisle AT, Flematti G. 2016. Karrikins identified in biochars indicate post-fire chemical cues can influence community diversity and plant development. *Plos One* 11(8): e0161234.
   Kochanek J, Swift RS, Kochanek MA, Cox J, Flematti GR. 2016. A systematic approach to recycling organics for horticulture: Comparing emerging and conventional technologies. *Acta Horticulturae* 1112: 327-334.
b) An invited industry magazine publication:
   Kochanek J. 2016. Potential new products for sustainable floriculture. *Australian Flower Industry Magazine*, Feb 2016.
c) The chief investigator (Kochanek) was an invited speaker at The World Science Festival, Science Chats at The Edge & live streaming UQ Science Radio interview.

Phase II project outputs

1. Phase II on-farm grower consultation (study 1b) was across SE Qld and central NSW with 12 growers & industry affiliates and non-confidential aspects of project aims and technology potential were communicated to industry via on-farm meetings and a field day. Growers exhibited unanimous support for potential VG15021 outcomes.

2. Crop resilience against stress studies (milestone 3b) commenced 12 months early and continued throughout

project life. PGR1 was demonstrated to behave as a multiprotectant that allows crops to maintain normal growth under stressful environments and overcome unwanted consequences, such as bolting, which reduce harvest quality for growers. For example, PGR1 priming with UQ-developed protocol:
a) Significantly reduced lettuce bolting and carrot seedling abnormalities across multiple repeat heat stress experiments within controlled temperature conditions. A longer duration of PGR1 exposure was shown to elicit the strongest effects.
b) Significantly reduced broccoli bolting by slowing floral initiation under stressful conditions and resulted in 16-30% heavier heads from PGR1 treatments relative to control treatments. The same results were confirmed across two repeat glasshouse (study 3b) and on-farm field (study 2b) studies. Significantly, PGR1 significantly outperformed a commercial GA suppressant (here named PGR2) which has been trialed previously for bolting suppression.

3. For Phase II technology development (study 4b) in-field prototypes were tentatively named *EmergUniform* since uniform emergence and growth are highly sought for Australian vegetable growers. Since the *EmergUniform* prototype utilized biochar as a carbon matrix for enhanced root formation, studies focused on optimized biochar development whereby:
a) A new biochar cleaning technique was validated by drastically improving plant growth outcomes relative to uncleaned biochar for two crops and PGR1 was successfully delivered via the biochar matrix,
b) Dosage response glasshouse trials validated that 30% biochar replacement of peat was optimal for plant growth and this rate was used in subsequent *EmergUniform* prototypes. This replacement rate agreed with earlier work conducted by the team (Kochanek et al 2016a, b).
c) Pyrolytic products were analysed for PGR1 content from three Australian pyrolysis companies and biochars from a USA company,
d) The discovery that PGR1 must be applied at variety-specific doses resulted in the abandonment of the PGR1-rich 'pyrolytic product' technologies,
e) Instead a cost-effective delivery mechanism for pure PGR1 was discovered and pursued going forward.

A second prototype – which later became the novel plant micro-dosing prototype - was developed for nursery-specific uses, utilising a UQ technology (previously used to deliver a commercial germination stimulant) with immense success to deliver PGR1. The innovative technology was validated by releasing PGR1 and broader PGRs as or more effectively than UQ priming and commercial seed priming protocol, for example, to slow or accelerate germination for thermo-inhibited broccoli and lettuce.

4. Grower trials testing Phase II prototypes (study 2b) were completed at a commercial vegetable farm in the Lockyer Valley or in UQ St Lucia field plots. Studies demonstrated real merit to introducing Phase II prototypes into grower systems, for example:
a) Under stressful conditions that initiate bolting, PGR1 delivery via the novel priming protocol resulted in 16% heavier broccoli heads relative to untreated plants and significantly delayed flowering. Also PGR1 outperformed a commercial GA suppressant (PGR2) that is being explored by other researchers as a potential bolting suppressant.
b) Study 1 with capsicum var. Katana was a glasshouse study at UQ and demonstrated that (i) media in *EmergUniform* prototypes drastically outperformed premium commercial peat media, (ii) UQ priming protocols drastically outperformed direct sowing and (iii) dose-optimised priming with PGR1 gave the highest number of premium seedlings (42%) versus only 10% for the untreated control. Seedlings were transplanted onto a commercial farm in the Lockyer Valley (results are detailed in study 2c).
c) Study 2 with capsicum var. Katana was an in-field study at UQ and demonstrated that (i) *EmergUniform* prototypes containing PGR1 doubled the number of seedlings emerged at one week after sowing and (ii) resulted in 26% taller plants at two weeks post emergence than the direct sow control.

Unfortunately subsequent on-farm trials with *EmergUniform* prototypes were devastated by drought as growers were unable to provide the irrigation quantity and frequency agreed to at project onset. This was a <u>major pivot point</u> for the project and the *EmergUniform* prototype were put on hold, with all focus being towards development of the novel plant micro-dosing prototype for phase III prototype development (study 4c).

5. A committee meeting (milestone 5c) was completed in June 2017. Additional outputs were:
a) VG15021 was reviewed and approved as being well on target since the stop-go milestone was met 1 year early,
b) A commercialization report for Phase II *EmergUniform* prototypes was produced by UniQuest with Business Masters students.

6. Sister project VG16028 was successfully completed by the VG15021 chief investigator (Kochanek) and communicated to vegetable levy payers. Technology transfer and extension activities included:

a) A face-to-face presentation to the vegetable levy board in Sydney by the chief investigator:

Kochanek J. presented VG16028 outcomes and future opportunities to the vegetable levy board in 2018.

b) Three industry articles were published:

Kochanek J. 2018. Grower input opportunity: Future R&D for improved on farm vegetable seed quality. Lockyer Valley Growers Magazine, May 2018.

Kochanek J. 2018. VG16028: On farm evaluation of vegetable seed viability using non-destructive techniques. Vegenotes magazine, March/April 2018.

Kochanek J. 2017. Turning seed quality failure into consistent success: How widespread is the problem? Summer 2017 HortLink Magazine: Horticulture Innovation Australia.

c) VG16028 final report was published:

Kochanek J. 2017. On farm evaluation of vegetable seed viability using non-destructive techniques. Final report for Horticulture Innovation Australia project VG16028.

d) Three presentations at a field day and conference:

Kochanek J. 2017 presented non-confidential VG15021 project outcomes at a field day in the Sydney Basin.

Kochanek J. 2018 presented non-confidential VG15021 project outcomes at a field day for the Lockyer Valley Growers BBQ.

PhD student presentation at an international conference: Dakuidreketi A, Aitken EAB, Furlong M, Flematti G, **Kochanek J.** 2018. The potential effect of karrikinolide (KAR_1) in inducing resistance against *Alternaria solani* on tomato, International Congress of Plant Pathology (ICPP), Boston, Massachusetts.

7. Committee meeting 5d was completed in June 2018. For this:
a) A new **Program Logic and Monitoring and Evaluation were approved by Hort Innovation.**

b) Version 1 of a 'living' exploitation plan document, updated as the project progressed, was constructed for commercialisation planning with input from Hort Innovation, vegetable industry, UniQuest and researcher stakeholders.

8. VG15021 chief investigator (CI, Kochanek) continued as active CI and project manager while on UQ-paid maternity leave and bolstered project outcomes with additional external *Advance Queensland grant* funding during 2017-18. Hence VG15021 received >8 months of fully supported R&D and salaries from UQ and the *Advance Queensland grant* towards project outcomes.

9. An independent midterm review was completed and the project continued. The report summary determined that (i) project management was successful, (ii) the project leader was well respected, (iii) internal communication between research personnel and growers was excellent, (iv) the project was progressing as contracted or with activities completed earlier than scheduled. Review feedback was incorporated into the second exploitation plan.

Due to high commercialisation potential of technologies Hort Innovation advised **no external communication** to ensure technology confidentiality was maintained.

10. Grower consultation and feedback from grower trials was collected (study 1c) and used to inform phase III technology development (study 4c). Drought conditions in 2018-19 pivoted VG15021 to nursery propagation as growers were unable to supply adequate irrigation for in-field *EmergUniform* prototype activation. Thus study 4c focused on validation of the novel plant micro-dosing prototype within commercial nursery and on-farm systems.

Phase III project outputs

1. Phase III technology development (study 4c) attained the 'holy grail':
a) PGR1 and PGR2 were successfully delivered via the novel plant micro-dosing prototype in grower demonstration trials at a commercial nursery in the Lockyer Valley to lettuce var. Blackbelt, broccoli var. Aurora, capsicum var. Katana and tomato var. Rebel.
b) The prototype delivered PGRs as or more effectively that optimized UQ priming protocol, which in turn outperformed commercial industry priming protocols.
c) Effects lasted until trials were terminated/destroyed (i.e. up to 10 weeks post sowing) hence confirmed an epigenetic plant memory triggered by the novel plant micro-dosing prototype. For example, PGR1 and PGR2 delivered via the prototype continued to display significant differences relative to controls for 4 weeks after they were transplanted to a commercial Lockyer Valley farm, while delivery via industry priming was not effective. Lettuce head diameter was *c.* 18% wider with both PGRs delivered via the plant micro-dosing prototype relative to the untreated controls. By contrast, commercial industry priming protocols resulted in short term effects (*c.* 2 weeks) but no longer-term effects, demonstrating that the UQ prototype outperformed current industry practice.
d) Lettuce, broccoli and tomato seedlings were transplanted to a commercial Lockyer Valley farm for harvest determination however all three trials were destroyed, being accidentally ploughed in just prior to harvest, eaten by hares due to inappropriate location of the test site or destroyed by immense herbicide spray drift, respectively. On-farm lettuce results are discussed in 1c above, while no on-farm data was salvageable from broccoli and tomato trials.

2. Grower consultation and feedback (study 1d) confirmed that VG15021 outcomes were highly useful for industry. Growers interviewed were from Kalfresh in the Fassifern Valley and Bowen, AustChilli, Sutton Farms, VegeFresh, Flavorite Tomatoes, Gibb Brothers, Story Fresh and others.

3. Additional grower trials tested Phase III technologies at a commercial farm (study 2c) in the Lockyer Valley under an extreme heat wave and drought conditions for capsicum var. Katana:
a) *EmergUniform* prototype components resulted in stronger plants with better structure and faster fruit formation,

b) Low dose PGR1 priming was the most important contributor to positive effects and these plants produced almost three times the number of quality fruit from the first two harvests relative to treatments without PGR1,

c) Growers confirmed that more rapid fruit development is highly desirable for industry as it minimizes grower risk by ensuring a more rapid fruit harvest.

4. The chief investigator's (Kochanek) team made the world-first discovery that PGR1 significantly suppressed disease via an upregulation of internal plant defense genes. Specifically:
a) PGR1 seed priming was more effective than root drenching or foliar spraying in suppressing disease. In fact, foliar sprays were entirely ineffective.
b) PGR1 seed priming halted disease progress and allowed normal fruit development, while the untreated crops were highly diseased and produced few fruit.
c) PGR1 seed priming was drastically more cost-effective (\$110-1100 ha^{-1}) than root drenching, while root drenching was entirely cost prohibitive (\$110,000 ha^{-1}).

5. A commercialisation strategy was reviewed by Hort Innovation Executive Dr Anthony Kachenko and Byron De Kock at a face-to-face meeting at UQ on 26th July 2019 with CI Dr Jitka Kochanek, UQ Dean of Agriculture Prof Neal Menzies and UniQuest Commercialisation Directors Dr Deon Goosen and Dr Kasra Sabermanesh.

- UQ presented excellent data and patent attorney advice that technologies had better than average potential to be patentable in multiple countries once a broadened scope via additional data collection was attained,
- Industry consultation across 17 separate businesses has confirmed that the technology requires further R&D before it can be commercialized,
- Upon advice from Hort Innovation, UQ are pursuing a Horticulture Frontiers Funded project and seeking external industry partner cash inputs,
- Additionally, support has been obtained from the Nursery Industry via letters of approval from key Australian businesses.

6. In late 2019 the team won the CSIRO ON Prime accelerator program for best innovation, team pitch and overall commitment before an audience of 150+ industry innovators. 130+ industry interviews created a technology

ecosystem and a pitch video was provided to Hort Innovation.

7. Technology transfer and extension activities:

a) Three peer reviewed papers/abstracts were published by the chief investigator (Kochanek) and her team:

   Zhong Xiang C, Harper S, O'Hare T, **Kochanek J**, Bell M. 2020. Zinc biofortification of immature maize and sweetcorn (*Zea mays* L.) kernels for human health. *Scientia Horticulturae,* 272, 109559.

   Tryggestad K, Krisantini S, Flematti G, **Kochanek J.** 2019. Beyond Arabidopsis: Paving the way to commercial applications of karrikin compounds, 4th Edition of the Global Conference on Plant Science and Molecular Biology, September 19-21st 2019, London, UK.

   Dakuidreketi A, Aitken EAB, Furlong M, Flematti G, **Kochanek J.** 2018. The potential effect of karrikinolide (KAR_1) in inducing resistance against *Alternaria solani* on tomato, International Congress of Plant Pathology (ICPP), Boston, Massachusetts.

b) Two PhD theses with R&D directly relevant to VG15021 will be submitted in early 2021 (Tryggestad and Dakuidreketi).

c) PhD student oral presentation at an international conference:

   Tryggestad K, Krisantini S, Flematti G, **Kochanek J.** 2019. Beyond Arabidopsis: Paving the way to commercial applications of karrikin compounds, 4th Edition of the Global Conference on Plant Science and Molecular Biology, September 19-21st 2019, London, UK.

Outcomes

New products and markets for growers were developed

Project outcomes were successful development of cost-saving new products that fit operational requirements and were deemed as having high potential benefit and impact for the vegetable sector by leading growers.

The outcome with potentially global impact was the development and validation of a novel precision application prototype to micro-dose crops with novel and classic plant growth regulators (PGRs), which is unlike anything available on the market today. Technology readiness level (TRL) is used to assess technology maturity using a scale of 1-9, where 1 is *basic principles observed and reported*, 7 is *prototype demonstration in an operational environment* and 9 is *routine use*. The plant micro-dosing prototype in VG15021 has reached a TRL of 6-7, since it was designed and tested with industry input and has been demonstrated in a commercial operating system, successfully delivering a novel PGR and commercially available PGR to four vegetable lines, namely lettuce var. Blackbelt, broccoli var. Aurora, capsicum var. Katana and tomato var. Rebel. Effects lasted until trial termination hence confirmed an epigenetic plant memory triggered by PGRs delivered via the technology.

In 2019 UQ presented excellent data and patent attorney advice to Hort Innovation that the technology had better than average potential to be patentable in multiple countries once a broadened scope via additional data collection was attained. In late 2019 the team won the CSIRO ON Prime accelerator program for best innovation, team pitch and overall commitment before an audience of 150+ industry innovators. 130+ industry interviews created a technology ecosystem and a pitch video was provided to Hort Innovation.

Advice from discussions with managers/personnel across 17 businesses is that the technology requires further R&D support before it can be commercialised. Hence UQ are now pursuing external industry partners towards a Hort Frontiers Funded study. Already the project has support from the Nursery Sector, with letters of support from key Australian businesses and CEO.

Grower interviews and consultation throughout project life confirmed that VG15021 outcomes were highly useful for industry and clear market opportunities existed for vegetable Levy Payers from VG15021 technologies. Growers consulted in 2019-20 included those from Kalfresh in the Fassifern Valley and Bowen, Sutton Farms, VegeFresh, AustChilli, Flavorite Tomatoes, Gibb Brothers, Story Fresh and others.

Also highly significant was validation that PGR1 was effective at enhancing crop resilience to environmental stress and disease and effective across a broad array of vegetable crops; in fact >90% of levied vegetable lines tested, including lettuce, carrot, capsicum, pumpkin, broccoli, cauliflower, sweet corn and baby spinach, were responsive to PGR1. More detail is provided below.

Significant productivity increases for growers

<u>Higher profits via enhanced crop establishment, growth and yield</u>

PGR1, which was successfully delivered by the novel plant micro-dosing prototype, was shown throughout the project to enhance grower productivity. Results throughout the project confirmed that PGR1 behaves as an important multiprotectant that allows crops to maintain normal growth under stressful environments and overcome unwanted consequences, such as bolting and disease, which reduce harvest quality for growers.

<u>Reduced costs of production via enhanced resource efficiency</u>

PGR1 delivery via the plant micro-dosing prototype was demonstrated to be 100-1000 times more cost-effective than current industry PGR application protocols, which were entirely cost prohibitive or did not work for PGR1. For example, for crop disease suppression via an upregulation of crop defense genes delivery of PGR1 via the new technology would cost \$110-1100 ha^{-1} (i.e. <1c plant) while delivery via root drenching would cost \$110,000 ha^{-1} (*c.* \$1 per plant). PGR1 foliar sprays were entirely ineffective.

More than 90% of lines tested were sensitive to PGR1 and the micro-dosing prototype was not only cost-effective but cost-saving relative to current industry PGR application techniques. For example, labour and other input costs were also reduced due to micro-dosing of the PGRs minimising user error and ease of process automation cutting labour costs of PGR application.

<u>Reduced risk via enhanced crop resilience to environmental variability</u>

Three clear examples whereby VG15021 enhanced crop resilience to environmental variability:

A. Broccoli bolting was significantly delayed with PGR1 treatments under stressful conditions across repeat glasshouse and field studies, and PGR1-treated plants produced 16-30% heavier heads relative to control un-treated plants. Further, PGR1 significantly outperformed the commercial GA suppressant and multiprotectant, here named PGR2, that has previously been trialed for bolting suppression.

B. The Chief Investigator's team made the world-first discovery that PGR1 significantly suppressed disease via an upregulation of crop defense genes. Additionally, PGR1 priming was demonstrated to be more effective than root drenching in suppressing disease while PGR1 foliar sprays were entirely ineffective.

C. Under an extreme heat wave and drought conditions for capsicum var. Katana the *EmergUniform* prototype components resulted in stronger plants with better structure and faster fruit formation. Seed priming with low PGR1 doses was the most important contributor to positive effects and these plants produced almost three times the number of quality fruit from the first two harvests relative to treatments without PGR1. Growers confirmed that more rapid fruit development is highly desirable for industry as it minimizes grower risk by ensuring a more rapid fruit harvest.

Spill-over benefits for society e.g. soil and environmental quality benefits

A novel plant micro-dosing prototype was developed by the project and delivery of a novel PGR via the prototype was demonstrated to be 100-1000 times more cost-effective than with current industry PGR application protocols, which were either entirely cost prohibitive (root drenching) or did not work (foliar sprays, commercial priming protocols).

Commercially available PGRs were delivered with similarly impressive outcomes: for example a GA suppressant (PGR2) used at ppm doses for potted colour growth control was successfully delivered to transplants at ppb doses by the prototype, therefore a 1000-fold reduction in dosing rate. Since the unit was developed for a commercial production setting, other critical elements for grower safety and practicality were incorporated into the design; for example, the user is never exposed to the chemical used and the technology is designed to be automated to drastically reduce labour requirements and to improve uniformity of outcomes. The broad plant families across which the unit was validated (Asteraceae, Solanaceae, Brassicaceae) and array of chemicals successfully delivered suggests applications to other plant sectors, such as broader horticulture production and chemicals beyond PGRs.

It is envisioned that the plant micro-dosing prototype will first be rolled out for the vegetable sector via the nursery transplant industry. If R&D funding towards Emerguniform prototype development is supported, there is additional scope for direct on-farm application of these technologies (i.e. not only via nurseries). Utilising the PGRs tested in this study, the technologies are expected to significantly improve crop disease outcomes and reduce the severity of effects from suboptimal conditions, such as heatwaves, in PGR-responsive crops such as capsicum and broccoli.

Further, the technology has the potential to significantly reduce chemical use within grower systems which will have immediate spill-over benefits to society via, for example, cleaner soils and waterways and fewer chemical residues on horticultural produce.

New linkages and opportunities

National and international reputation: In late 2019 the team won the CSIRO ON Prime accelerator program for best innovation, team pitch and overall commitment before an audience of 150+ industry innovators. 130+ industry interviews created a technology ecosystem and a pitch video was provided to Hort Innovation. Industry leaders in Australia and internationally in the USA, Europe and Asia were made aware of the innovation.

Building horticulture teaching capacity in Australia: The chief investigator (Kochanek) is a research academic at the University of Queensland and throughout the project supervised four PhD, four Masters, five Honours and 13 undergraduate research student projects. All student projects provided labour and expertise that enabled the immense success of VG15021. The students are now skilled professionals in horticulture and many will contribute to the industry into the future.

Monitoring and evaluation

The overall project performance was exceptional in that all Key Evaluation Questions (KEQs) were answered during VG15021. Below is listed how the project answered KEQs for the Australian Vegetable Industry & Hort Innovation Australia against SIP outcomes within the M&E Plan.

New products, new uses, new markets (Strategy 2.4) and using transformational R&D to enhance the productivity of the Australian Vegetable industry (3.1)

MEETS AUSTRALIAN GOVERNMENT CRITERIA: Enhanced productivity & adding value

Grower interviews and consultation throughout project life confirmed that VG15021 outcomes were highly useful for industry and clear market opportunities existed for vegetable Levy Payers from VG15021 technologies. Growers consulted in 2019-20 included those from Kalfresh in the Fassifern Valley and Bowen, AustChilli, Sutton Farms, VegeFresh, Flavorite Tomatoes, Gibb Brothers, Story Fresh and others.

A commercialisation strategy was reviewed by Hort Innovation Executive Dr Anthony Kachenko and Byron De Kock at a face-to-face meeting at UQ on 26th July 2019 with CI Dr Jitka Kochanek, UQ Dean of Agriculture Prof Neal Menzies and UniQuest Commercialisation Directors Dr Deon Goosen and Dr Kasra Sabermanesh. UQ presented excellent data and patent attorney advice that technologies had better than average potential to be patentable in multiple countries once a broadened scope via additional data collection was attained. Upon recommendation from Hort Innovation, UQ are actively pursuing a Hort Frontiers project and are seeking external industry partners. Strong support has already been obtained from the Australian Nursery Production Industry.

In late 2019 the team won the CSIRO ON Prime accelerator program for best innovation, team pitch and overall commitment before an audience of 150+ industry innovators. 130+ industry interviews created a technology ecosystem and a pitch video was provided to Hort Innovation.

Develop promising novel products and technologies (2.4.2)

VG15021 attained the 'holy grail' by delivering PGRs within commercial systems via the novel plant micro-dosing prototype as or more effectively than UQ priming protocol which in turn outperformed commercial protocols, including industry priming, root drenching and foliar sprays.

The novel PGR1 and commercially available PGR2 were successfully delivered in grower demonstration trials to lettuce var. Blackbelt, broccoli var. Aurora, capsicum var. Katana and tomato var. Rebel at a commercial nursery in the Lockyer Valley and also on a commercial vegetable farm, with PGR effects lasting until trial termination (i.e. 10 weeks). This confirmed an epigenetic plant memory triggered by PGR priming via the UQ developed micro-dosing prototype.

Give Australian products a competitive advantage over imported products (2.3.4) and manage risks and improve industry environmental/natural resource management (3.3) including those risks associated with increased future climatic variability (3.4)

PGR1 was successfully delivered by the plant micro-dosing prototype and was shown throughout the project to enhance grower productivity. Results throughout the project confirmed that PGR1 behaves as an important multi-protectant that allows crops to maintain normal growth under stressful environments and overcome unwanted consequences, such as bolting, which reduce harvest quality for growers.

Three examples:

A. Bolting was significantly delayed under stressful conditions for broccoli plants treated with PGR1, with such plants producing 16-30% heavier heads than from un-treated control plants; and results were re-confirmed across repeat glasshouse and on-farm field studies. Further, PGR1 significantly outperformed a commercial GA suppressant and multi-protectant, named PGR2.

B. The team made the world-first discovery that PGR1 significantly suppressed disease via an upregulation of internal plant defense genes. Additionally, PGR1 seed priming was demonstrated to be more effective than root drenching in suppressing disease while PGR1 foliar sprays were entirely ineffective.

C. Under an extreme heat wave and drought conditions for capsicum var. Katana the *EmergUniform* prototype components resulted in stronger plants with better structure and faster fruit formation. Dosing with low PGR1 concentrations was the most important contributor to positive effects and these plants produced almost three times the number of quality fruit from the first two harvests relative to treatments without PGR1. Growers confirmed that more rapid fruit development is highly desirable for industry as it minimizes grower risk by ensuring a more rapid fruit harvest.

Reduce the costs of inputs such as labour, fuel, energy, fertiliser and other costs (3.5)

The potential new products from VG15021 are an entirely new way to precisely and cost-effectively deliver plant growth regulators or other chemistries to crops within a grower's system. Importantly, the prototype is activated to release these compounds to the plant at a precise dose within the growers system, making its design completely different to other delivery technologies on the market today.

PGR1 delivery via the prototype was demonstrated to be 100-1000 times more cost-effective than currently used PGR application protocols, which were entirely cost prohibitive or did not work. For example, to deliver crop disease suppression via an upregulation of crop defense genes delivery of PGR1 via the prototype would cost *c.* \$110-1100 ha^{-1} (<1c plant) while delivery via root drenching would cost *c.* \$110,000 ha^{-1} (*c.* \$1 per plant). PGR1 foliar sprays were entirely ineffective.

More than 90% of lines tested were sensitive to PGR1 and prototypes were cost-effective and cost-saving relative to current industry PGR application techniques.

Specific answer to Key Evaluation Questions (KEQs):

Effectiveness – the technology is highly effective
The project developed a prototype that is useful for industry. The delivery of PGRs via the prototype was cost-effective and cost-saving for growers – in fact making PGRs far more useful, safe, accessible and cost-effective than with currently used application techniques.

Relevance – the technology is highly relevant

The project met the needs of industry levy payers since more than 90% of lines tested were sensitive to PGR1 and prototypes were cost-effective relative to currently used PGR application techniques, providing PGRs 100-1000 times cheaper via lower doses.

The technology is process appropriate

Regular project updates were provided through linkage with the industry steering committee and SIAP.

The project engaged with industry levy payers, regularly sharing information that was not confidential and sharing deeper information where a confidentiality agreement was in place. Levy payers were consulted throughout the project and were on the steering committee. Non-confidential aspects were communicated to industry throughout the project at field days, presentations, via bulletins, face-to-face or phone consultation and via an industry pitch video.

The project was highly efficient - The project considered on-farm efficiency at every stage of the technology research, development and design. This was imperative to the successful design of the prototype.

Recommendations

Recommendations to industry/growers

The project has developed a novel plant micro-dosing prototype, designed to safely, precisely and cost-effectively deliver PGRs to plants. It is envisioned that the plant micro-dosing prototype will be rolled out for the vegetable sector via the nursery transplant industry, at least initially. Hort Innovation and the Nursery Production Industry support a Hort Frontiers Funded project to complete the R&D needed to bring about commercialisation of the technology.

The prototype delivered PGRs as or more effectively within grower systems as all commercially available PGR delivery mechanisms trialed, including root drenches, foliar sprays and seed priming.

A novel PGR (PGR1) and a commercially available micro-protectant (PGR2) were successfully delivered via the novel prototype in commercial demonstration trials for lettuce var. Blackbelt, broccoli var. Aurora, capsicum var. Katana and tomato var. Rebel. Positive effects lasted until trial termination, including within commercial on-farm field trials, hence confirmed an epigenetic plant memory triggered by the PGRs delivered via the novel plant micro-dosing prototype. By contrast, PGR delivery via conventional industry application protocols (root drenches, foliar sprays and seed priming) were less or not effective.

The prototype reduced PGR doses 100-1000-fold hence also cut PGR costs 100-1000-fold within grower systems. Cost savings can further be made since the prototype removes user-error from PGR delivery and is designed to be fully automated, thus reducing labour costs and frustrations for growers who struggle to find skilled workers. Immediate spill-over benefits to society will be, for example, via cleaner soils and waterways, fewer PGR residues and improved profitability and productivity of horticulture production systems.

Novel plant growth regulators present real opportunities for crops to cope with environmental variability and stress in real-time. The novel PGR1 tested in this study was demonstrated to be an important crop multiprotectant and optimised delivery of PGR1 maintained normal plant growth under suboptimal conditions.

Examples of enhanced crop resilience included:

(a) Significantly delayed flowering and hence reduced bolting under stressful conditions for PGR1-treated broccoli plants and, as a result, 16-30% heavier heads than from untreated control plants,

(b) For capsicum grown on-farm under an extreme heat wave and drought conditions, PGR1-treated plants were stronger, had better structure (thicker stems, shorter internodes) and faster fruit formation. Growers confirmed that the more rapid quality fruit development is highly desirable for industry as it minimizes grower risk by ensuring a more rapid quality fruit harvest,

(c) The team also made the world-first discovery that PGR1 significantly suppressed disease via an upregulation of internal crop defense genes. Specifically, PGR1 treated plants displayed halted disease progress and normal fruit development, while the untreated crops were highly diseased and produced few fruit.

Recommendations to R&D investment decision makers

The project has developed a novel plant micro-dosing prototype designed to safely, precisely and cost-effectively deliver PGRs to plants. In mid-2019 UQ presented excellent data, patent attorney advice and a R&D strategy towards commercialisation to Hort Innovation. In late 2019 the team won the CSIRO ON Prime accelerator program for best innovation, team pitch and overall commitment before an audience of 150+ industry innovators. 130+ industry interviews created a technology ecosystem and a pitch video was provided to Hort Innovation. Hort Innovation and the Nursery Production Industry support a Hort Frontiers Funded project to complete the R&D needed to bring about commercialisation of this highly promising technology and UQ is actively seeking external industry funding to progress this next exciting project phase.

Refereed scientific publications

Peer reviewed papers/abstracts/reports published by the CI and her team:

Zhong Xiang C, Harper S, O'Hare T, Kochanek J, Bell M. 2020. Zinc biofortification of immature maize and sweetcorn (*Zea mays* L.) kernels for human health. *Scientia Horticulturae,* 272, 109559.

Tryggestad K, Krisantini S, Flematti G, Kochanek J. 2019. Beyond Arabidopsis: Paving the way to commercial applications of karrikin compounds, 4th Edition of the Global Conference on Plant Science and Molecular Biology, September 19-21st 2019, London, UK.

Dakuidreketi A, Aitken EAB, Furlong M, Flematti G, Kochanek J. 2018. The potential effect of karrikinolide (KAR_1) in inducing resistance against *Alternaria solani* on tomato, International Congress of Plant Pathology (ICPP), Boston, Massachusetts.

Kochanek J. 2017. On farm evaluation of vegetable seed viability using non-destructive techniques. Final report for Horticulture Innovation Australia project VG16028.

Kochanek J, Long RL, Lisle AT, Flematti G. 2016. Karrikins identified in biochars indicate post-fire chemical cues can influence community diversity and plant development. *Plos One* 11(8): e0161234.

Kochanek J, Swift RS, Kochanek MA, Cox J, Flematti GR. 2016. A systematic approach to recycling organics for horticulture: Comparing emerging and conventional technologies. *Acta Horticulturae* 1112: 327-334.

More articles are in preparation.

Industry bulletins/articles published by the CI and her team:

Kochanek J. 2018. Grower input opportunity: Future R&D for improved on farm vegetable seed quality. Lockyer Valley Growers Magazine, May 2018.

Kochanek J. 2018. VG16028: On farm evaluation of vegetable seed viability using non-destructive techniques. Vegenotes magazine, March/April 2018.

Kochanek J. 2017. Turning seed quality failure into consistent success: How widespread is the problem? Summer 2017 HortLink Magazine: Horticulture Innovation Australia.

Kochanek J. 2016. Potential new products for sustainable floriculture. *Australian Flower Industry Magazine*, Feb 2016.

Theses

Two PhD theses with R&D directly relevant to VG15021 will be submitted in early 2021 (Tryggestad and Dakuidreketi).

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Intellectual property, commercialisation and confidentiality

This report is public and non-confidential.

Acknowledgements

We acknowledge with gratitude the immense assistance of growers across horticulture industries who allowed our team to conduct face-to-face interviews, visit farms and conduct crop growth trials within their production systems. In particular we wish to acknowledge the team at VegeFresh nursery in the Lockyer Valley whose exceptional attention to detail allowed for the development of our innovative technologies within a commercial production system, and hence drastically boosted our technology readiness level. Without their immense assistance we would not have made the discoveries that won the CSIRO ON Prime program in 2019. We also thank Byron De Kock for his kindness, enthusiasm and support throughout VG15021.

We also acknowledge with gratitude the immense support and assistance of UQ staff towards project outcomes, primarily of Dr Krisantini who worked tirelessly since 2016 to ensure R&D success across all trials and tribulations; UniQuest staff Dr Deon Goosen, Dr Karin Taylor and Dr Kasra Sabermanesh for their endless support and enthusiasm in guiding the technologies towards a successful outcome; UQs Dr Ali Malekizadeh who assisted with biochar cleaning protocols, Dr Harshi Gamage who assisted with histology of samples and Prof Elizabeth Aitken, Dr Robyn Cave and Prof Bhesh Bhandari with whom we co-advised PhD and other students; and the 4 PhD, 4 Masters, 5 Honours and 13 undergraduate research students who provided labour and expertise that enabled the immense success of VG15021, with special mention to Kenneth Tryggestad, Aloesi Dakuidreketi, Yunwei Wang and Georgie McGregor, without whom the project would not have been possible. Finally, we wish to acknowledge the excellent help of Dr Gavin Flematti from UWA for synthesising PGR1 for the study and analysing samples for compound quantification.

Appendices

NA