Project title:	Blackcurrants: Hot water treatment (HWT) of cuttings
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Project leader:	John Atwood, ADAS
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Previous report:	None
Key staff:	John Atwood, ADAS Harriet Roberts, ADAS Chris Dyer, ADAS
Location of project:	ADAS Boxworth
Industry Representative:	Rob Saunders, GSK
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The results and conclusions in this report are based on an investigation conducted over a one-year period. The conditions under which the experiments were carried out and the results have been reported in detail and with accuracy. However, because of the biological nature of the work it must be borne in mind that different circumstances and conditions could produce different results. Therefore, care must be taken with interpretation of the results, especially if they are used as the basis for commercial product recommendations.

AUTHENTICATION

We declare that this work was done under our supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

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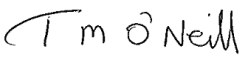

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GROWER SUMMARY

Headline

- Hot water treatment of four cultivars of blackcurrant cuttings (up to 50^{o}C for up to 20 minutes) did not improve establishment of cuttings.

Background and expected deliverables

The objective of this project was to evaluate different water temperatures and durations of hot water treatment (HWT) to cuttings, for crop safety and potential beneficial effects of establishment and vigour on four varieties of blackcurrant, Ben Gairn, Ben Starav, Ben Dorain and Ben Tirran.

Heat treatment is an established method used to eradicate diseases and pests from plant material and has been utilised in many crops. In blackcurrant, hot water treatment has been used in Denmark and Finland for eradication of gall mite, by immersing cuttings in water baths at 45ºC for 20 minutes. HWT is also used in grape vines for the eradication of various pests and diseases including nematodes and *Phytophthora*. However there can be a narrow temperature/duration margin between achieving pest eradication and damaging the cutting. Different vine varieties have been shown to have different tolerances to temperature and duration of HWT. To investigate whether blackcurrant varieties also vary in their tolerance to HWT, four commonly grown newer cultivars were selected for this trial. Researchers have also reported incidental improvements in crop vigour following HWT regimes when used for pest eradication. As cutting establishment and vigour is important to ensuring plantations are brought into crop on target, this was a further aspect investigated in this project.

Summary of the project and main conclusions

Cuttings of four varieties Ben Gairn, Ben Starav, Ben Dorain and Ben Tirran were kept in a cold store until treatment on 16 April 2012. Cuttings were removed from cold store and allowed to reach room temperature before immersing in a hot water bath, according to the treatment schedule (Table 1). After the allotted times, blackcurrants were removed and rapidly brought back to room temperature, before planting into 3 L pots filled with a sterilised loam. Irrigation was supplied overhead and pesticides were applied as required.

Table 1. Treatment schedule for HWT applied to four cultivars of blackcurrant cuttings, 16 April 2012, ADAS Boxworth

Treatment number	Temperature (^{o} C)	Duration (minutes)
1	Untreated (not soaked)	-
2	Ambient	5
3	Ambient	10
4	Ambient	15
5	Ambient	20
6	40	5
7	40	10
8	40	15
9	40	20
10	45	5
11	45	10
12	45	15
13	45	20
14	50	5
15	50	10
16	50	15
17	50	20

Establishment and growth stage was assessed on 5 May 2012, three weeks after planting and was scored according to blackcurrant growth stages. At this first assessment, establishment was similar across treatments 1 to 12. However for treatments 13-15 (45^{o}C for 20 minutes and all 50^{o}C treatments) there was a significant delay in establishment or an increase in the proportion of dead plants. Shoot growth was assessed on four occasions through the growing season. Figures 1 and 2 show the average growth across all the varieties at both the June assessment and at the final growth assessment carried out in October. Table 2 shows the same data but split by variety. The same result was clear across all assessments and varieties. None of the water treatments (with or without heat) enhanced establishment or growth when compared to an untreated cutting. There were, however, significant effects of hot water treatment, with growth suppression occurring at higher temperatures and longer durations. At 45^{o}C for 20 minutes there was some initial growth reduction for varieties apart from Ben Tirran. At 50 ^{o}C, there was significant growth suppression at all soak durations, with cutting death occurring at durations of 15 and 20 minutes. Ben Tirran appeared to be the most resilient to the hottest treatment showing more consistent growth at 50 ^{o}C for up to 15 minutes than the other three varieties; Ben Gairn showed marginally less heat tolerance.

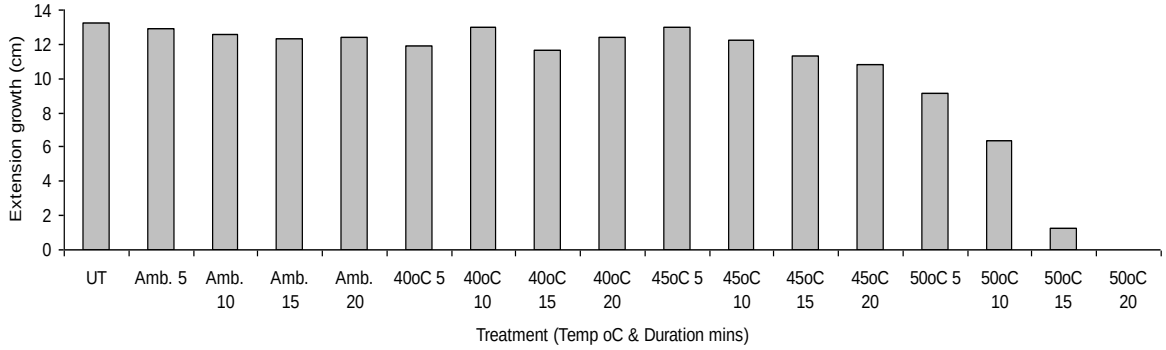

Figure 1. Effect of hot water treatment on average growth (cm) of blackcurrant cuttings (averaged across four varieties) – June 2012

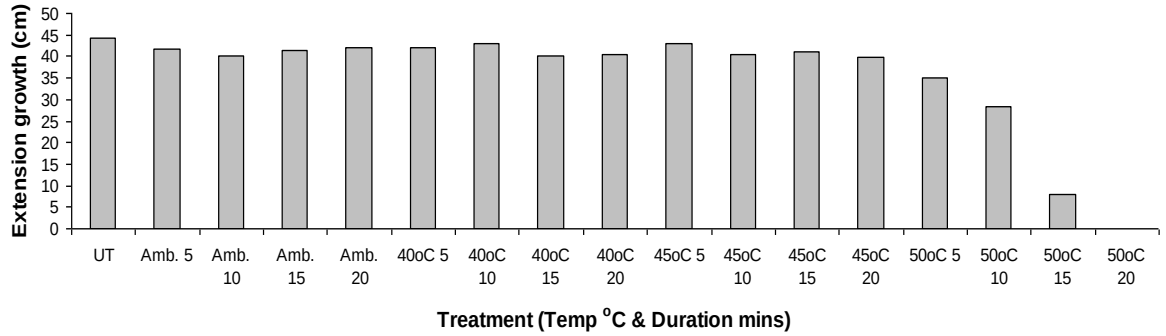

Figure 2. Effect of hot water treatment on average growth (cm) of blackcurrant cuttings (averaged across four cultivars) - October 2012

Table 2. Effect of hot water treatment on average growth of four cultivars of blackcurrant cuttings - assessment 4, October 2012

	Average shoot growth in cm			
Treatment	**Ben Gairn**	**Ben Starav**	**Ben Dorain**	**Ben Tirran**
Untreated	52.35	41.15	45.40	38.10
Ambient 5 min	48.95	34.95	43.75	38.75
Ambient 10 min	45.30	37.05	43.80	34.90
Ambient 15 min	49.50	39.65	43.35	33.05
Ambient 20 min	51.35	39.20	41.80	35.45
40°C 5 min	50.45	40.95	43.25	33.45
40 °C 10 min	52.20	43.50	43.55	33.00
40 °C 15 min	50.65	36.75	40.25	32.45
40 °C 20 min	48.90	37.00	42.30	33.05
45 °C 5 min	52.35	42.90	40.80	35.60
45 °C 10 min	47.90	35.50	42.45	35.50
45 °C 15 min	48.50	37.85	40.65	37.30
45 °C 20 min	44.00	36.70	42.70	35.40
50 °C 5 min	30.40	33.15	40.25	36.40
50 °C 10 min	19.70	21.50	32.15	40.50
50 °C 15 min	2.30	1.70	5.05	23.25
50 °C 20 min	0.00	0.00	0.00	0.00

Financial benefits

This project did not demonstrate improved establishment and vigour resulting from soaking cuttings. Therefore any financial benefit would have to be associated with pest or disease eradication. Establishing a safe and practical method for heat treating cuttings may enable cutting material to be produced with reduced pest and disease pressure from pathogens such as gall mite and *Phomopsis ribicola.* The Plant Health Propagation Scheme (PHPS) in the UK already minimises the level of gall mite contamination in propagation material. Reduction in inoculum of *Phomopsis ribicola,* however, would have a large financial benefit for growers. Further research is required to establish the significance of cutting infection versus other infection routes and also to establish the temperatures needed to eradicate this specific pathogen and to make the process practical on a larger scale.

Action points for growers

At present there are no action points. Adoption of HWT would first require further research to confirm the role of cutting infection in spread of *Phomopsis ribicola*, then to establish the temperature required for eradication.

SCIENCE SECTION

Introduction

Heat treatment is an established method used to eradicate diseases and pests from plant material and has also been reported to improve establishment and vigour in cutting material. Blackcurrant cultivars are known to respond differently to hot water treatment (HWT) regimes and to date HWT has not been tested on the main UK cultivars. This one year project evaluates the safety and benefit of different hot water treatments (HWT) regimes to four commercial standard cultivars, assessing cutting establishment and vigour in the first season.

Heat treatment is utilised in many crops for control of a range of pests and diseases. In blackcurrant, HWT has been used in Denmark (Pedersen *et al.,* 2009) and Finland (Tuovinen *et al.,* 2008) for eradication of gall mite by immersing cuttings in water baths at 45 ºC for 20 minutes. HWT is also used in grape vines for the eradication of various pests and diseases, including nematodes and Phytophthora (Waite & Morton, 2007). However there can be a narrow temperature/duration margin between achieving pest/disease eradication and damaging the cutting. Different vine varieties have been shown to have different tolerances to temperature and duration of HWT (Waite & Morton 2007).

To investigate whether blackcurrant cultivars also vary in their tolerance to HWT, four commonly grown cultivars were selected for this trial. Pedersen *et al.* (2009) also reported incidental improvements in crop vigour following HWT regimes when used for pest eradication. As cutting establishment and vigour is important to ensuring plantations are brought into crop on target this was a further aspect investigated in this project.

The objective of this project was to evaluate different water temperatures and durations of hot water treatment (HWT) to cuttings, for crop safety and potential beneficial effects of establishment and vigour on four cultivars of blackcurrant: Ben Gairn, Ben Starav, Ben Dorain and Ben Tirran.

Materials and methods

Graded, high quality 20cm hardwood cuttings of four cultivars: Ben Gairn, Ben Starav, Ben Dorain and Ben Tirran were supplied from Welsh Fruit Stocks. These were cut in early February and kept in cold store until treatment on 16 April 2012. The cuttings were removed

from the cold store and allowed to reach room temperature before carrying out treatments. Treatments were carried out in controlled temperature water baths according to Table 3. Care was taken to ensure that the baths were not overloaded with material so that target temperatures were regained rapidly after the cuttings were introduced and so hot water would surround all woody material. All cuttings of one cultivar for a test temperature were put into the bath and batches of cuttings were removed as each time period elapsed. Temperature loggers were placed in the water bath to monitor the temperature throughout the treatments. Loose net hammocks were used to allow both heat to penetrate and also rapid removal of the cuttings. Once treated, the cuttings were rapidly cooled and allowed to dry in the Class 2 laminar flow cabinet before planting.

Table 3. Treatments applied to blackcurrant cuttings 16 April 2012 – ADAS Boxworth

Treatment number	**Temperature (^{o} C)**	**Duration (minutes)**
1	Untreated (not soaked)	-
2	Ambient	5
3	Ambient	10
4	Ambient	15
5	Ambient	20
6	40	5
7	40	10
8	40	15
9	40	20
10	45	5
11	45	10
12	45	15
13	45	20
14	50	5
15	50	10
16	50	15
17	50	20

After treatment the cuttings were potted up individually without delay in 3 litre pots filled and firmed to within 2.5 cm of the pot surface with sterilised sandy loam with 20% grit. The cuttings were potted so that the base of the cutting was at least 5 cm from the base of the pot. The top of the cutting was trimmed to leave two buds from the surface of the soil. Cutting material was slightly variable in thickness from 5 -10mm and was therefore graded and blocked according to size (block one thickest cuttings, block four thinnest cuttings).

The trial was replicated four times and each plot consisted of five cuttings (trial plan in Appendix 1). Once growth began the plants were inspected fortnightly for possible pest or

disease problems and direct effects due to water treatment. Full details of pesticide and fertiliser application can be found in Appendix 2

On 5 May 2012 all plants were inspected and growth stage recorded along with the number of dead plants for each plot. A further three assessments were carried out on 18 June, 8 August and 8 October 2012, measuring total stem growth in cm for each plant.

Results and discussion

Establishment and growth stage was assessed on 5 May 2012, three weeks after planting, and was scored on a 1-5 basis according to blackcurrant growth stages 1 = A (dormant bud), 2 = B1 (bud break), 3 = B2 (bud burst), 4 = C1 (1 leaf unfolded) and 5 = C3 (3 leaves unfolded). At this first assessment, establishment was similar across treatments 1 to 12. However for treatments 13-15 (45^{o}C for 20 minutes and all 50^{o}C treatments) there was a significant delay in establishment (Table 4) or an increase in the proportion of dead plants (data not shown). Shoot growth (in cm) was assessed on 18 June, 8 August and 8 October. Averages across all cultivars for each assessment are shown in Table 4 and for each cultivar individually in Tables 5 - 8.

The same result was clear across all assessments and varieties. None of the water treatments (with or without heat) enhanced establishment or growth when compared to an untreated cutting. There were, however, significant effects of hot water treatment, with growth suppression occurring at higher temperatures and longer durations. At 45^{o}C for 15 or 20 minutes there was some growth reduction for cultivars apart from Ben Tirran. At 50^{o}C, there was significant growth suppression at all soak durations, with cutting death occurring at durations of 15 and 20 minutes. Ben Tirran appeared to be the most resilient to the hottest treatment, showing more consistent growth at 50^{o}C for up to 15 minutes than the other three varieties; Ben Gairn showed marginally less heat tolerance.

No phytotoxicity or differences in leaf quality were noted from any of the treatments and no treatment differences in pest or disease levels was observed, however pest and diseases were all at a very low level (pest control was applied). There were no significant block effects, indicating that the cutting thickness did not affect the results.

Table 4. Average growth stage score and shoot growth across four cultivars – ADAS Boxworth 2012.

Treatment	Growth stage score 5 May	Average stem growth in cm 18 June	8 August	8 October
Untreated	2.70	13.27	41.68	44.25
Ambient 5 min	2.75	12.93	37.18	41.60
Ambient 10 min	2.43	12.57	36.25	40.26
Ambient 15 min	2.58	12.36	36.84	41.39
Ambient 20 min	2.58	12.40	39.21	41.95
40°C 5 min	2.75	11.93	38.38	42.03
40 °C 10 min	2.58	12.99	39.70	43.06
40 °C 15 min	2.40	11.63	36.03	40.03
40 °C 20 min	2.46	12.42	36.57	40.31
45 °C 5 min	2.60	12.98	38.60	42.91
45 °C 10 min	2.13	12.24	36.46	40.34
45 °C 15 min	2.16	11.30	37.05	41.08
45 °C 20 min	1.53	10.81	36.57	39.70
50 °C 5 min	1.28	9.10	30.58	35.05
50 °C 10 min	0.74	6.36	22.41	28.46
50 °C 15 min	0.70	1.27	6.54	8.08
50 °C 20 min	0.64	0.00	0.00	0.00
P value	<0.001	<0.001	<0.001	<0.001
LSD	0.574	1.498	5.172	5.16

Growth stage scores 1 =A Dormant bud, 2=B1, 3=B2, 4=C1 and 5=C3

Table 5. Average growth stage score and shoot growth for cv. Ben Gairn – ADAS Boxworth 2012.

Treatment	Growth stage score	Average stem growth in cm Assessment 2	Assessment 3	Assessment 4
Untreated	2.65	14.21	47.50	52.35
Ambient 5 min	2.65	12.17	39.27	48.95
Ambient 10 min	2.70	12.21	39.39	45.30
Ambient 15 min	2.50	12.25	39.97	49.50
Ambient 20 min	2.50	14.04	45.11	51.35
40°C 5 min	2.80	11.17	41.05	50.45
40 °C 10 min	2.40	14.73	45.12	52.20
40 °C 15 min	2.55	12.25	42.77	50.65
40 °C 20 min	2.50	12.06	44.17	48.90
45 °C 5 min	2.50	13.33	43.91	52.35
45 °C 10 min	1.95	13.15	40.05	47.90
45 °C 15 min	2.25	12.58	42.09	48.50
45 °C 20 min	1.30	9.88	39.30	44.00
50 °C 5 min	0.90	5.08	24.00	30.40
50 °C 10 min	0.70	2.58	10.83	19.70
50 °C 15 min	0.65	0.38	2.25	2.30
50 °C 20 min	0.75	0.00	0.00	0.00
p value	<0.001	<0.001	<0.001	<0.001
LSD	0.788	3.005	10.433	8.8750

Growth stage scores 1 =A Dormant bud, 2=B1, 3=B2, 4=C1 and 5=C3

Table 6. Average growth stage score and shoot growth for cv. Ben Starav – ADAS Boxworth 2012.

Treatment	Growth stage score	Average stem growth in cm		
		Assessment 2	Assessment 3	Assessment 4
Untreated	3.50	12.50	38.27	41.15
Ambient 5 min	4.00	12.13	32.17	34.95
Ambient 10 min	3.25	12.50	31.01	37.05
Ambient 15 min	3.75	12.63	33.97	39.65
Ambient 20 min	3.90	12.67	36.69	39.20
40°C 5 min	3.80	12.04	38.55	40.95
40 °C 10 min	3.95	12.88	41.82	43.50
40 °C 15 min	3.50	12.25	33.66	36.75
40 °C 20 min	3.65	13.21	29.61	37.00
45 °C 5 min	3.85	13.04	37.85	42.90
45 °C 10 min	2.74	10.46	30.44	35.50
45 °C 15 min	2.80	9.71	31.25	37.85
45 °C 20 min	2.15	8.79	33.18	36.70
50 °C 5 min	1.60	8.19	30.15	33.15
50 °C 10 min	0.70	4.88	14.63	21.50
50 °C 15 min	0.65	0.38	1.50	1.70
50 °C 20 min	0.60	0.00	0.00	0.00
p value	<0.001	<0.001	<0.001	<0.001
LSD	0.609	1.868	6.785	5.830

Growth stage scores 1 =A Dormant bud, 2=B1, 3=B2, 4=C1 and 5=C3

Table 7. Average growth stage score and shoot growth for cv. Ben Dorain – ADAS Boxworth 2012.

Treatment	Growth stage score	Average stem growth in cm		
		Assessment 2	Assessment 3	Assessment 4
Untreated	3.10	14.23	43.21	45.40
Ambient 5 min	2.85	15.27	43.14	43.75
Ambient 10 min	2.60	14.50	43.38	43.80
Ambient 15 min	2.55	13.27	41.78	43.35
Ambient 20 min	2.65	13.65	40.30	41.80
40°C 5 min	3.00	12.56	41.21	43.25
40 °C 10 min	2.80	13.35	41.70	43.55
40 °C 15 min	2.55	11.94	38.68	40.25
40 °C 20 min	2.55	14.65	41.26	42.30
45 °C 5 min	2.90	13.42	40.08	40.80
45 °C 10 min	2.35	13.67	41.71	42.45
45 °C 15 min	2.10	11.90	38.46	40.65
45 °C 20 min	1.60	13.00	41.33	42.70
50 °C 5 min	1.10	10.77	32.51	40.25
50 °C 10 min	0.70	6.75	24.60	32.15
50 °C 15 min	0.65	0.60	4.75	5.05
50 °C 20 min	0.45	0.00	0.00	0.00
p value	<0.001	<0.001	<0.001	<0.001
LSD	0.573	1.448	4.854	4.702

Table 8. Average growth stage score and shoot growth for cv. Ben Tirran – ADAS Boxworth 2012.

Treatment	Growth stage score	Average stem growth in cm		
		Assessment 2	Assessment 3	Assessment 4
Untreated	1.55	12.15	37.75	38.10
Ambient 5 min	1.50	12.15	34.14	38.75
Ambient 10 min	1.15	11.08	31.22	34.90
Ambient 15 min	1.50	11.31	31.65	33.05
Ambient 20 min	1.25	9.23	34.74	35.45
40°C 5 min	1.40	11.96	32.73	33.45
40 °C 10 min	1.15	11.02	30.18	33.00
40 °C 15 min	1.00	10.06	29.02	32.45
40 °C 20 min	1.15	9.77	31.24	33.05
45 °C 5 min	1.15	12.15	32.59	35.60
45 °C 10 min	1.50	11.69	33.62	35.50
45 °C 15 min	1.50	11.00	36.41	37.30
45 °C 20 min	1.05	11.58	32.49	35.40
50 °C 5 min	1.50	12.38	35.66	36.40
50 °C 10 min	0.85	11.23	39.59	40.50
50 °C 15 min	0.85	3.73	17.64	23.25
50 °C 20 min	0.75	0.00	0.00	0.00
p value	<0.001	<0.001	<0.001	<0.001
LSD	0.377	2.520	8.386	7.193

Growth stage scores 1 =A Dormant bud, 2=B1, 3=B2, 4=C1 and 5=C3

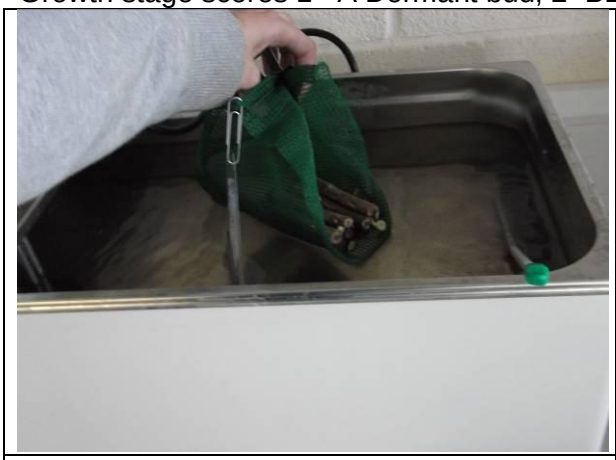

Figure 3. Water bath and cuttings

Figure 4. Cuttings at planting

Figure 5. Three weeks after planting, Treatment 17, 50°C for 20 minutes

Figure 6. Three weeks after planting, Treatment 1, Untreated control

Conclusions

- No improvement to blackcurrant cutting establishment was demonstrated with water soaking with or without heat treatment (up to 50^{o}C for up to 20 minutes).
- Heat treatment of up to 45^{o}C for 20 minutes has been shown to be safe for four the blackcurrant cultivars Ben Gairn, Ben Starav, Ben Dorain and Ben Tirran but caused an initial check in all but Ben Tirran.
- Ben Tirran was moderately tolerant of HWT at 50^{o}C for 15 minutes
- The plants will need to be grown on to determine whether there are any lasting effects in terms of yield potential as a result of the HWT.
- Further research is required to establish if these hot water soak conditions will eradicate important pathogens and to make the process practical at on a larger scale.

Knowledge and Technology Transfer

No knowledge transfer activities were carried out during this project.

References

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Tuovinen, T., Parikka, P. and Lemmetty, A. 2008. Plant protection in currant production in Finland. Acta Horticulturae 777: 333-338

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Appendices

Appendix 1. Trial plan

Ben Gairn

Plot	Block	Treatment	Plot	Block	Treatment	Plot	Block	Treatment	Plot	Block	Treatment
1	1	3	**18**	2	4	**35**	3	15	**52**	4	7
2	1	2	**19**	2	6	**36**	3	13	**53**	4	5
3	1	8	**20**	2	8	**37**	3	6	**54**	4	13
4	1	14	**21**	2	5	**38**	3	17	**55**	4	14
5	1	4	**22**	2	12	**39**	3	9	**56**	4	8
6	1	7	**23**	2	16	**40**	3	11	**57**	4	2
7	1	1	**24**	2	10	**41**	3	12	**58**	4	10
8	1	10	**25**	2	14	**42**	3	4	**59**	4	3
9	1	13	**26**	2	9	**43**	3	8	**60**	4	6
10	1	16	**27**	2	2	**44**	3	3	**61**	4	12
11	1	5	**28**	2	3	**45**	3	10	**62**	4	4
12	1	11	**29**	2	15	**46**	3	2	**63**	4	16
13	1	9	**30**	2	17	**47**	3	14	**64**	4	11
14	1	6	**31**	2	1	**48**	3	5	**65**	4	15
15	1	12	**32**	2	13	**49**	3	7	**66**	4	1
16	1	17	**33**	2	7	**50**	3	1	**67**	4	9
17	1	15	**34**	2	11	**51**	3	16	**68**	4	17

Ben Starav

Plot	Block	Treatment	Plot	Block	Treatment	Plot	Block	Treatment	Plot	Block	Treatment
1	1	3	**18**	2	2	**35**	3	15	**52**	4	14
2	1	6	**19**	2	15	**36**	3	6	**53**	4	2
3	1	16	**20**	2	1	**37**	3	13	**54**	4	6
4	1	7	**21**	2	13	**38**	3	8	**55**	4	9
5	1	15	**22**	2	9	**39**	3	9	**56**	4	3
6	1	1	**23**	2	10	**40**	3	4	**57**	4	11
7	1	10	**24**	2	7	**41**	3	16	**58**	4	13
8	1	9	**25**	2	5	**42**	3	7	**59**	4	12
9	1	14	**26**	2	11	**43**	3	12	**60**	4	10
10	1	12	**27**	2	6	**44**	3	3	**61**	4	7
11	1	4	**28**	2	4	**45**	3	10	**62**	4	16
12	1	13	**29**	2	16	**46**	3	14	**63**	4	5
13	1	17	**30**	2	14	**47**	3	2	**64**	4	8
14	1	5	**31**	2	17	**48**	3	17	**65**	4	1
15	1	11	**32**	2	12	**49**	3	1	**66**	4	15
16	1	8	**33**	2	3	**50**	3	11	**67**	4	17
17	1	2	**34**	2	8	**51**	3	5	**68**	4	4

Ben Dorain

Plot	Block	Treatment	Plot	Block	Treatment	Plot	Block	Treatment	Plot	Block	Treatment
1	1	17	**18**	2	9	**35**	3	17	**52**	4	9
2	1	14	**19**	2	7	**36**	3	16	**53**	4	14
3	1	11	**20**	2	15	**37**	3	6	**54**	4	15
4	1	9	**21**	2	6	**38**	3	7	**55**	4	2
5	1	2	**22**	2	1	**39**	3	9	**56**	4	4

6	1	5	**23**	2	12	**40**	3	10	**57**	4	1
7	1	12	**24**	2	11	**41**	3	3	**58**	4	12
8	1	7	**25**	2	13	**42**	3	2	**59**	4	7
9	1	16	**26**	2	3	**43**	3	15	**60**	4	13
10	1	13	**27**	2	4	**44**	3	13	**61**	4	3
11	1	3	**28**	2	2	**45**	3	5	**62**	4	5
12	1	6	**29**	2	16	**46**	3	1	**63**	4	16
13	1	4	**30**	2	17	**47**	3	12	**64**	4	11
14	1	8	**31**	2	10	**48**	3	14	**65**	4	17
15	1	10	**32**	2	8	**49**	3	11	**66**	4	10
16	1	1	**33**	2	14	**50**	3	4	**67**	4	8
17	1	15	**34**	2	5	**51**	3	8	**68**	4	6

Ben Tirran

Plot	Block	Treatment	**Plot**	Block	Treatment	**Plot**	Block	Treatment	**Plot**	Block	Treatment
1	1	10	**18**	2	8	**35**	3	4	**52**	4	12
2	1	4	**19**	2	3	**36**	3	1	**53**	4	7
3	1	11	**20**	2	10	**37**	3	10	**54**	4	15
4	1	3	**21**	2	6	**38**	3	6	**55**	4	11
5	1	2	**22**	2	1	**39**	3	7	**56**	4	16
6	1	6	**23**	2	4	**40**	3	16	**57**	4	5
7	1	14	**24**	2	15	**41**	3	12	**58**	4	14
8	1	5	**25**	2	7	**42**	3	17	**59**	4	13
9	1	17	**26**	2	16	**43**	3	15	**60**	4	9
10	1	16	**27**	2	13	**44**	3	8	**61**	4	10
11	1	15	**28**	2	14	**45**	3	5	**62**	4	8
12	1	1	**29**	2	17	**46**	3	9	**63**	4	3
13	1	9	**30**	2	11	**47**	3	13	**64**	4	4
14	1	13	**31**	2	2	**48**	3	14	**65**	4	2
15	1	12	**32**	2	9	**49**	3	11	**66**	4	6
16	1	8	**33**	2	5	**50**	3	2	**67**	4	17
17	1	7	**34**	2	12	**51**	3	3	**68**	4	1

Appendix 2. Crop husbandry

Date	**Treatment**	**Rate**
19/6/12	Calypso	0.25 L/ha
19/6/12	Draza	3.75 kg.ha
28/6/12	Floranid permanent	2 g/pot