CRDC

FINAL REPORT

For Public Release

Part 1 - Summary Details

CRDC ID: 15-16PRP079

Project Title: Agronomy for Resilient Future Cotton Systems

Project Start Date: 1/07/2015 **Project Completion Date:** 30/06/2019

Research Program: 1 Farmers

Part 2 – Contact Details

Administrator: Ms Suzanne Blankley

Organisation: CSIRO Agriculture and Food

Postal Address: Locked Bag 2, Glen Osmond, SA, 5064

Ph: 08 8303 8709 E-mail: Suzanne.Blankley@csiro.au

Principal Researcher: Dr Michael Bange

Organisation: CSIRO Agriculture and Food

Postal Address: Locked Bag 59, Narrabri, NSW, 2390

Ph: 02 67991540 Fax: 02 67931186 E-mail: Michael.Bange@csiro.au

Signature of Research Provider Representative:

Date submitted: ______________________

Final Report

Background

A key challenge for the Australian cotton industry is to ensure that its' reputation for high quality is maintained and year to year variation in yield is minimised. There is also continued pressure to explore changes in agronomic practice to deal with rising costs, reduced terms of trade, need for improved resource use efficiencies for crop inputs, and in response to technological changes such as new varieties, hormones, and precision agriculture innovations. Recent analyses by the CSIRO cotton breeding program have also demonstrated that yield improvement has been realized from improvements in varieties and in crop management.

To maintain progress, research is needed to update existing agronomic recommendations as well as identify new practices or tools that increase yield and provide resilience to crop stress in both irrigated and dryland systems. A fundamental tool to accomplish this is the ability to accurately assess crop development against growth and management (e.g. nutrition sampling, first irrigation) milestones for that season's climate. Currently, the 'day degree' approach is used to achieve this, but it is not robust to extremes of climate (heat/frost/cold). With the expansion of cotton regions and access to new knowledge on crop development this approach needs refinement. There have also been advances made in formulations of growth hormone and regulant compounds that could assist in managing stresses (water and heat) in cotton. Past research has demonstrated the utility of some of these hormones, but this was done in lower yielding crops in the USA where their use was often not economically viable. Recent successful research in Australia using an ethylene inhibitor on waterlogged cotton to reduce fruit shedding has highlighted that the use of hormones should be reconsidered for both managing stress and assisting with novel approaches to agronomic management to improve resilience and profit.

Objectives

This project will assist industry in developing strategies to minimise the year to year variation in yield and quality in both irrigated and dryland systems, supporting a more consistent financial return for Australian cotton. This should make it more attractive for growers to consider as a crop, raise average yields, and encourage expansion of cotton growing regions.

Industry outcomes: (i) novel agronomic approaches utilising various plant hormones to raise yield and build crop resilience to stress, raising profit in both irrigated and dryland systems (ii) assist in identifying novel dryland cotton systems (iii) An alternative approach to day degree that delivers more precise predictions and assessments of crop development for all cotton regions that will facilitate more accurate growth assessment and management decisions (iv) Maintain crucial independent research capacity in cotton agronomic research.

Science outcomes: (i) Published alternative methodologies on cotton's crop development in response to wider temperature extremes (ii) Develop new understanding of the potential of novel plant growth hormones and regulants in modern Australian cotton systems (iii) Publish strategies that build resilience into cotton systems associated with a variable and changing climate.

Objectives and milestones achieved throughout this project are summarised in Table 1.

Table 1: Objectives and Milestones undertaken and achieved as part of this project.

Obj No.	Objective Description	No.	Milestone Description	Performance Indicator	Start/Finish Date *(dd/mm/yy)*	Achieved Yes/No
1	Improved crop agronomy through industry engagement in research project	**1.1**	Involve CSD E&D and Cottoninfo teams in capturing and extending research in resilient agronomic solutions related to this project	Documented and agreed project engagement plan developed with CSD and the Cottoninfo team.	01/07/2015 30/09/2015	Yes, new ideas were instigated from interactions
		1.2	Provide opportunities for broader industry feedback and direction into cotton agronomic research.	With assistance from CRDC program leaders and the CottonINFO team coordinate 1 workshop/meeting conducted by August 30th each year involving growers (dryland and irrigated) and industry to review outcomes and assist in planning the next research program.	01/07/2015 30/06/2018	Yes, new ideas were instigated from interactions
2	Improve the industry's ability to predict seasonal cotton crop development	**2.1**	Utilising existing data collated from a range of experiments already completed assess three alternative methods to predict crop development	A report generated that compares the predictive ability of alternative methods	01/07/2015 30/06/2016	Yes, A new day degree algorithm has been tested and delivered to industry
		2.2	Using data collected from 4 sites throughout the industry over two seasons validate alternative means of predicting crop development	A report generated that highlights the preferred method of predicting cotton development. Completed in conjunction with an industry extension initiative promoting its use.	01/10/2015 30/06/2017	As above
3	Identify opportunities to utilise plant hormones/growth regulators to improve cotton crop resilience and performance	**3.1**	In consultation with grower/consultant groups identify opportunities and strategies for novel use of hormones for crop management.	Ideas documented in CRDC progress report and used in planning of field experiments.	01/07/2015 30/09/2015	Yes, new ideas were instigated from interactions
		3.2	In consultation with chemical companies identify the most suitable hormones/regulators to be used in research.	Hormones/regulators identified, material transfer agreements in place, and available for research.	01/07/2015 30/09/2015	Access to a range of hormones was provided by companies.
4	Novel approaches for agronomic	**4.1**	Undertake detailed research to assess anti-ethylene agents in	Four comprehensive seasonal evaluations (reflecting more than one	01/10/2015 30/06/2019	Completed

	management utilising plant hormones/regulators validated in the field – Anti-ethylene agents		providing crop resilience to water and heat stress.	field experiment) at ACRI completed assessing a range of anti-ethylene agents with various rates and treatments (water and heat).		
		4.2	Undertake on-farm satellite experiments assess anti-ethylene agents in providing crop resilience to water and heat stress.	Eight (two per season) satellite on-farm experiments completed assessing anti-ethylene agents applied at flowering and cutout.	01/10/2015 30/06/2019	Completed although some experiments were affected by extreme climate events.
		4.3	Undertake research to investigate the combination of anti-ethylene agents in combination with foliar fertilisers to reduce the impact of waterlogging	One experiment conducted at ACRI with various combinations of anti-ethylene agents and foliar fertilisers	01/10/2018 30/06/2019	Experiment was setup but was destroyed by hailstorm at ACRI.
5	Novel approaches for agronomic management utilising plant hormones/regulators validated in the field – Cytokynins for promoting root growth (Initiative was led by Claire Welsh)	**5.1**	Undertake detailed research to assess cytokynins for promoting root growth in limited water situations.	Two comprehensive field experiments at ACRI completed assessing cytokynins with various rates and treatments (water and various skip rows).	01/10/2015 30/06/2019	Yes, experiments initiated by this project, but further detailed work was ultimately led by the PhD project by Claire Welsh. Technical support for her research was provided by this project.
		5.2	Undertake on-farm satellite experiments assessing cytokynins for promoting root growth in limited water situations	Two satellite on-farm experiments completed assessing cytokynins.	01/10/2017 30/06/2019	Yes, experiments initiated by this project but further detailed work was ultimately led by the PhD project by Claire Welsh. Technical support for her research was provided by this project.
6	Novel approaches for agronomic management utilising plant hormones/regulators validated in the field – Hormones for delaying crop growth (Initiative now led by Claire Welsh)	**6.1**	Undertake detailed research to assess use of plant hormones to delay onset of fruiting in a stressed environment.	Two comprehensive field experiments at ACRI completed assessing hormones to delay the onset of reproductive growth with various rates and treatments (irrigated and dryland).	01/10/2016 30/06/2018	Yes, experiments initiated by this project, but further detailed work was ultimately led by the PhD project by Claire Welsh. Technical support for her research was provided by this project.

		6.2	Undertake on-farm satellite experiments assessing hormones to delay onset of fruiting in a stressed environment.	Four (two per season) satellite on-farm experiments completed assessing hormones that delay the onset of reproductive growth.	01/10/2016 30/06/2018	Yes, experiments initiated by this project, but further detailed work was ultimately led by the Phd project by Claire Welsh. Technical support for her research was provided by this project.
7	Novel approaches for agronomic management utilising plant hormones/regulators validated in the field – Auxins to delay fruit opening	**7.1**	Undertake detailed research to assess use Auxins to delay fruit maturation.	A pilot glasshouse experiment completed assessing the effects of Auxins on fruit maturation.	01/10/2017 30/06/2018	Not completed due to demands of other experiments.
8	Communicate results of studies to scientific community and industry	**8.1**	Publish articles and participate in conference and/or industry presentations	1 journal article, 3 cottongrower articles along with related CRDC spotlight articles, and at least one major industry presentation per year.	01/07/2015 30/06/2019	Industry presentations achieved although no specific CottonGrower articles were delivered as results were not complete to enable this. Results of new day degree approach was delivered to industry. Used in development of day degree calculator delivered by Cotton Seed Distributors. Permission to distribute was gained by CRDC.
		8.2	Outcomes delivered to chemical companies to that growers across industry benefit	In consultation with the CRDC discuss and deliver appropriate messages to chemical companies on what opportunities be exploited (exploitation plan developed if necessary)	01/03/2019 30/06/2019	No specific recommendation was generated from products and approaches used in this study. No exploitation plan was developed.

Methods

Key research activities included:

Novel hormones/growth regulants for crop resilience – Experiments at the Australian Cotton Research Institute and satellite experiments across the industry were used to evaluate new plant hormones with an aim to improve crop resilience. Approaches used were based around the used of plant hormones /regulators in the following way:

• Use of anti-ethylene agents (e.g. aminoethoxyvinylglycine (AVG) and 1-Methylcyclopropene (1-MCP)) prevent the production of ethylene to reduce the effects of stresses (e.g. fruit loss associated with heat, water stress and waterlogging).

• The use of cytokynins to promote effective exploration of roots in limited water situations. This research was ultimately led by Claire Welsh and will be reported in her thesis.

• In dryland and water limited situation utilise plant growth regulators to delay plant development such to improve growth and water use. This may allow growers to utilise planting rainfall event but move the fruit growth period of a crop into a less stressful part of the season. This research was ultimately led by Claire Welsh and will be reported in her thesis.

• Undertake a detailed assessment the plant growth hormone (auxin) to delay opening of early bolls without affecting yield.

In all experiments standard commercial cultivars and crop husbandry was undertaken. Specific details are not reported herein. Measurements on experiments undertaken included yield, yield components, fibre quality, crop maturity, and when necessary resource use efficiency (water and/or nutrition). Further detailed methods will be presented in the results section under the key research questions that follow.

New cotton day degree function – New approaches were tested to see if they can better capture cotton development and accommodate extremes in temperatures and different regions. This will reduce reliance on day degree targets and adjustments made using cold shocks. This should improve predictive capabilities within the industry and improve management recommendations that rely on this approach. To validate new day degree modelling approaches developed, existing crop development data (first square, flower, open boll) taken across the industry will be used, with new data collected if necessary.

Results - Novel hormones/growth regulants for crop resilience

Over the course of the four years many experiments were conducted to evaluate key research questions. This was a challenging project where experiments were compromised by hail (1 on-farm experiment in 15/16 season, and most experiments at ACRI in 2018/19 season), extreme cold then extreme heat and disease (verticillium) (all ACRI experiments in the 2016/17 season), waterlogging when not required (1 on-farm experiment in 17/18 season), and extreme rainfall events removing lint from the plant (2 on-farm experiments in Emerald in 2016/17).

Results will be presented and summarised around key research questions which were:

- Can yield and quality be improved on fully irrigated crops using consecutive applications of anti-ethylene agents?

- Can various combinations of anti-ethylene agents reduce the effects of mild stress in irrigated cotton?

- Can anti-ethylene agents improve yield and quality by retaining fruit at cutout using anti-ethylene agents?

- Can the use of anti-ethylene agents help with yield reduction associated with a skipped irrigation?

- Can a combination of anti-ethylene agents and foliar fertiliser reduce the impacts of a waterlogging event? This will be the first research conducted where they will be assessed in combination.

Can yield and quality be improved on fully irrigated crops using consecutive applications of anti-ethylene agents?

To assess this question experiments were conducted at ACRI and at on-farm locations in Emerald Qld (two in 15/16) and at Maules Creek NSW (in 15/16 and in 17/18). Comprehensive experiments were also conducted at ACRI in the 15/16, 16/17, 17/18 and 18/19 cotton seasons (Table 2).

The intent of the multiple application approach is to reduce the impact of carbon stress associated with cotton's continuous flowering pattern that exists throughout the season; and thus, reduce fruit loss and raise retention. Stress in cotton is related to the production of ethylene which triggers the production of abscisic acid that leads to abscission of young fruit. The anti-ethylene chemicals used across all experiments were:

- AVG [aminovinylglycine] – an inhibitor of the conversion of S-adenosylmethionine (SAM) to 1-aminocyclopropane-1-carboxylate (ACC) which is the direct precursor to ethylene.

- 1-MCP (1-Methylcyclopropene) - a competitive inhibitor to ethylene for receptor sites. 1-MCP has a higher affinity for the ethylene receptor sites.

- NAA [naphthalene acetic acid] - a synthetic auxin which downregulates cellulase and polygalacturonase expression, key enzymes in the fruit abscission events. Auxin application is also associated with increased ethylene synthesis.

These anti-ethylene agents AVG and 1-MCP utilise different mechanisms to reduce the effect of ethylene upon plants. AVG acts on the precursors to ethylene production whilst 1-MCP binds to receptors, competitively inhibiting ethylene recognition. Alongside NAA, a synthetic auxin which inhibits key enzymes involved in formation of the fruit abscission zone, various combinations of these plant growth regulators were applied in an attempt to reduce the impact of multiple pathways that can lead to fruit abscission, and ultimately yield losses.

Outline of the ACRI experiments conducted to address the question of the impact of multiple applications are detailed in Table 2 below. In the first season (15/16) the concept was tested with only two hormones (AVG and 1-MCP) applied close to the time of first square and first flower. This was based on research undertaken in Brazil by Brito *et al.* (2013) where effects in rainfed cotton had been recorded. During the project improvements were also made to application technologies (i.e. nozzle changes from large drop size to flat fan) to improve coverage of the chemical on the crop.

Across the four seasons there were three hormones applied at a water rate of 100L/ha:

AVG [aminovinylglycine]	1250ppm (125g ai/ha – 833.3g product)
1-MCP [1-Methylcyclopropene]	250ppm (25g ai/ha – 660g product)
NAA [naphthalene acetic acid]	20ppm (2g ai/ha – 100mL product)

Maxx Organosilicone Surfactant was used with AVG treatments. If temperature was forecasted to exceed 30°C a rate of 50mL/100L was used. If lower than 30°C a rate of 100mL/100L was utilised.

All experiments utilised a Randomised Complete Block Design with 4 replicates. Plot sizes were 4 rows wide and 10m in length. Where experiments had similar treatments across seasons, they were re-randomised within each experiment for each season.

Table 2: Experiments and treatments undertaken to address the research question of '*Can yield and quality be improved on fully irrigated crops using consecutive applications of anti-ethylene agents?'*

Experiment	Year	Location	Treatments	Replication/Plots	Comments
A3 Hormone Timing Trial	2015/16	ACRI	Trt1 Single rate AVG @ 1st Square	4 Reps 28 Plots	
			Trt2 Single rate AVG @ 1st Flower		
			Trt3 @ 1st SquareAVG + 7 days		
			Trt4 @ 1st Flower AVG + 7 days		
			Trt5 1-MCP Std rate @ Flowering		
			Trt6 1 - MCP Double Rate @Flowering + 7 days		
			Control		
Emerald Hormone Timing East	2015/16	Emerald	Trt1 Control	4 Reps 20 Plots	Yield measurements compromised by heavy rainfall at maturity
			Trt2 AVG @ 1St Square + 7 days		
			Trt3 AVG @ 1st Flower + 7 days		
			Trt4 1-MCP @ 1st Square + 7 days		
			Trt5 1-MCP @ 1st Flower + 7 days		
Emerald Hormone Timing West	2015/16	Emerald	Trt1 Control	4 Reps 9 Plots	
			Trt2 AVG @ 1st Square		
			Trt3 1-MCP @ 1st Square		
Bellevue Hormone Timing	2015/16	Maules Creek	Trt 1 Control	4 Reps 20 Plots	Yield measurements compromised by hail at maturity
			Trt 2 AVG @ 1st Square + 7 days		
			Trt 3 AVG @ 1st flower + 7 days		
			Trt 4 1-MCP @ 1st square + 7 days		
			Trt 5 1-MCP @ 1st Flower + 7 days		
D2 Anti-Ethylene Consecutive Spray Trial	2016/17	ACRI	Trt 1 Nil control	4 reps 72 plots	Results compromised by Verticillium outbreak
			Trt 2 AVGx1		
			Trt 3 AVGx2		
			Trt 4 AVGx3		
			Trt 5 AVGx4		
			Trt 6 AVGx5		
			Trt 7 AVGx6		
			Trt 8 AVGx2 FS FF		
			Trt 9 1-MCPx1		
			Trt 10 1-MCPx2		
			Trt 11 1-MCPx3		
			Trt 12 1-MCPx4		
			Trt 13 1-MCPx5		
			Trt 14 1-MCPx6		
			Trt 15 1-MCPx2 FS FF		
			Trt 16 AVG/NAAx3		
			Trt 17 AVG/1-MCPx3		
			Trt 18 1-MCP/NAAx3		
B2 Anti-Ethylene Consecutive Spray Trial	2017/18	ACRI	Trt 1 Nil control	4 reps 72 plots	
			Trt 2 AVGx1		
			Trt 3 AVGx2		
			Trt 4 AVGx3		
			Trt 5 AVGx4		
			Trt 6 AVGx5		
			Trt 7 AVGx6		
			Trt 8 AVGx2 FS FF		
			Trt 9 1-MCPx1		
			Trt 10 1-MCPx2		
			Trt 11 1-MCPx3		
			Trt 12 1-MCPx4		
			Trt 13 1-MCPx5		
			Trt 14 1-MCPx6		
			Trt 15 1-MCPx2 FS FF		
			Trt 16 AVG/NAAx3		
			Trt 17 AVG/1-MCPx3		
			Trt 18 1-MCP/NAAx3		
Bellevue Hormone Timing	2017/18	Maules Creek	Trt 1 Control	4 Reps 20 Plots	Crop waterlogged at a number of times
			Trt 2 AVG @ 1st Square + 7 days		
			Trt 3 AVG @ 1st flower + 7 days		
			Trt 4 1-MCP @ 1st square + 7 days		
			Trt 5 1-MCP @ 1st Flower + 7 days		
A3 Anti-Ethylene Consecutive Spray Trial	2018/19	ACRI	Trt 1 Nil control	4 reps 56 plots	This was a replanted experiments due to the first attempt being destroyed by hail. Not taken through to yield.
			Trt 2 AVGx1		
			Trt 3 AVGx2		
			Trt 4 AVGx3		
			Trt 5 AVGx4		
			Trt 6 AVGx2 FS FF		
			Trt 7 1-MCPx1		
			Trt 8 1-MCPx2		
			Trt 9 1-MCPx3		
			Trt 10 1-MCPx4		
			Trt 11 1-MCPx2 FS FF		
			Trt 12 AVG/NAAx3		
			Trt 13 AVG/1-MCPx3		
			Trt 14 1-MCP/NAAx3		

For ACRI experiments conducted in 16/17 and 17/18 seasons treatments were applied starting from first square and then applied every 10 days following (Table 3). In the 18/19 season following the hailstorm replanting was necessary and only a subset of treatments was implemented (Table 2). These restrictions were a results of planting time restrictions associated with the use of Bollgard III. Note that the treatment numbers in the table do not align but the name of the treatments are consistent across seasons. In addition, due to the lateness of the 18/19 crop only final fruit retention was measure.

Table 3: Treatment combinations and timing used in experiments at ACRI. Beginning at first square (day 0) each plot was sprayed as below. X denotes where a spray was not applied.

Trt	Chemical	1st square	+10 days	+20 days	+30 days	+40 days	+50 days
1	Nil control	x	x	x	x	x	x
2	AVGx1	Spray	x	x	x	x	x
3	AVGx2	Spray	Spray	x	x	x	x
4	AVGx3	Spray	Spray	Spray	x	x	x
5	AVGx4	Spray	Spray	Spray	Spray	x	x
6	AVGx5	Spray	Spray	Spray	Spray	Spray	x
7	AVGx6	Spray	Spray	Spray	Spray	Spray	Spray
8	AVGx2 FS FF	Spray	x	Spray	x	x	x
9	1-MCPx1	Spray	x	x	x	x	x
10	1-MCPx2	Spray	Spray	x	x	x	x
11	1-MCPx3	Spray	Spray	Spray	x	x	x
12	1-MCPx4	Spray	Spray	Spray	Spray	x	x
13	1-MCPx5	Spray	Spray	Spray	Spray	Spray	x
14	1-MCPx6	Spray	Spray	Spray	Spray	Spray	Spray
15	1-MCPx2 FS FF	Spray	x	Spray	x	x	x
16	AVG/NAAx3	Spray	Spray	Spray	x	x	x
17	AVG/1-MCPx3	Spray	Spray	Spray	x	x	x
18	1-MCP/NAAx3	Spray	Spray	Spray	x	x	x

Results

Results of all experiments addressing the question whether yield and quality could be improved on fully irrigated crops using consecutive applications of anti-ethylene agents are summarised in Table 4. A considerable number of these experiments were compromised during the project which meant that there were not conclusive or consistent outcomes generated.

In the 15/16 season fibre length was affected although the treatments that generated differences had fibre length less than the control.

In the 16/17 season there were significant differences generated for lint yield and gin-turnout. The lint yields varied by 18% across the treatments, although there were no treatments that had a lint yield greater than the control (lint yield 11.85 bales/ha). There was also no relationship between gin-turnout and the final lint yield. No other variables were significant and were related to yield. Note that this experiment was heavily compromised by a verticillium wilt outbreak in this field that noticeably affected some plots more than others. There was no relationship between verticillium wilt affected plots and treatments.

In the 17/18 season seed cotton yield and fibre micronaire were significantly different. Seed cotton yield was improved by 13% by 5 treatments over the control (seed cotton yield 0.66 kg/m^2). The treatments were 1-MCPx6, AVGx1,1-MCPx1,1-MCPx2 FS FF, and 1-MCP/NAAx3. This did not however, translate into significant differences in lint yield. once ginning had occurred. It is possible subtle changes in boll weight, ginning, and boll number together did not allow for these differences to be translated into lint yield. Micronaire was also affected across the treatments by a difference of 11%, where treatments (AVGx5, 1-MCPx2 FS FF) were significantly different from the control (micronaire 4.99) they had micronaire that was high and would have incurred financial discounts. In the 18/19 season where only final fruit retention was measured and there were no significant differences detected.

Table 4: Summary of results of the consecutive spraying experiments conducted in this project. NS (no significant difference) at the 95% level of confidence; * significant difference at the 95% level of confidence; ** significant difference at the 99% level of confidence. P<0.1 is significant difference at the 90% level of confidence. A dash denotes that a measurement was not taken.

Experiment	Seed-Cotton Yield	Boll Mass	Boll Number	Gin-Turnout	Fibre Lint Yield	Fibre Micronaire	Fibre Length	Fibre Strength
15/16	NS	NS	NS	NS	NS	NS	**	NS
16/17	NS	NS	NS	P<0.1	*	NS	NS	NS
17/18	*	NS	NS	NS	NS	P<0.1	NS	NS
18/19	-	-	-	-	-	-	-	-
Emerald 15/16 East	NS	NS	NS	-	-	-	-	-
Emerald 15/16 West	NS	NS	NS	-	-	-	-	-
Maules Creek 15/16	NS	NS	NS	-	-	-	-	-
Maules Creek 17/18	NS	NS	NS	-	-	-	-	-

To better understand why there may have been little difference in lint yield across experiments correlation analysis was conducted to identify if there were any yield components (gin-turnout, boll size or boll number) related to yield. It was postulated that small differences in these components may be a reason for lack of significant differences. This analysis was conducted on each of the large experiments at ACRI. We established that there was a significant relationship between boll size and boll number in the 17/18 season (the only complete experiment that was unaffected by external factors). Where boll size was reduced there were more bolls (see Figure 1). This highlights that there may be compensatory mechanisms at play with the retention of more fruit. That is, when more fruit are retained, they produce smaller bolls negating the effects of the hormone treatments.

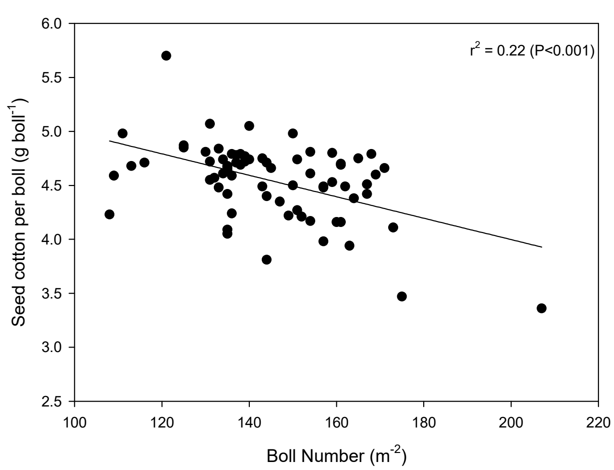

Figure 1: The significant linear response of boll size with changes in boll number in the consecutive spraying experiment conducted in the 17/18 season. It is possible that the lack of lint yield responses is because of compensatory mechanisms at play where the retention of more bolls simply reduces the size of the bolls already present.

Can various combinations of anti-ethylene agents reduce the effects of mild stress in irrigated cotton?

To assess this question experiments were conducted at ACRI in 15/16, 16/17, and 17/18 seasons. Applications attempted to alleviate the impact of a stress event post first flower. Applications were applied as a pre-emptive response to post-anthesis skipped irrigations. As for the previous question the same anti-ethylene chemicals were used at the same rates. All experiments utilised a Randomised Complete Block Design with 4 replicates. Plot sizes were 4 rows wide and 10m in length. Where experiments had similar treatments across seasons, they were re-randomised each season. Details of treatments applied is detailed in Table 5.

Table 5: Experiments and treatments undertaken to address the research question of '*Can various combinations of anti-ethylene agents reduce the effects of mild stress in irrigated cotton?*'

Experiment	Year	Location	Treatments	Replication	Timing/Rates
A3 Hormone Timing Trial	2015/16	ACRI	Trt1 = Single rate @ 1st Square	4 Reps 28 Plots	7 Trts @ 1st Square or 1st flower + some 7days later 1MCP after 1st Flower. Water stressed (Missed 3 irrigations).
			Trt2 Single rate @ 1st Flower		
			Trt3 @ 1st Square + 7 days		
			Trt4 @ 1st Flower + 7 days		
			Trt5 1-MCP Std rate @ Flowering		
			Trt6 1-MCP Double Rate @Flowering + 7 days		
			Trt7 Nil-Control		
D2 PGR Combos	2016/17	ACRI	Trt1 Nil - Control	4 reps 32 plots	8 trts with 3 different chemicals with skipped irrigation at time of application (post 1st flower)
			Trt2 AVG		
			Trt3 1-MCP		
			Trt4 NAA		
			Trt5 AVG/NAA		
			Trt6 AVG/1-MCP		
			Trt7 1-MCP/NAA		
			Trt8 AVG/1-MCP/NAA		
B2 PGR Combos	2017/18	ACRI	Trt1 Nil - Control	4 reps 32 plots	8 trts with 3 different chemicals with skipped irrigation at time of application (post 1st flower)
			Trt2 AVG		
			Trt3 1-MCP		
			Trt4 NAA		
			Trt5 AVG/NAA		
			Trt6 AVG/1-MCP		
			Trt7 1-MCP/NAA		
			Trtr8 AVG/1-MCP/NAA		

*Note that the 16/17 experiment was compromised with Verticillium wilt.

Results

Results of all experiments addressing the question of whether yield and quality could be improved with various combinations of anti-ethylene where mild water stress occurs in irrigated cotton are summarised in Table 6.

Across all three experiments the only variable that was affected was fibre micronaire in the 15/16 and 16/17 season. In the 15/16 season the single rate of AVG at first square (micronaire 4.26) and the single rate of 1-MCP at first flower (micronaire 4.19) had micronaire that was significantly less than the control (micronaire 4.58). In the 16/17 season the single application of Auxin at first flower had a micronaire (4.99) that was greater than the control (4.82).

Table 6: Summary of results of the experiments conducted in this project to investigate the impact of ant-ethylene agents to reduce the effects of mild stress. NS (no significant difference) at the 95% level of confidence; * significant difference at the 95% level of confidence; ** significant difference at the 99% level of confidence. P<0.1 is significant difference at the 90% level of confidence.

Experiment	Seed-Cotton Yield	Boll Mass	Boll Number	Gin-Turnout	Fibre Lint Yield	Fibre Micronaire	Fibre Length	Fibre Strength
15/16	NS	NS	NS	NS	NS	P<0.1	NS	NS
16/17	NS	NS	NS	NS	NS	P<0.1	NS	NS
17/18	NS	NS	NS	NS	NS	NS	NS	NS

Can anti-ethylene agents improve yield and quality by retaining fruit at cutout using anti-ethylene agents?

To assess this question experiments were completed at ACRI in 16/17, 17/18, and 18/19 seasons (Table 7). The experiment in 18/19 at ACRI was destroyed by hail and was not completed. Off station experiments were conducted in all seasons from 15/16 until 18/19 and numerous locations (Table 7). Results of off station experiments in 15/16 were compromised by hail and extremely heavy rainfall at harvest. The experiment at Bellevue was heavily waterlogged mid flowering.

Applications attempted to reduce the impact of abscission of fruit at cutout due to known increases of ethylene leading to the production of abscisic acid that triggers fruit abscission.

As for the previous question the same anti-ethylene chemicals were used at the same rates (see above). Mepiquat chloride (Pix) was applied with a moderate cutout rate (1l/ha). The four hormones were applied at a water rate of 100L/ha at the time of cutout (6 nodes above white flower); 2nd application occurs at 4 NAWF. All experiments utilised a Randomised Complete Block Design with 4 replicates. Plot sizes were 4 rows wide and 10m in length.

Table 7: Experiments and treatments undertaken to address the research question of '*Can anti-ethylene agents improve yield and quality by retaining fruit at cutout using anti-ethylene agents?'*

Experiment	Year	Location	Treatments	Replication	Timing/Rates	Comments
Emerald Hormone Timing East	2015/16	Emerald	Trt1 Nil-Control	4 Reps 8 Plots	Handpick results only.	Yield measurements compromised by hail at maturity
			Trt2 AVG @ Cutout			
Emerald Hormone Timing West	2015/16	Emerald	Trt1 Nil-Control	4 Reps 12 Plots	Handpick results only.	Yield measurements compromised by hail at maturity
			Trt2 AVG @ Cutout			
			Trt3 1- MCP @ Cutout			
Bellevue Hormone Timing	2015/16	Maules Creek	Trt1 Nil-Control	4 Reps 8 Plots	Handpick results only	Yield measurements compromised by hail
			Trt2 AVG @ Cutout			
D2 Cut out Trial	2016/17	ACRI	Trt1 Nil - Control	4 reps 20 plots	6 trts of different hormones applied at cut out	
			Trt2 AVG/1-MCP			
			Trt3 1-MCP/NAA			
			Trt4 AVG/NAA			
			Trt5 AVG/1-MCP/NAA			
B2 Cut out Trial	2017/18	ACRI	Trt1 Nil-Control	4 reps 36 plots	9 trts of different hormones/rates applied at cut out, 2X means there was a second application	
			Trt2 AVG/1-MCP			
			Trt3 1-MCP/NAA			
			Trt4 AVG/NAA			
			Trt5 AVG/1-MCP/NAA			
			Trt6 PIX			
			Trt7 2 x AVG/1-MCP			
			Trt8 AVG/PIX			
			Trt9 2 x AVG/1-MCP/NAA			
Bellevue Hormone Timing	2017/18	Maules Creek	Trt 1 Nil-Control	4 Reps 8 Plots	6 Trts with hormones applied @ different times. Handpick results only	Crop waterlogged at a number of times
			Trt2 AVG @ Cutout			
Cudgewa Cut Out Trial	2018/19	Wee Waa	Trt1 Nil-Control	4 reps 24 plots	6 trts of different chemicals applied at cut out, 2X means there was a second application	
			Trt2 AVG/1-MCP			
			Trt3 PIX			
			Trt4 2 x AVG/1-MCP			
			Trt5 AVG/PIX			
			Trt6 2 x AVG/1-MCP/NAA			

Results

Results of all experiments addressing the question of whether anti-ethylene agents can improve yield and quality by retaining fruit at cutout using anti-ethylene agents are summarised in Table 8. Across all experiments and season there was ability of the hormone treatments to influence yield or fibre quality outcomes. This was despite attempts to change the product and/or increase concentrations of the product within the canopy.

Table 8: Summary of results of the consecutive spraying experiments conducted in this project. NS (no significant difference) at the 95% level of confidence; * significant difference at the 95% level of confidence; ** significant difference at the 99% level of confidence. P<0.1 is significant difference at the 90% level of confidence.

Experiment	Seed-Cotton Yield	Boll Mass	Boll Number	Gin-Turnout	Fibre Lint Yield	Fibre Micronaire	Fibre Length	Fibre Strength
Emerald East 15/16	NS	NS	NS	-	-	-	-	-
Emerald West 15/16	NS	NS	NS	-	-	-	-	-
Bellevue Maules Creek 16/17	NS	NS	NS	-	-	-	-	-
ACRI D2 Cutout 16/17	NS	NS	NS	NS	NS	NS	NS	NS
ACRI B2 Cutout 17/18	NS	NS	NS	NS	NS	NS	NS	NS
Bellevue Maules Creek 17/18	NS	NS	NS	-	-	-	-	-
Cudgewa 18/19	NS	NS	NS	-	-	-	-	-

*Note that all experiments in the 15/16 season were compromised by hail or very heavy rainfall prior to harvest.

Can a combination of anti-ethylene agents and foliar fertiliser reduce the impacts of a waterlogging event?

This was the first research attempted where both types of products have been used in combination in an attempt to reduce waterlogging impacts. Past research has shown that each used separately applied to a crop before a significant waterlogging event can reduce the severity of impacts. This experiment was only attempted in the last season of the project. Following the hailstorm in Dec 2018 the experiment was planted late in January. As the experiment was planted so late there was no intention to take the experiment through to yield.

Due to restriction in the area of replant associated with Bollgard 3 licence conditions the treatments in the experiment were not fully replicated. There was a waterlogged and non-waterlogged area with two plots of each treatment in those areas. The outcome was to assess final fruit retention and to observe if any differences could be detected.

Treatments were applied on both non-waterlogged and waterlogged areas the day before the waterlogging treatment soon after first flower. Waterlogging was achieved by continuing the irrigation for a full 48 h. Treatments were control (no application of product), AVG standard rate, Foliar fertiliser, and AVG+Foliar fertiliser. The rate of AVG is the same rate used in other experiments described above. The foliar product was called K-Flow (a liquid potassium nitrate fertiliser by Yarra (4% N, 12% K)) and was applied at the rate used was 30L/ha.

Data collected was highly variable and no discernible differences were observed.

Discussion - Novel hormones/growth regulants for crop resilience

The conditions in which this project was undertaken was challenging with the climate extremes experienced. Variability within many experiments was far greater than effects caused by the treatments making it difficult to discern any consistent treatment effects.

It was hoped based on the waterlogging experiments conducted in the past that rates and timings would have led to differences. These results potentially highlight that unless there is a severe stress imposed (like a waterlogging event) to prevent significant fruit loss there may be little utility in retaining fruit in less stressful situations. Lack of differences could simply be a result of cotton's ability to compensate the loss of fruit to allow assimilates to support the growth of existing fruit (resulting in larger fruit; evidenced in this study). This is a known mechanism that cotton uses to overcome stress in milder situations. In these experiments there were no significant stress events like that of an extreme waterlogging event. Severe stress caused by water deficits and any response to the hormones used in this study is being investigated by Claire Welsh. Opportunities to quantify plant stress using canopy temperature sensors that separate soil and plant temperatures will offer opportunities in the future to support these studies. Overall at the present time given the current high cost of these hormones, the multiple application strategies that generated differences would be currently cost prohibitive. Future research should be explored in more controlled conditions, with greater replication, and along with an explicit ability to quantify the stressed conditions. The ability to utilise a technique that can quantify the ethylene hormone response will also aid this research.

At this time no clear recommendation of the use of these growth regulators to answer the questions addressed in this study can be made.

Results – New Cotton Day Degree Function

Another significant objective of the project was to seek new ways to improve the industry's ability to predict crop development. During the course of this project we have compiled data from multiple seasons where first square, first flower, and sometimes first open boll were recorded. Data was collated from both Australian and USA locations.

We compared a number of approaches: 1. The existing industry day degree approach and targets; 2. A modified approach using the existing approach with a maximum temperature threshold and existing thresholds; 3. A published method used in the USA in Arizona; and 4. A method that uses an alternative approach calculating a rate of progress from data measured in the Canberra Phytoton previously published by Bange and Milroy (2001).

This study was able to demonstrate that there were improvements in the predictability of time of first square and first flower measured in cotton crops. Two functions were able to better predict these phenological stages compared to the existing function used currently in the Australian industry (Constable and Shaw, 1988). The best performing functions were a variable temperature day degree function that used a base temperature of 15.6 °C and an optimum of 32 °C, and a physiological rate function that reflected similar temperature characteristics as the variable function. The use of these functions should be considered in the development of new cotton crop predictive capabilities as they will be able to account for more temperature extremes (high and low, that maybe more prevalent in a changing climate) and where cotton production moves into new regions. The analyses of functions here also support the use of a base temperature of 15.6 °C (60 °F) used in USA cotton systems.

Michael Bange began promotion of the understanding relating to the use of these new functions throughout the industry. He presented outcomes at the 2018 Crop Consultants Association's meeting in Narrabri and at the Association of Australian Cotton Scientist conference also in 2018. An industry Youtube video was also developed on the use of day degree functions and included outcomes generated in this study. The approach was shared with Steve Buster who used this in his analysis of data across the southern regions. CSD have implemented the new function as part of their online suite of agronomy tools. Comprehensive detail is provided in the document provided with this report.

Project Extension

Industry Presentations

- Presentation at a Dryland Association meeting at Bellata in July 2016 (approximately 40 attendees).
- Presentation at a rain-fed field day at Bellata 2017 (47 participants)
- Walgett Field Day 5th April 2017– Cotton Physiology Topics – Approx. 30 Attendees.
- Liverpool Plains grower and consultant meeting Dec 15th 2017 - Cotton Physiology Topics - Approx. 30 Attendees.
- Presentation to Cotton Consultants Australia Dubbo Jun 2017– Physiology of Cotton – Elements of research undertaken in this project were presented.
- Verbal Presentation to the Texas High Plains Cotton Growers Association – Aug 2017 – Approx. 100 people on the collaborative research being undertaken on cotton crop development between USDA and CSIRO.
- Verbal Discussion to Dryland Cotton Growers Spring Ridge (Aug 2018) – Approx. 30 people.
- Darling Downs Field Day 28th Feb 2018 – Cotton Defoliation > 100 attendees
- Crop simulations provided to dryland cotton growers association on impacts of planting time on yield and associated risk – Nov 2018.
- Macintyre Field Day 14th Mar 2018 – Cotton Defoliation > 100 attendees
- Crop Consultants Australia meeting in Narrabri in August 2018 – A presentation on cotton crop development and temperature impacts on production.
- CSD Field Day – 27th Mar 2019 – End of Season Management and discussion on research into growth regulators.
- Australian Cotton Collective Sep 2019 – Plenary Speaker – Trends affecting Australian Cotton Production.

Research Presentations

- Presentation and Paper at the Australian Agronomy Conference – Sep 2017 - Assessment of individual fruit maturation (boll) period in modern cotton.
- Presentation and Abstract at the Association of Australian Cotton Scientists – Aug 2017 - Re-evaluating the Prediction Models for Cotton Development
- Guest Presentation at the Beltwide Cotton Conference Jan. 2018 – Cotton Physiology
- Australian Agronomy Conference 2019 - Validation of novel in-field monitoring techniques to assist harvest aid timing in cotton.
- Presentation and Abstract at the Association of Australian Cotton Scientists – Oct 2019 - Predicting Micronaire Accounting for Integrated Effects of Environment and Crop Growth.
- Presentation at the Association of Australian Cotton Scientists – Oct 2019 – Trends affecting future cotton production.
- International Cotton Advisory Committee Dec 2019 – Climate Change and Future Cotton Production.

Others

- Cotton Crop Physiology Lecture to Sydney University Students – 2017 to 2019
- Cottoninfo Youtube videos – Defoliation Topics and Cutout principles, and Day Degrees.
- Revision of Australian Cotton Production Chapters (6 chapters per year) on rainfed Production, Cotton Physiology, use of growth regulators, managing for quality, preparing for harvest, and harvesting

Publications and Online Resources

No specific publications resulted from the growth regulator/hormone research. The full report on the day degree research is attached to this report. M. Bange is currently developing a refereed publication for an international journal on the day degree research. Chris Nunn (CSIRO) is assisting with this publication.

The day degree function developed in this project has been provided to Cotton Seed Distributors and they have developed an online calculator (see below). The CRDC has been acknowledged supporting the research to generate this.

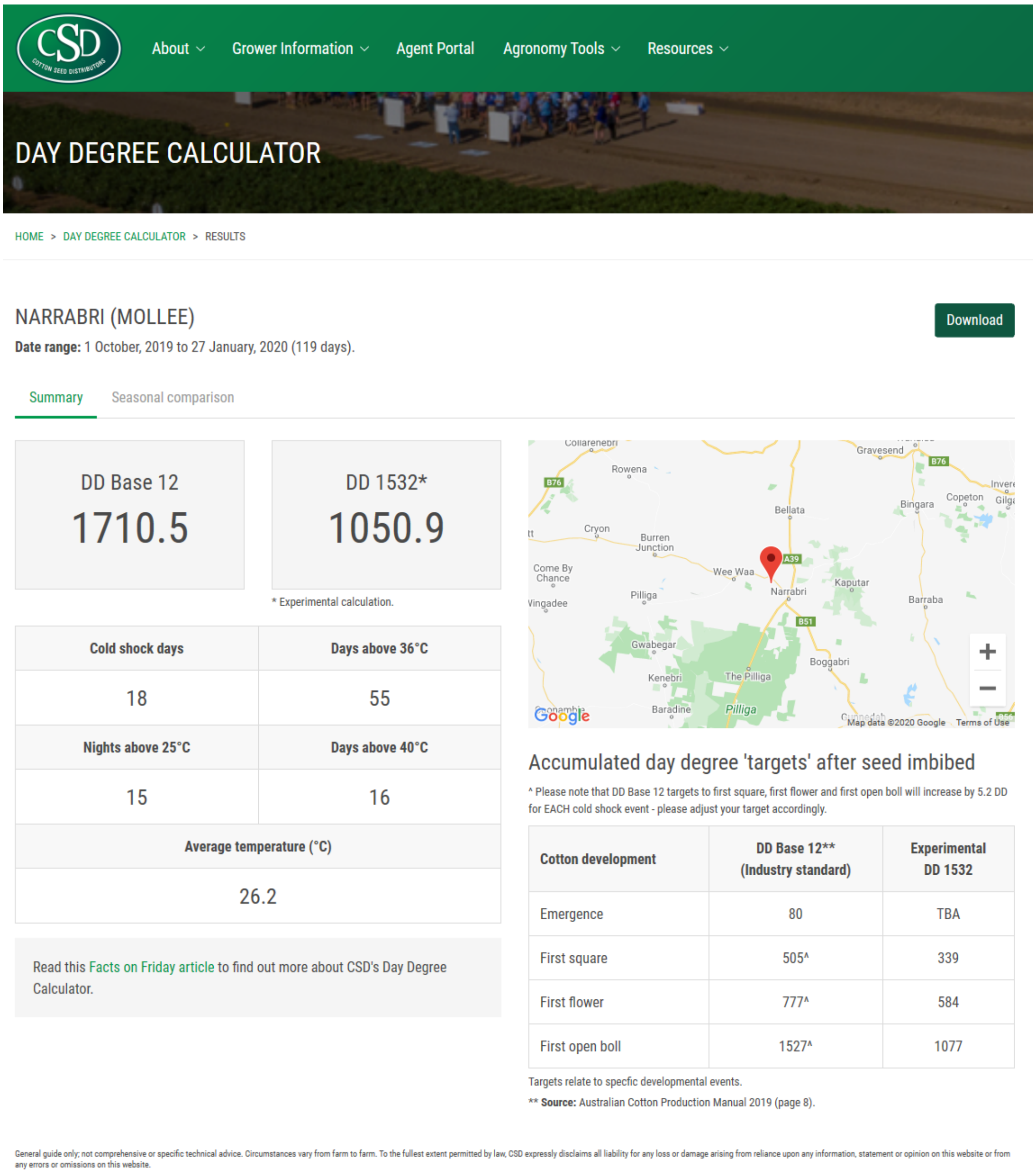

Figure: A snapshot of the Cotton Seed Distributor’s day degree calculator which includes the predictions made using the new day degree function developed in this study.

A key challenge for the Australian cotton industry is to ensure that its' reputation for high quality is maintained and year to year variation in yield is minimised. There is also continued pressure to explore changes in agronomic practice to deal with rising costs, reduced terms of trade, need for improved use efficiencies for crop inputs, and in response to technological changes such as new varieties, plant hormones, and precision agriculture innovations.

To maintain progress, research is needed to update existing agronomic recommendations as well as identify new practices or tools that increase yield and provide resilience to crop stress in both irrigated and dryland systems. There have been advances made in growth hormone and regulant compounds that could assist in managing stresses (water and heat) in cotton. Past research has demonstrated the utility of some of these hormones, but this was done in lower yielding crops in the USA where their use was often not economically viable. Recent successful research in Australia using an ethylene inhibitor on waterlogged cotton to reduce fruit shedding has highlighted that the use of hormones should be reconsidered for both managing stress and assisting with novel approaches to agronomic management to improve resilience and profit.

This project addressed the following research objectives (i) investigate whether the use of novel agronomic approaches utilising various plant hormones could raise yield and build crop resilience to stress, raising profit in both irrigated and dryland systems; (ii) assess an alternative approach to day degree that delivers more precise predictions and assessments of crop development for all cotton regions that will facilitate more accurate growth assessment and management decisions; and (iii) Maintain build crucial independent research capacity in cotton agronomic research through the support of Claire Welsh's PhD studies in rainfed cotton systems.

Growth regulator/hormone research - Over the course of the four years many experiments were conducted to evaluate key research questions. This was a challenging project where experiments were compromised by hail (1 on-farm experiment in 15/16 season, and most experiments at ACRI in 2018/19 season), extreme cold then extreme heat and disease (verticillium) (all ACRI experiments in the 2016/17 season), waterlogging when not required (1 on-farm experiment in 17/18 season), and extreme rainfall events removing lint from the plant (2 on-farm experiments in Emerald in 2016/17).

Research addressed the following key questions:

- Can yield and quality be improved on fully irrigated crops using consecutive applications of anti-ethylene agents?

- Can various combinations of anti-ethylene agents reduce the effects of mild stress in irrigated cotton?

- Can anti-ethylene agents improve yield and quality by retaining fruit at cutout using anti-ethylene agents?

- Can the use of anti-ethylene agents help with yield reduction associated with a skipped irrigation?

- Can a combination of anti-ethylene agents and foliar fertiliser reduce the impacts of a waterlogging event? This was the first research conducted where they will be assessed in combination.

The conditions in which this project was undertaken was challenging with the climate extremes experienced. Variability within many experiments was far greater than effects caused by the treatments making it difficult to discern any consistent treatment effects.

It was hoped based on the waterlogging experiments conducted in the past that rates and timings would have led to differences. These results potentially highlight that unless there is a severe stress imposed (like a waterlogging event) to prevent significant fruit loss there may be little utility in retaining fruit in less stressful situations. Lack of differences could simply be a result of cotton's ability to compensate the loss of fruit to allow assimilates to support the growth of existing fruit (resulting in larger fruit; evidenced in this study). This is a known mechanism that cotton uses to overcome stress in milder situations. Overall at the present time, and given the current high cost of these hormones, the multiple application strategies that generated differences would be currently cost prohibitive. Future research should be conducted in more controlled conditions, with greater replication, and with an explicit ability to quantify the stressed conditions. Ability to utilise a technique that can quantify the ethylene hormone response would also aid this research. Therefore, at this time no clear recommendation of the use of these growth regulators to answer the questions addressed in this study can be made.

New Day Degree Calculator - Key management recommendations rely on accurate estimates of crop development and boll periods using the day degree approach. The day degree approach is a fundamental tool used to assess crop development against growth and management (eg nutrition sampling, first irrigation) milestones for that particular season's climate. Currently, the 'day degree' approach is not robust to accommodate extremes of climate (heat/cold). There is a need to refine this approach to ensure the accuracy of this critical tool to accommodate temperature extremes and ensure we can use it confidently for management decisions in new cotton regions (eg. Griffith). New approaches will be developed to accommodate temperature extremes improving predictive capabilities and management recommendations that rely on this approach.

During the course of this project we have compiled data from multiple seasons where first square, first flower, and sometimes first open boll were recorded. Data was collated from both Australian and USA locations. We compared a number of approaches: 1. The existing industry day degree approach and targets; 2. A modified approach using the existing approach with a maximum temperature threshold and existing thresholds; 3. A published method used in the USA in Arizona; and 4. A method that uses an alternative approach calculating a rate of progress from data measured in the Canberra Phytoton previously published by Bange and Milroy (2001).

This study was able to demonstrate that there were improvements in the predictability of time of first square and first flower measured in cotton crops. Two functions were able to better predict these phenological stages compared to the existing function used currently in the Australian industry (Constable and Shaw, 1988). The best performing functions were a variable temperature day degree function that used a base temperature of 15.6 °C and an optimum of 32 °C, and a physiological rate function that reflected similar temperature characteristics as the variable function. The use of these functions should be considered in the development of new cotton crop predictive capabilities as they will be able to account for more temperature extremes (high and low, that maybe more prevalent in a changing climate) and where cotton production moves into new regions. The analyses of functions here also support the use of a base temperature of 15.6 °C (60 °F) used in USA cotton systems.

Michael Bange began promotion of the understanding relating to the use of these new functions throughout the industry. An industry you tube video was also developed on the use of day degree functions and included outcomes generated in this study. CSD have also implemented the new function as part of their online suite of agronomy tools