Project title:	Narcissus: Investigation into the effects of a range of potential biocides in hot water treatment
Project number:	BOF 077
Project leader:	Rob Lillywhite, University of Warwick
Report:	Second annual report, December 2017
Previous report:	First annual report, December 2016
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Date project commenced:	1st January 2016
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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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CONTENTS

GROWER SUMMARY

Headline

At the end of the second year of the project, growers should be aware of the following:

- Under laboratory conditions, thermal, ultra-violet radiation and chlorine dioxide treatments provided effective control of Fusarium spores
- Filtration and UV radiation equipment has been successfully retrofitted to a commercial HWT system
- Filtration proved difficult to implement under commercial conditions because most bioload was composed of very fine soil particles
- Field trials were established in Cornwall but as yet any benefit of treatments has yet to be determined

Background

Hot water treatment (HWT) of narcissus bulbs is used to control pests and diseases, notably stem nematodes, bulb scale mites and Fusarium basal rot. This has been the standard approach for at least 70 years. For most of that time, formalin was added to HWT tanks as a general biocide i.e. to reduce inoculum in the tank water, however approval for formalin was withdrawn in 2008. Work in BOF 061a (Lole, 2010) identified FAM 30 as a viable alternative and this has since become standard practice in the UK. However, FAM 30 is expensive in comparison to formalin and the result has been that growers do not always use it at the required rate and this issue is exacerbated since FAM 30 rapidly depletes in tanks under a high bioload.

Other biocide alternatives have been considered, notably chlorine dioxide which was demonstrated to be effective against spread of Fusarium (Chastagner and Riley, 2002) and is believed to be currently used by American Narcissus growers. However, in AHDB Horticulture project BOF 061a (Lole, 2010), chlorine dioxide was assessed alongside a number of alternative biocides, but was not considered further as FAM 30 was found to be more effective. The use of chlorine dioxide was further reviewed in BOF 070 (Hanks, 2010) which suggested that additional investigations were required before it could be recommended to growers.

Other biocides previously examined include peroxyacetic acid (Hanks and Linfield, 1999), hydrogen peroxide and UV (Stewart-Wade, 2011) but tank bioload was again found to reduce their efficacy so further commercial scale evaluation is required before they can be recommended. Non-chemical biocidal approaches, e.g. UV and thermal treatment, have been used in other water-based treatment systems and appear to offer a viable alternative to chemical approaches but their efficacy is known to very dependent of water clarity, which is a problem with high bioload HWT (Petit, 2016). The issue of high HWT tank bioload was reported in BOF 070 (Hanks, 2010) and generating a solution to this issue is probably key in improving the efficacy of all biocides and biocidal approaches (and probably fungicides as well).

Project aim

The aim of this project is to examine a range of candidate biocides (chlorine dioxide, hydrogen peroxide and didecyl dimethyl ammonium chloride) and physical approaches (thermal and UV treatment) for their efficacy and ease of use against stem nematode and Fusarium basal rot.

The project has been divided into eight objectives:

1. Review of the literature
2. *In vitro* laboratory tests
3. Assess the feasibility and cost of retrofitting biocide delivery systems to existing HWT tanks
4. Assess impact of different treatments on infrastructure
5. Small-scale tank tests
6. Commercial scale testing
7. Field trials
8. Health and safety considerations

Summary

This report covers the period January 2017 to December 2017 which is the second year of the four year project to investigate new or improved biocidal approaches in the hot water treatment of daffodil bulbs. This period saw the cessation of laboratory testing and the start of commercial on-farm testing. A literature review, details of the laboratory testing, including a feasibility study on microwave sterilization, and a feasibility and cost/benefit analysis of HWT modifications can be found in the 1st annual report.

Results from year 1 had shown that chloride dioxide was an effective biocide but also that tank bioload had a negative effect of its efficacy. Work in 2017 therefore concentrated on

reducing tank bioload and examining if biocides were more effective under cleaner water conditions. The use of UV radiation was also investigated.

Filtration

The earlier laboratory work carried out during this project had established that for all chemical based fungicide or biocide treatments there was a negative effect of dirty water on the efficacy of the treatment. It is also well established that for UV sterilization to be effective the water needs to be of good clarity. This data, along with some of the feasibility work carried out, was presented to the growers at the AHDB growers meetings of spring 2017. There was a general agreement from the growers that cleaner tank water would be beneficial and a good level of interest in trialling some sort of filtration. Therefore work continued to establish what technology would be best suited to the continual clean-up of tank water.

This investigation into filtration had two aims: firstly a general clean-up of tank water to allow chemical treatments to work more effectively. It was assumed that filtration to a level of approximately 50-100µm would be effective to achieve this aim. Secondly to clean tank water to allow UV sterilization to be effective, which would require filtration to approximately the 5-10µm level. Given the high flow rate of some HWT systems, these parameters caused some problems in identifying suitable filtration equipment that the research team considered would be acceptable to growers and cost-effective. A full description is provided in the Science Section but in summary, it was decided to examine a set-up that comprised two different sized filters in series (150 and 25 micron) and to run two of these in parallel to cope with the flow rate. This system was installed at Carwin Farm in July 2017 in preparation for bulb dipping (Figure 1).

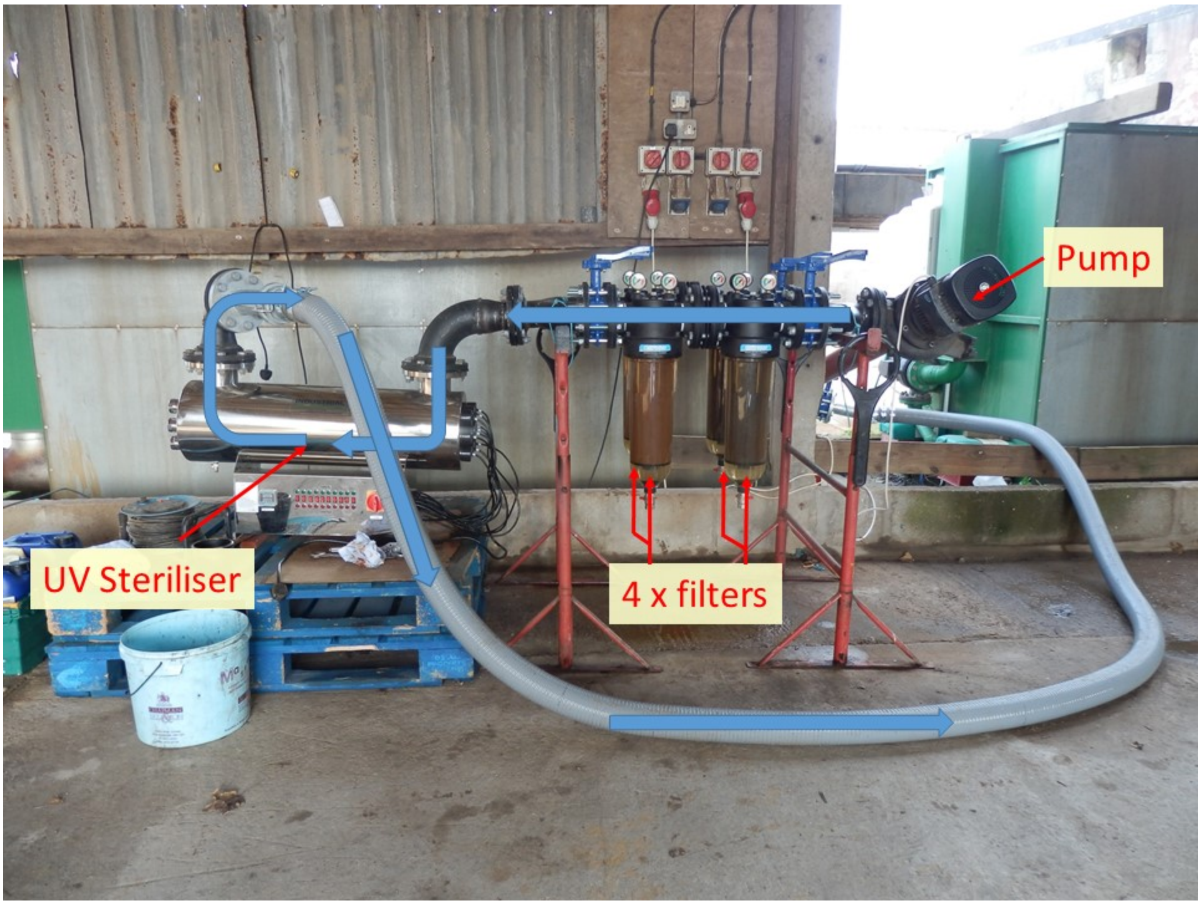

Figure 1. Retro-fitted filtration and UV system at Carwin Farm, Cornwall

The cost of the purchase and installation of the filtration system was £3,057 + VAT which was made up of four filters plus replacement screens (£1505), plumbing and fittings (£1072) and labour (£480).

The system was initially tested using fresh water without bulbs to ensure that it functioned properly. This initiated a considerable release of rust, which quickly clogged the filter screens and necessitated their removal and cleaning a number of times. It was noted that this rust deposit is normally removed by the first batch of bulbs.

The introduction of bulbs introduced organic and mineral contaminants into the system and the filters required cleaning multiple times in order to keep the water flowing through the system. The conclusion, at this early stage, was that unless there was a very clear benefit to filtration, a system that required stopping and the manual cleaning of filters during every run was not going to be commercially viable. In order to explore the effect of filter size on water flow, all the filters were changed to 150 µm which resulted in a marked improvement in water flow.

Whilst this filtration setup did remove some of the contaminants from the tank it was clearly not an appropriate set up to recommend without modification and further testing. With filter screens of pore size smaller than 100 µm (50, 25 and 10 µm) the flow of water was slowed to the point of not moving through the bulb tanks effectively and this problem became worse as the filters became clogged. Observations both on farm and in the lab suggest that filtering to 100 µm will only have a limited positive effect and will not be sufficient to clean the water adequately (to allow the use of UV sterilization although it may improve the efficacy of chlorine dioxide). To filter the water to a fine enough level it is likely that a different approach is required. One consideration was to install more powerful pumps, however, it is unclear at this stage whether greater pressure would improve the flow of water through the tanks and filters or stress the whole system somewhere else. Alternatively, a more sophisticated and expensive filtration system would be required that include automated removal of the collected contaminants and debris. This would be the preferred approach since it could be retrofitted without major modifications to the existing HWT system.

Further investigation on the suspended particle sizes present in tank water, revealed that a majority of them measured between 0.4 and 20µm which is why the 150 and 25 µm filters had proved ineffective. At this size range it is likely that the particles are fine silt and/or clay particles released from any soil adhering to the bulbs rather than fragments of bulb scale; this was contrary to our assumption that bulb scale would make up a significant proportion of the suspended solids. While it is possible to obtain filtration systems to remove these particles, it would be a considerable investment and preventing more of the soil and scale from entering the tanks is likely be a more promising option.

At this stage of the testing, it is hard to draw any conclusions from the work on which recommendations to change of practice could be made. While it demonstrated that it is possible to retrofit filters (and a UV source) to an existing HWT system, the results of filtration were disappointing. It is planned to repeat these tests in Lincolnshire in 2018 to investigate if these results are particular to Cornwall or shared across different production areas. If similar results are observed, it is likely that a simple and inexpensive filtration system is unlikely to be the solution to dirty water in HWT systems and that if filtration is considered necessary, then more expensive and sophisticated systems will be required.

Ultraviolet radiation

Ultraviolet (UV) radiation is a proven sterilization technology that is often used to control pathogens in irrigation water. It is a non-chemical approach that is basically ‘fit and forget’ although its efficacy is known to be much severely reduced in dirty water conditions. A twofold approach was followed which used laboratory testing to assess its efficacy in clean water conditions, and this was followed by on-farm testing to judge whether it was viable under commercial conditions.

In laboratory tests, UV reduced the amount of Fusarium chlamydospores in water samples by 99% after two hours and to no detectable amounts after five hours. However, because UV operates on a continuous basis, we assume that there were still some viable spores circulating in the water early in the test before complete control was exerted. Following treatment, bulbs were air dried overnight before being incubated for 28 days at 25°C. After incubation the bulbs were dissected and scored from 0-10 for basal rot.

Although there was a significantly lower level of infection in the bulbs from the UV treated tank than in the non-UV treated tank the reduction was nowhere near as big as that seen in the water samples. This would suggest that even the very low level of fusarium that was still present was sufficient to cause a substantial level of infection. Even though the level at which the tanks were inoculated was massively higher than would be expected in a commercial tank, the results were encouraging and show that the concept of using UV to control Fusarium was proven.

A commercial scale UV unit was trialled at Carwin Farm in August 2017. The cost of the purchase and installation was £2,450 + VAT. The combination of the filters and UV source was tested with one batch of bulbs and while it had some positive effects it was clearly not an appropriate set up to recommend without modification. With filter membranes of pore size smaller than 100 µm (50, 25 and 10 µm) the flow of water was slowed to the point of not moving through the bulb tanks effectively, this problem became worse as the filters became clogged. Because the UV technology relies on water flowing through the UV treatment tube, any reduction in water flow will reduce the control of water-borne pathogens. This is a double burden because clear water is required for effective UV treatment. Observations both on farm and in the lab suggest that filtering to 100 µm will only have a limited effect and will not be sufficient to clean the water adequately to allow UV treatment to be effective.

The key to improving water clarity and treatment efficacy is to reduce the amount of bioload entering HWT tanks. Even though bulbs are cleaned and graded prior to dipping, enough soil still adheres to them to produce sufficient suspended particles to dirty tank water. During 2018, it is hoped to examine if additional cleaning of bulbs prior to HWT can reduce tank bioload and improve the efficiency of filtration and therefore the efficacy of UV treatment.

Chlorine dioxide

Commercial testing of chlorine dioxide was undertaken at Carwin Farm in August 2018. Chlorine dioxide (in the form of Activ-ox, supplied by Feedwater) was added to the tank with in-tank levels quantified using a Palintest meter. Despite increasing levels of chemicals added (until the supply was exhausted) the in-tank level of chlorine dioxide did not at any point reach a detectable level on the measuring equipment. Consequently, it was decided not to run a load of bulbs as it was unlikely to give a meaningful result and the bulbs were unlikely to be protected from Fusarium. This result confirms the difficulties experienced by other researchers and has triggered a rethink of the approach. It is planned to test chloride dioxide again in 2018 but to use an automated dosing system supplied by the Scotmas Group to try and overcome some of the operational difficulties. However, despite these problems, chloride dioxide remains the most promising of the chemical biocides as it is widely used in other industries and its use is unlikely to be regulated in the foreseeable future.

Thermal treatment

Work in year one of the project demonstrated that thermal treatment was a very effective biocidal approach with complete control of Fusarium spores being achieved at temperatures above 60°C. The concept that supports this approach was that tank water could be either continuously or batch treated to provide another form of control. In practice, this would mean tank water being sterilised overnight, when empty of bulbs, by increasing the temperature to 60°C thereby ensuring that the water was sterile at the start of every day. This was tested at both Carwin and Bosahan Farms in Cornwall and was successfully achieved without any issues. The effectiveness of this approach is currently unknown although the treated bulbs will be monitored over the next two years to see if they show any advantage.

During the Narcissus Growers Workshops in May 2017 there was also interest in using thermal treatments on bulbs themselves. Although bulbs are normally dipped at 44°C to avoid tissue damage, while at the same time controlling stem nematodes and bulb scale mites, the effect of short-term dipping at 60°C or more was unknown.

Trials were undertaken at Carwin Farm and Warwick Crop Centre to investigate the effect of water temperature and dipping time on daffodil bulbs. Bulbs were dipped at either 60°C, 65°C or 70°C for either 3, 5 or 10 minutes. Bulb core temperature increased in line with water temperature and dipping time. However, it was noted that the data obtained at Carwin Farm shows a much higher core temperature than the equivalent data obtained at Warwick Crop Centre. This effect was more noticeable at the shorter dipping times. It may be that this is due to the addition of bulbs to the small tanks at Warwick Crop Centre causing a drop in water temperature, which is likely to be less of an issue in commercial tanks.

Post-dipping, half of the bulbs were incubated at 25°C for 30 days while the other half were planted into individual pots and will be assessed in spring 2018.

Financial Benefits

At this stage of the project, it is not possible to make any assessment of the financial benefits arising from the results of the research. Although the costs of the different approaches are known, we cannot yet quantify any financial savings that might arise from the biocidal approaches.

Action Points

Growers should ensure that bulbs destined for hot water treatment are as clean as possible as that will reduce the build-up of tank bioload. Reduced bioload will increase the efficacy of both fungicides and chemical biocides.

Growers should be cautious of using manual chlorine dioxide treatments (premixed liquids) as repeated trials have shown that target concentrations cannot be achieved by single dosing.

SCIENCE SECTION

Introduction

Hot water treatment (HWT) of narcissus bulbs is used to control pests and diseases, notably stem nematodes, bulb scale mites and Fusarium basal rot. This has been the standard approach for at least 70 years. For most of that time, formalin was added to HWT tanks as a general biocide, however, approval for formalin was withdrawn in 2008. Work in BOF 61 (Lole, 2010) identified FAM 30 as a viable alternative and this has since become standard practice in the UK. However, FAM 30 is expensive in comparison to formalin and the result has been that growers do not always use it at the required rate and this issue is exacerbated since FAM 30 rapidly depletes in tanks under a high bioload.

Other biocide alternatives have been considered, notably chlorine dioxide which was demonstrated to be effective against spread of *Fusarium* (Chastagner and Riley, 2002) and is apparently used by American narcissus growers. However, in AHDB Horticulture projects BOF 61 and 61a, chlorine dioxide was assessed alongside a number of alternative biocides, but was not considered further as FAM 30 was more effective. The use of chlorine dioxide was further reviewed in BOF 70 which recommended that further investigations were required before it could be recommended to growers.

Other biocides examined previously include peroxyacetic acid (Hanks and Linfield, 1999), hydrogen peroxide and UV (Stewart-Wade, 2011) but require further commercial scale testing before they can be recommended. Non-chemical biocidal approaches, e.g. UV and thermal treatment, have been used in other water-based treatment systems and appear to offer a viable alternative to chemical approaches but their efficacy is known to very dependent on water clarity, which is a problem with high bioload HWT. The issue of high HWT tank bioload was reported in BOF 70 and generating a solution to this issue is probably key in improving the efficacy of all biocides and biocidal approaches (and probably fungicides as well).

The aim of this project is to examine a range of candidate biocides (chlorine dioxide, hydrogen peroxide and didecyl dimethyl ammonium chloride) and physical approaches (thermal and UV treatment) for their efficacy and ease of use against stem nematode and Fusarium basal rot.

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Objectives and results

The study of biocides in HWT is a very specialised area of research and the majority of evidence is to be found in AHDB commissioned research, namely BOF 61a (Hole, 2010) and BOF 70 (Hanks, 2010) and BOF 70a (Hanks, 2012) and the HDC Narcissus Manual (Hanks, 2013). No new scientific articles on this topic have been published since Chastagner and Riley in 2002. Therefore there is no new direct evidence which this project can build on. However, the literature on biocides is slightly more forthcoming as they are used as disinfectants in a number of industries. Perhaps the most useful of these is the treatment of irrigation water, particularly in recirculating systems, where the control of pathogens is critical. Stewart-Wade (2011) reviewed a number of biocides and technological innovations and discussed the advantages and disadvantages of a number of approaches and her work is used to inform this project.

The primary objective in year two was to trial some of the candidate biocidal treatments under commercial conditions. The treatments chosen were filtration, as it benefits both chemical biocides and fungicides, UV and chloride dioxide.

Small scale tank tests - materials and methods

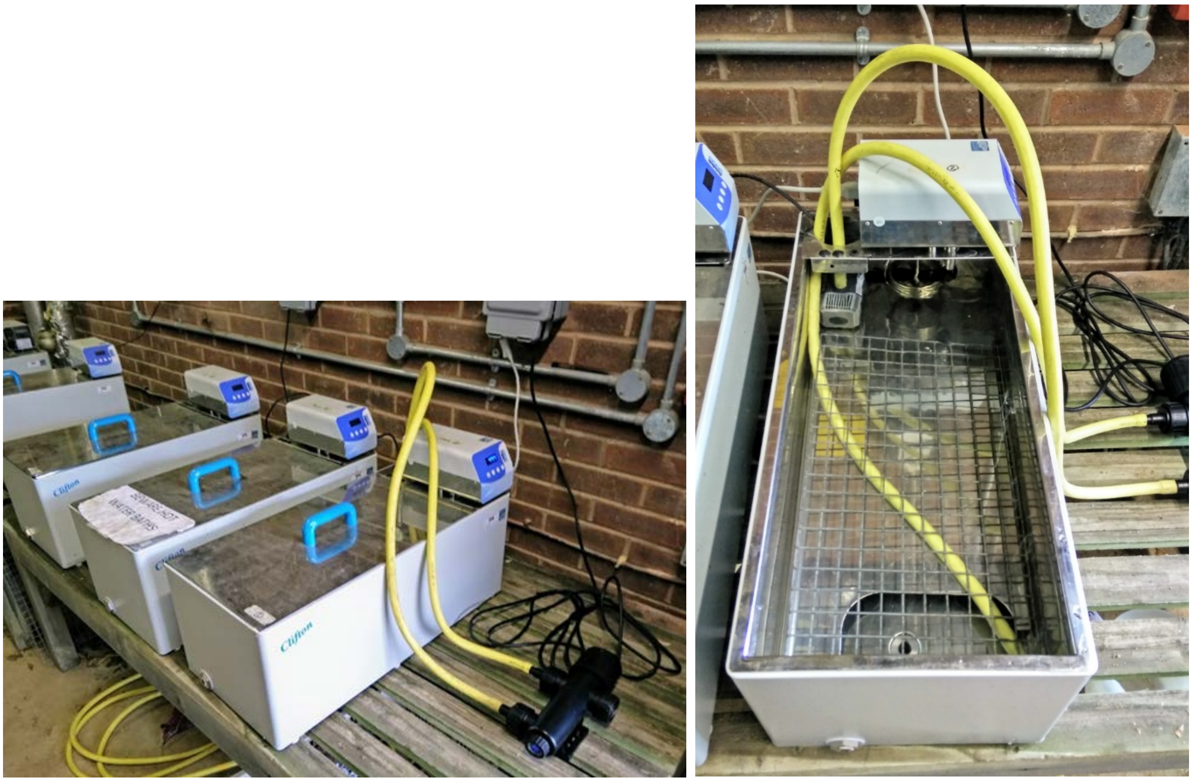
Small scale tank tests for the majority of the methods to be investigated were reported in full in the Year 1 (2016) report. The work to investigate the efficacy of UV sterilization was still outstanding at that time and is reported here.

Ultra-violet sterilization.

Production of Fusarium chlamydospores. Chlamydospores of *Fusarium oxysporum f.sp. narcissus* isolate 139 were prepared by using the method described in full in the 2016 report for this project. Prior to use chlamydospore concentration was established by counting on a

Neubauer improved cell counting chamber. All samples were pooled before being aliquoted into appropriate portions for the tank tests.

<u>Test protocol.</u> Three 35l water baths were used in for testing, one of these (tank 3) had an additional pump added to supply a steady flow of tank water through the UV steriliser. The experimental setup is shown in Figures 2 and 3.



Figures 2 and 3. The three water baths used for small-scale testing. The water bath in the foreground in Figure 1 shows the UV unit with yellow tubing while Figure 2 shows the inlet and outlet setup.

All three tanks were filled with clean fresh water and heated to 44.4°C and the additional pump on tank 3 was also turned on. Once the required temperature was reached, chlamydospores of *Fusarium oxysporum f.sp. narcissus* isolate 139 were added to tanks 2 and 3 at a rate of 6500 chlamydospores per ml.

After five minutes to allow for thorough mixing of the spores, 50ml samples were taken from each tank. The UV unit was then turned on. After 2hr at 44.4°C and with water being circulated through the UV unit in the case of tank 3, a further sample was taken from each tank. An onion bag containing 40 bulbs was then added to each tank. After a further 3hrs 15 minutes the bulbs were removed and a final water sample was taken from each tank.

The bulbs were then air dried overnight before being incubated for 28 days at 25°C. After incubation the bulbs were dissected and scored from 0-10 for basal rot.

Serial dilutions of the water samples were then performed and each dilution (100 µl) plated onto three replicate plates. Plates were incubated at 18°C (+/- 2) for five days, the number of colony forming units (cfu) recorded and the concentration of viable spores/ml calculated.

Results

Culture from water samples. The control tank (tank 1) to which no chlamydospores had been added showed zero Fusarium colonies on all plates until the final time point. Two colonies were counted on one of the plates containing undiluted samples, suggesting that a small number of spores may have been released from the bulbs during the 3.25hr dipping time. Tanks 2 & 3 both showed high levels of fusarium at time point 1 (immediately following addition of chlamydospores). The levels in tank 2 (no UV) dropped significantly over the course of the 5.25 hour experiment, which may be due to adhesion of spores to either the bulbs or the experimental equipment as well as loss of viability of some of the less robust material. By time point 3 (3.25 hours at 44.4°C) however the detectable level of fusarium remained at 74% of the inoculated level. In tank 3 (with UV treatment) by time point 2 (2 hours at 44.4°C) the detectable level of fusarium had dropped by >99.9% and by time point 3 there was no detectable fusarium (Figure 4).

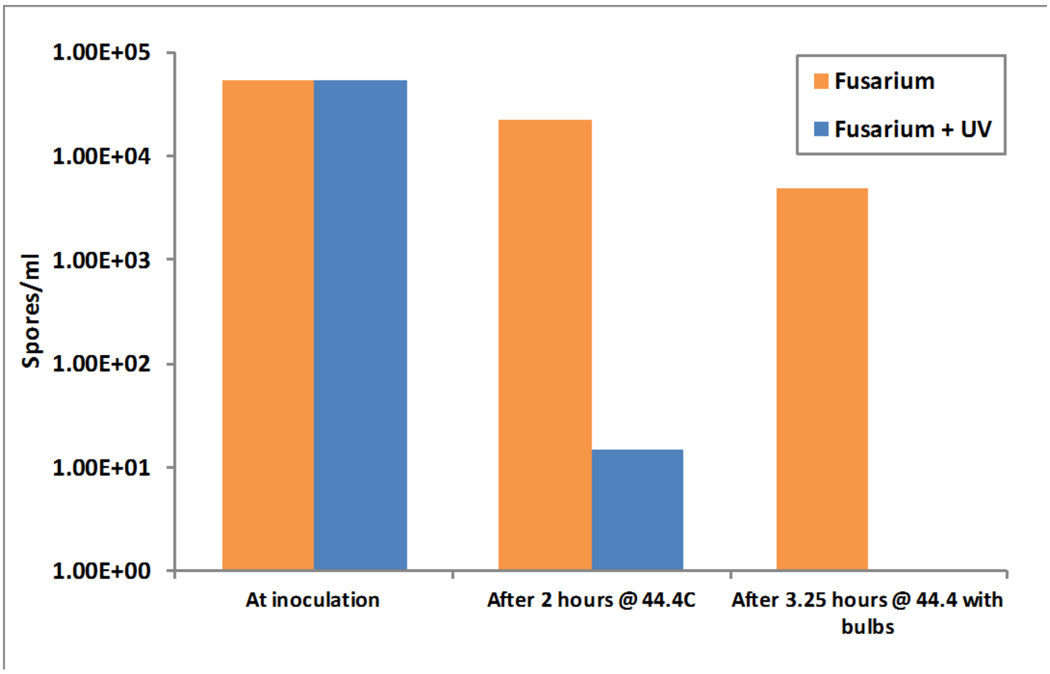


Figure 4. Spore counts following control and UV treatments in small-scale tank tests.

Bulb dissections. The control bulbs from tank 1 showed 12.5% infection, with 35 of 40 bulbs being scored zero on the basal rot scale. The bulbs from tank 2 showed 100% infection with most (82.5%) being categorized as highly infected (6-10 on the scale). Bulbs from tank 3 showed 75% infection with 32.5% being categorized as medium (scored between 3 and 5) and 42.5% being categorized as highly infected (Figure 5).

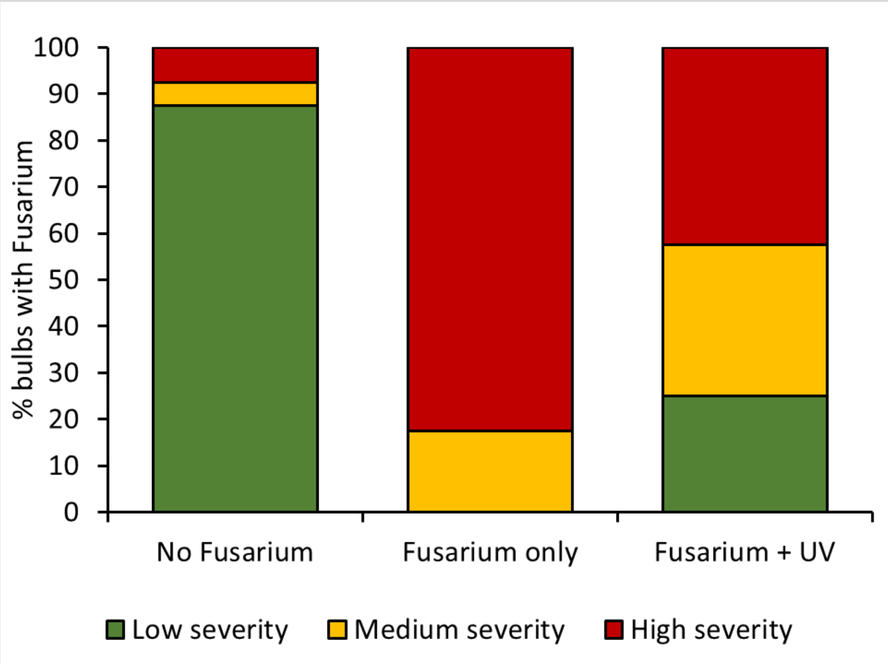


Figure 5. The level and severity of infection following incubation for 28 days at 25°C.

Discussion.

The effectiveness of UV treatment as measured by culturing of water samples was extremely promising with a reduction in measurable level of 99.9% before the addition of the bulbs and zero fusarium detectable by the end of the experiment. Although there was a significantly lower level of infection in the bulbs from the UV treated tank than in the non-UV treated tank the reduction was nowhere near as dramatic as that seen in the water samples. This would suggest that even the very low level of fusarium that was still present when the bulbs were added to tank 3 was sufficient to cause a substantial level of infection. The possibility of latent infection was considered by the use of a control treatment. The level at which the tanks were inoculated was far higher than would be expected in a commercial tank and so the level of control achievable may be more impressive than shown in these tests.

This test demonstrated that UV is a very effective biocide but that there is a delay between the bulbs entering the water and full control being implemented. As a proof of concept, the results are compelling but further testing at spore levels closer to those found commercially is required before any further conclusions can be drawn.

Water turbidity and effect on UV treatment

The efficacy of UV is known to reduce dramatically in water with high levels of suspended soils (Figure 6). Since typical HWT water is dirty, this is potentially a barrier to the use of UV (Figure 7). When turbidity is 5 NTU* or greater and/or total suspended solids are greater than 10 ppm, pre-filtration of the water is highly recommended. Normally, it is advisable to install a 5 to 20 µm filter prior to a UV disinfection system[1]

* Nephelometric Turbidity Unit (NTU) is a measure of turbidity where scattered light is measured at 90 degrees from the incident light beam.

Figure 6. Examples of 5, 50 and 500 NTU

[1] Water Research Centre. www.water-research.net.

Figure 7. Examples of tank water: fresh, after 1 dip, after 2 dips and end of season (left to right)

In order to help understand the effect of tank bioload on any potential UV treatments, spectrophotometric analysis was carried out. End-of-season water (sample in Figure 7) proved very effective at blocking the transmission of light, particularly at shorter wavelengths (Table 1).

Table 1. The absorbance (at 350nm) of end of season tank water at varying concentration

Concentration (%)	Absorbance at 350nm	Transmission (%)
10	1.709	2
5	0.983	10.4
2.5	0.553	27.7
1.25	0.353	44
0.625	0.237	57.6
0	0.	100

To try and replicate used tank water, daffodil bulb scale was finely ground and added to water at varying rates (Table 2).

Table 2. The absorbance (at 350nm) of end of manufactured dirty water

Scale (g/l)	Absorbance at 350nm	Transmission (%)
15	1.863	1.4
10	1.12	7.6
7.5	0.837	14.5
5	0.537	29.2
2.5	0.218	60.4
0	0	100

Whilst this allowed a calculation of the amount of ground scale required to absorb the same light as end-of-season water (i.e.14.6g/l is equivalent to a 10% tank water solution) it was noted that visually these two solutions appeared quite different. This would suggest that scale particles may not be the main constituent of the suspended solids in the tank water.

Filtration

Background

The earlier laboratory work carried out during this project established that for all chemical based fungicide or biocide treatments there was a negative effect of dirty water on the efficacy of the treatment. It is also well established that for UV sterilization to be effective the water needs to be of good clarity. This data, along with some of the feasibility work carried out, was presented to the growers at the AHDB growers meetings of spring 2017. There was a general agreement from the growers that cleaner tank water would be beneficial and a good level of interest in trialling some sort of filtration. Therefore work continued to establish what technology would be best suited to the continual clean-up of tank water.

This investigation into filtration had two aims: firstly general clean-up of tank water allowing chemical treatments to work more effectively. It was assumed that filtration to a level of approximately 50-100µm would be effective to achieve this aim. Secondly in order to clean up the tank water to allow UV sterilization to be effective, which would require filtration to approximately the 5-10µm level.

Initial Considerations were established through discussion with growers, these included:

- fairly high flow rate, assumed 100m^3/hour as general target
- Filtration should not significantly impede flow
- Minimal user input required, too much cleaning/maintenance would be a barrier to implementation

- Cost – different attitude to cost expressed between growers but the expectation was that any investment would need to be justified by expected benefits
- How would filtration affect chemical treatments?

Once discussions began with manufacturers a second set of considerations were established:

- solids loading, size profile and specific gravity (relevant to vortex/kinetic systems) of filtrate
- pressure of system/ required pressure for filtration
- multi stage(/method) filtration

Filter types:

Vortex/ kinetic

Vortex or kinetic separators are designed to force the fluid down in a vortex and then back up through the centre of the vortex (Figure 8). Sediments drop out of the bottom of the vortex rather than being pushed back up and can be periodically removed as a waste sludge. The advantage of this type of system is that the dirty water does not pass through a filter and therefore there is no drop in flow rate due to clogging of the filter material.

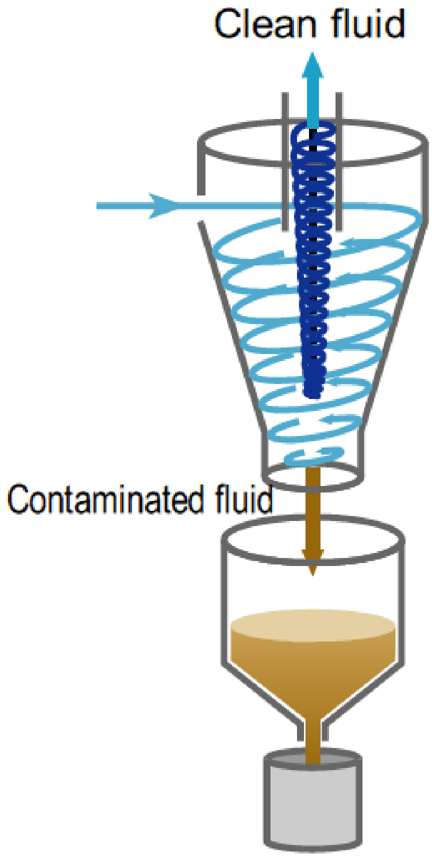


Figure 8. Representation of a vortex type filter

There are various designs along this model (Nikuni, Amiad, Toshiba, Socotec, Bell & Gosset), however they require a steady working pressure of between 36psi (Nikuni) and 125psi (Bell and Gossett). The efficiency of sediment removal in this type of system is dependent largely on the specific gravity (SG) of the particulates. In the Nikuni system a SG of >2.5 is recommended for single pass removal of particles with removal efficiency between 65% (3µ² and 99% (25µm). The SG of tank water particulates has not been ascertained but sand and grit are likely to be the highest SG components and these have SGs considerably lower than 2.5 (email communication from Peter Pridham, Aeration & Mixing Ltd). This issue is somewhat mitigated by the fact that we would be working with an enclosed system where the water is cycled through the system approximately 10 times per hour.

Conventional Filtration – metal, polymer, micro fibre - Russell Finex, Orival Eaton, Forsta, Tekleen, Hydro-gen, Rotorflush, Secker Welding, Eliquo Hydrok

- Automatic/semi-automatic cleaning, requiring minimal volume loss with discharge

Conventional filtration, where the fluid passes through a porous material comes in many configurations, the most common materials are a metal or polycarbonate sheet with pores of appropriate size although some devices use fibrous sheets. The advantage of this type of system is that the filter represents a physical barrier to particles larger than the pore size (as compared to the vortex separators), the disadvantage is that there is a build-up of dirt on the filter that can lead to clogging and a reduction in flow rate. All filter systems will require periodic cleaning to avoid clogging, a workable system for this application will require easy or automatic cleaning of the filter.

Russell-Finex, Orival, Eaton, Forsta and Tekleen all produce similar metal or polycarbonate self-cleaning filters employing either a back flush or rotating cleaning system.

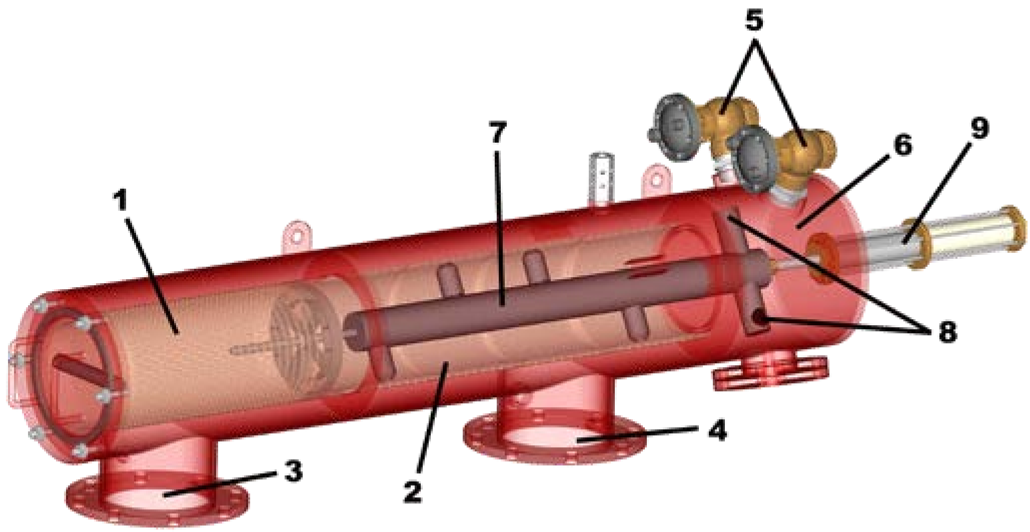


Figure 9. Orival OR series as an example of self-cleaning filters

Discussions with a leading UK treatment tank manufacturer resulting in the design of a similar system tailored to their tank designs. The design featured a polycarbonate screen with a 10µm pore size and a back wash cleaning system triggered by a drop in pressure across the screen.

Eliquo-Hydrok suggested a pile cloth media filtration system where the water is passes through a series of pile "carpets" with an automated systems raising and cleaning each carpets as it becomes clogged.

Other technology– **Lamella Clarifier**

A lamella clarifier removes sediment by passing the fluid over a series of inclined plates. This technology is easily scaled up to handle any required flow rate, although the size of the unit can become significant.

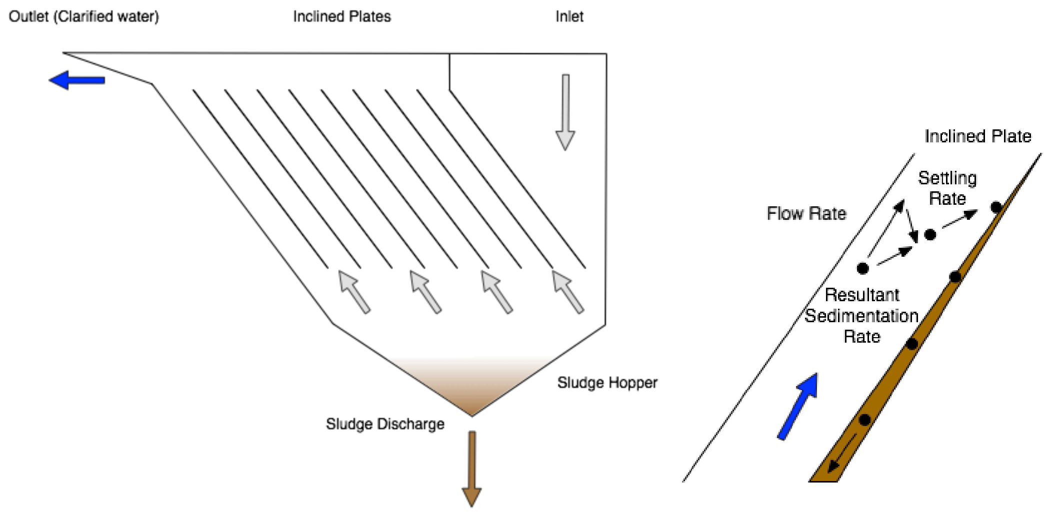


Figure 10. A Lamella clarifier

Lamella plates rely on the sedimentation of the particles in the water offering a huge surface area on which the sediment can collect, however given the expected low SG of the treatment tank load it is not certain how effective this technique would prove.

Micro bubble/ Dissolved air flotation

This method raises low SG particles to the top of the vessel on a cushion of fine bubbles where they can be easily skimmed off. Due to the complementary nature of this method to vortex separation it could possibly be combined with such a system to remove the larger less dense material prior to entry into the vortex separator.

Centrifugal filters

Cintropur manufacture a range of relatively small "centrifugal" filters. These filters combine elements of the vortex separators with standard filtration, essentially the return flow of the vortex passes through a filter sleeve resulting in the larger, heavier particles collecting at the bottom of the vortex and the smaller particles being trapped on the filter sleeve.

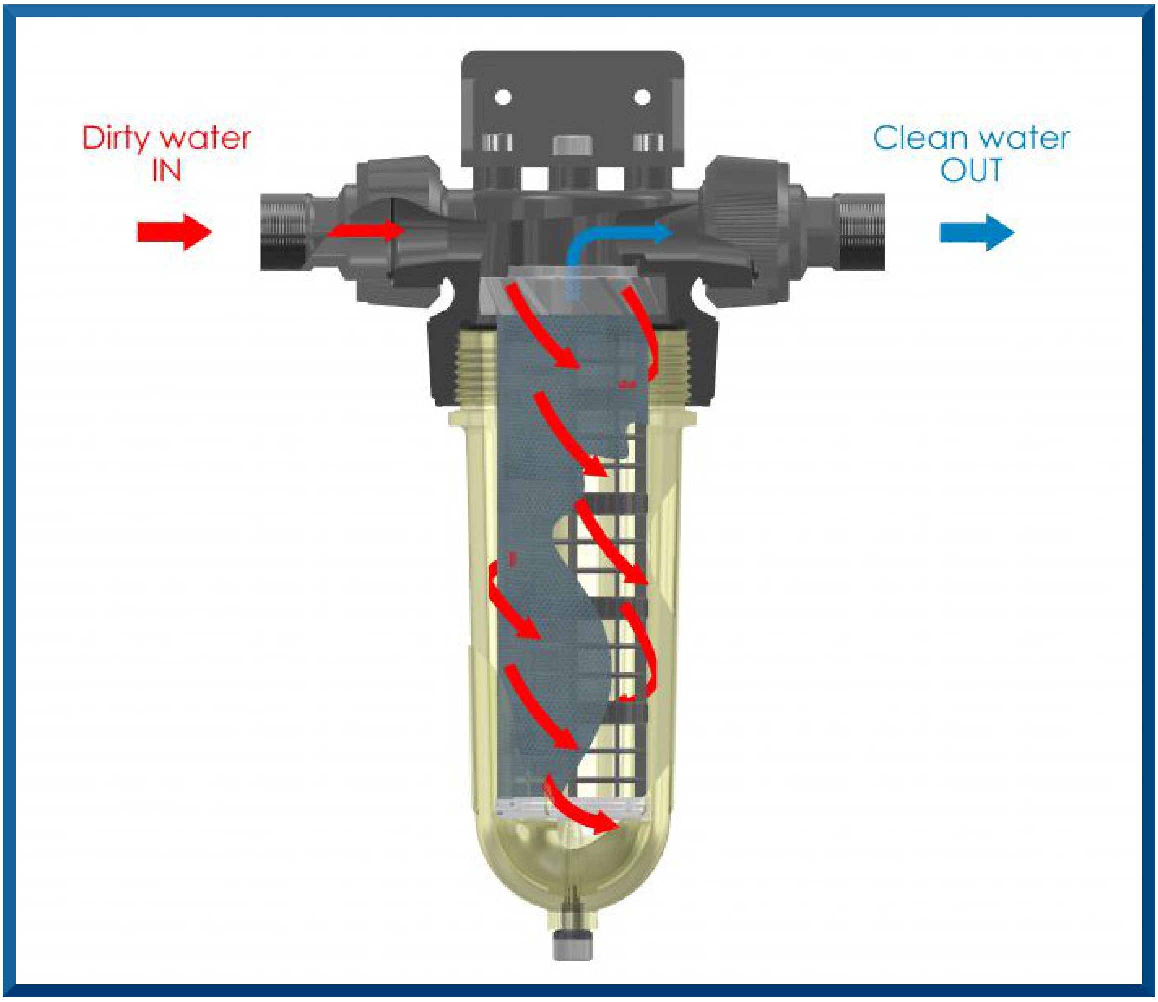


Figure 11. A centrifugal filter

This combination of processes would be expected to work well with the mixed nature of the particles found in tank water. The Cintropur units also have the advantage of being considerably less expensive than any other filtration device considered, with the largest unit the NW800 costing ~£500. However the NW800 is only rated for flow rates of 32 m^3/hr meaning that depending on the current tank setup 2 or even 3 of these units would need to be fitted in parallel in order to cope with the flow rate. The Cintropur units also do not have any automated cleaning system, there is a release valve for removal of sludge from the bottom of the unit but the units require disassembly and removal of the filter sleeve for washing.

Commercial scale testing

During the early part of 2017, discussions were held with a number of commercial growers who had expressed an interest in trailing filtration either on its own or in conjunction with other treatments. In addition, some growers independently investigated the use of filtration although to our knowledge none of them installed filtration equipment for the 2017 dipping season.

Commercial scale testing at Carwin Farm

We are grateful to Andrew Richards of Carwin Farm who agreed to host commercial scale testing on his bulbs stocks during 2017. The test setup included centrifugal filtration, UV sterilization and chlorine dioxide. Equipment was ordered late spring and installed during August 2017 on one of their tanks. The total cost of purchasing and fitting the equipment was £6,273.

Equipment:

4 x NW800 centrifugal filters - £1,782

10 and 25 µm filter sleeves

150 µm nylon filter sleeves

1 x SDB 550PH UV sterilizer - £2,913

Assorted ancillary fittings and fixtures - £1,098

Installation and welding - £480

Installation and set up

The equipment was retro-fitted to one of the tanks at Carwin Farm (Figure 12). In addition, to the filters and UV unit, associated flanges, valves, elbows and hose were required to connect the existing pump to the filters and UV unit. Directly following the pump outlet (3” pipe) the water flow was split into two (2” pipes) with each flow passing through two filters. The flow was then recombined (3” pipe) before passing through the UV sterilizer and being directed back to the treatment tank. Filter sleeves were changeable, the first filter in each series was 150 µm and the second was changed between 10, 25 and 150 µm as needed. The water flowed through the UV unit at all times but the lamps were only turned on when appropriate. Butterfly valves (blue handles in Figures 12 and 13) were installed to isolate the four filters to allow the filter sleeves to be cleaned as required.

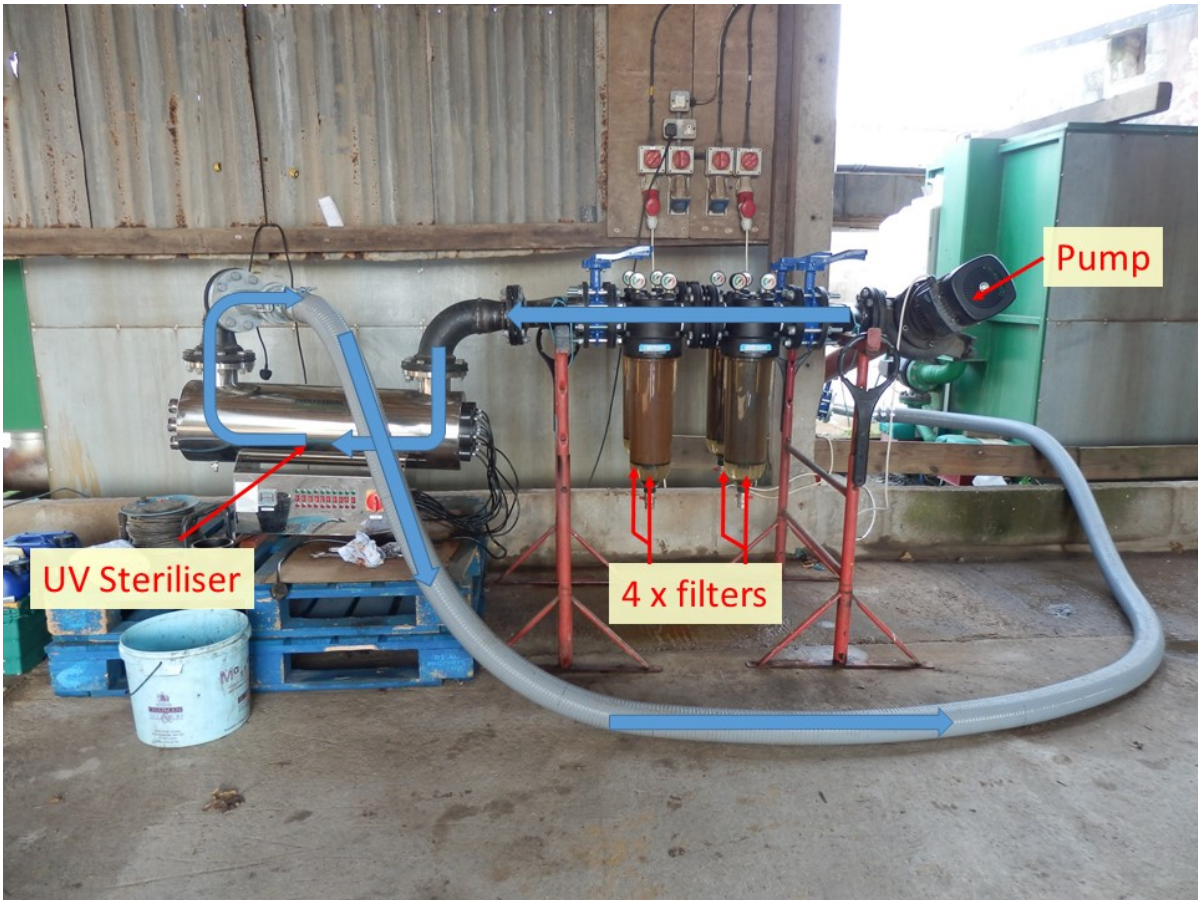


Figure 12. Retro-fitted filtration and UV system at Carwin Farm, Cornwall

Figure 13. Retro-fitted filtration system at Carwin Farm, Cornwall

Testing

The retrofitted filtration and UV system was commissioned and tested by filling and running the tank with just fresh water. Prior to filling, it was noted that the tank was lightly rusted (Figure 14) and turning on the pump encouraged a considerable release of rust, which quickly clogged the filters and necessitated removal and cleaning of the filter sleeves multiple times. Apparently, this rust deposit is normally removed by the first batch of bulbs which turns them a noticeable orange colour.

A protocol to cope with this high loading on the first filling of the season would need to be developed in the future. The size of the rust particles was not determined.

Figure 14. The bottom of an empty tank at Carwin Farm

The commercial testing was divided into six batches of bulbs, with each batch contains two bins of bulbs (approximately 600 kg). The individual batches and their respective treatments are detailed in Table 3. For batch 1, the tank was heated to 44°C, a water sample was taken (sample 1), bulbs were loaded and 150 and 25 µm filter sleeves were installed; the UV unit was turned off. The water at the start of this run was the "cleanest ever seen" although this might be because the normal rust load had already been removed. During the run the filter screens required cleaning in order to keep the water flowing and even at this early stage, it was obvious that unless there was a very clear benefit to the filtration, a system that required the filter screens to be manually cleaned during every run was not going to be practical. Sample 2 was taken from the tank at the end of this run.

Table 3. Summary of batch testing and treatments

Bulb batch	Water status	Filter 1 (size µm)	Filter 2 (size µm)	UV (on or off)	Respective water sample
1	Clean	150	25	Off	1,2
2	After 1 batch	150	25/150*	On	3
3	End-of-season	150	150	Off	5,7
4	End-of-season	-	-	-	6
5	End-of-season	150	150	On	8
6	End-of-season	-	-	-	9

* Filter was changed half-way through the run to evaluate change in water flow

For batch 2, the water was unchanged and the filters were again run at 150 and 25 µm. The UV unit was turned on for this run. During this run the tank water took on a green hue and an oily scum developed on top of the tank, it was not clear what the cause of this was but it was assumed to be related to the use of the UV steriliser for the first time. Again the filter screens required removal and cleaning and the decision was taken to change both screens to 150 µm (from 150 and 25 µm pairs). The change provided a marked increase in water flow. Sample 3 was taken at the end of this run. During this run three (out of ten) UV lamps gave a fault warning with two of these being resolved by restarting the unit. On investigation after the run the third fault appeared to be a wiring issue as the bulb functioned when changed to a different fitting and other functioning bulbs failed to work in the failing fitting.

The next batch was due to be run with chlorine dioxide (in the form of Activ-ox, supplied by Feedwater). A Palintest meter was used to monitor in tank chlorine dioxide concentrations but despite adding increasing levels of chemicals (until the supply was exhausted) the in-tank level of chlorine dioxide did not at any point reach a detectable level. Sample 4 was taken for testing at this point but it was decided not to run a load of bulbs as it was unlikely to give a meaningful result and the bulbs were unlikely to be protected from Fusarium. This tank of water was disposed of and the tank was refilled with water stored from the end of normal season testing. This water was considerably dirtier compared to batch 1 and 2 and also contained FAM30, Storite and Bravo at typical levels and any dirt or pathogens built up over the dipping season.

Batch 3 used end-of-season water. Water sample 5 was taken after heating but before the bulbs were introduced. Bulbs were added and the batch was run using 2 x 150 µm filter screens. The UV was turned off. Water sample 7 was taken at the end of the run. The filter sleeves were dirty at the end of the run but didn’t require cleaning mid-run. No other chemicals were added.

Batch 4 was the same as batch 3 except that no filters were used. Batch 4 acts as a control for batch 3. Water sample 6 was taken at the end of the run.

Batch 5 was the same as batch 3 except that the UV was on. Water samples 8 was taken at the end of this run. There was no repeat of the green hue and oily scum as observed during the previous UV run. It is possible that the faster flow of water obtained through use of only 150 µm filters was the reason for this. The filter sleeves were dirty at the end of the run but didn’t require cleaning mid-run. No other chemicals were added.

Batch 6 was the same as batch 5 except that no filters or UV were used. Batch 6 acts as a control for batch 5. Water sample 9 was taken at the end of the run.

Once treated, each batch of bulbs was labelled and dried before being planted in marked plots in the commercial fields. Assessments will be made in the spring of 2018 and 2019.

Pathogen testing on water samples

All the water samples taken from commercial testing at Carwin Farm were transported back to Warwick Crop Centre for analysis. Sub-samples (neat, 1 in 10 and 1 in 100 dilutions) were spread onto malachite green agar plates to detect/quantify fusarium. No fusarium was detected but other colonies did form on some plates giving some indication of the overall biocidal nature of the various treatments (Table 4).

Sample 1 (fresh tank, filled the previous day, with no chemicals) showed a low level of colony forming units (CFU). This level increased approximately 20-fold following addition of bulbs to the tank (samples 2 &3). The increase following the second run was less than that seen after one run, possibly due to some action of the UV treatment. Treatment by ClO_2 reduced the CFU count almost to zero, this was despite the level of ClO_2 in the tank not reaching a detectable level on the meter. Despite having a whole season's build-up of dirt all end of season water samples showed zero or close to zero CFUs suggesting that the remaining fungicide/biocide levels were effective.

Additional chemical analysis on water samples

Although the investigation of fungicide active ingredients was not within the project remit, the opportunity was taken to test for the presence of popular actives. Water samples were tested by HPLC to quantify the levels of thiabendazole (Storite) and chlorothalonil (Bravo). Levels detected were well below the recommended rates (thiabendazole 275ppm (10 litres of Storite/Tezate in an 8000 litre tank) and chlorothalonil 500ppm (8 litres of Bravo in an 8000 litre tank).

Levels of thiabendazole decreased fairly uniformly with each run in both tanks, this suggests that the product is lost, either through adherence to the bulbs or degraded through heat action. There did not appear to be any difference in the reduction between filtered and unfiltered tanks.

The chlorothalonil level decreased by 14ppm and then 15.4ppm for each run in tank 4. For tank 1 the level after 2 runs showed no significant change (49.4 to 54.0ppm). This difference across the filtered and unfiltered tanks may suggest some loss of product due to filtration. If this is proved to be the case then a higher top up rate may need to be used for Bravo if filtration is used.

Table 4. Water sampling regime and conditions

Sample	Date (& time)	Treatments	CFU/ml	Thiabendazole ppm	Chlorothalonil ppm
1	26/09/17 (08.30)	No bulbs. Fresh water with filtration. No chemicals.	310	*	*
2	26/09/17 (12:15)	With bulbs. One batch water with filtration. No chemicals.	6000	*	*
3	26/09/17 (16.30)	With bulbs. Filtration plus UV. No chemicals.	7000	*	*
4	27/09/17 (09.00)	No bulbs. End-of-season water with ClO_2.	40	*	*
5	27/09/17 (13.30)	With bulbs. End-of-season water.	0	11.6	85.1
6	27/09/17 (13.30)	With bulbs. End-of-season water.	0	9.9	49.4
7	27/09/17 (17.30)	With bulbs. End-of-season water with filtration.	20	7.7	71.1
8	27/09/17 (21.30)	With bulbs. End-of-season water with filtration and UV.	0	6.8	55.7
9	27/09/17 (21.40)	With bulbs. End-of-season water.	0	5.2	54.0

0* - not tested as fresh water with no added Storite or Bravo

Additional testing

Further testing was carried out at Carwin Farm to investigate some of the points raised by the initial tests. Flow rates were monitored using combinations of either 150/100 µm or 150/50 µm filter sleeves. It was observed that the 50 µm filters were too fine to allow a good flow however the 100 µm filters were acceptable.

Reflections on commercial scale testing

The use of filter screens with pore sizes smaller than 100 µm (50, 25 and 10 µm) reduced the flow of water to the point where we judged that it was not moving through the bulb tanks effectively; and this problem became worse as the filters became progressively more blocked. On-farm and laboratory observations lead us to believe that filtering to 100 µm will only have a small positive effect and will not be sufficient to clean the water adequately to allow UV treatment to be effective.

One solution to obtain cleaner water would be to specify higher pressure pumps, however, it is not clear at this stage whether the additional pressure would result in improved flow through the HWT system or simply reduce the amount of time to block the filters. Our trial required filters to be manually cleaned so higher pressure pumps would also require auto cleaning filters to avoid imposing too great a workload on the operator.

The nature of UV sterilisation, like that of chemical biocides, means that there is no observable effect of the treatment. However, bearing in mind the fact that filtration appeared to provide only a minor improvement in water clarity, we assume that dirty tank water severely hampered the ability of the UV light to penetrate through the sterilization chamber and as such no broad conclusions can be drawn from these results. However, the treated bulbs will be monitored in spring 2018 and 2019 for any differences in flower performance.

Whilst the filtration setup had some positive effects, it also revealed a number of limitations so further work is required on screen sizes and flow rates before any recommendations can be made. It is also possible that the filtration setup used will not be able to cope with either the particle size and flow rate and that more supplicated and expensive filtration technologies will be required to significantly improve the cleanliness of tank water.

The chlorine dioxide results proved interesting. The advice from Feedwater Ltd was that to work effectively an automated dosing system was required to provide a steady supply of low levels of chlorine dioxide and that if the water was dirty the efficacy would be lessened. We did not fit an automated dosing system for these tests and the water was not filtered to the level we had hoped for. In this context, the results from the pathogen testing were promising, showing a greater than 99% reduction in CFUs. It is not clear however whether this was the effect of a one-off sterilization of the tank water when the chemicals were added or whether there would be any lasting effect over the course of a 3 hour run. In retrospect, it was a mistake not to run a batch of bulbs although it is planned to rectify this in 2018 using an automated dosing system.

The levels of both thiabendazole and chlorothalonil were found to be low in the end of season tank water. Despite the water having a considerable build-up of debris and potentially pathogens from the season's dipping this water proved to be very low in pathogens when samples were cultured.

Estimation of suspended solids

The assumption at the beginning of the project was that a majority of the particulates in tank water would be derived from bulb scale and therefore be relatively large. However as testing progressed, it became apparent that this was not the case and that a greater understanding of particle size and origin was required. Consequently, a one litre water sample was collected from one of the treatment tanks, during operation, approximately one month into the dipping season.

The water sample was passed through a series of filters starting with a milk filter (pore size ~250 µm, followed by 125, 80, 20 and finally 0.4 µm filters). The sample passed through each of the filters from 250-20 µm without the removal of any solids. On passing through the 0.4 µm filter a layer of silt was deposited on the filter and the liquid appeared to be clear of any further suspended material (Table 4).

Table 4. Total suspended solids in dirty water

Particle size (µm)	Suspended solids (g)
0.4 to 20	1.6
>20	0
Total	1.6

Conclusion:

All of the suspended solids came out at between 0.4 and 20µm and are therefore very small particles. It is likely that at this size range these particles are mainly soil based fine silt or clay particles rather than fragments of bulb scale. The expectation was that bulb scale would make up a significant proportion of the suspended solids. It is therefore important to find that in fact the size of suspended particles is very much concentrated in the very fine range. Any filter system designed to remove these particles will need a pore size of less than 20µm and preferably 10µm or lower.

Filtration conclusions

It has been established that commercial tanks are low pressure systems with mainly very small, low specific gravity particles but also some larger particles – enough to clog small pore filters but less than expected. Although, we plan to conduct further filtration tests in 2018, if one of the conclusions of this project is that clean water is a pre-requisite for future treatment, then more sophisticated filtration systems will be required to achieve it. To use 5-10 µm screens for successful UV operation, would likely require (new) higher pressure pumps, to maintain an adequate flow rate through the tanks, and auto-cleaning filters. If the use of chlorine dioxide was the objective, rather than UV, filtration at 10-20 µm is likely to be required (at least in Cornwall) to provide clean enough water to allow it to be effective and to improve efficacy of other chemical treatments. Both of these systems would be more expensive than the test system and more difficult to retrofit to an existing HWT system.

Increased temperature HWT treatment

In addition to continuous filtration and UV treatment, static batch sterilisation was investigated. This involves raised the temperature of tank water to 60°C overnight (without bulbs) as this temperature has been shown to provide effective control of Fusarium.

Testing was carried out at two farms in Cornwall. At farm 1, raising the water temperature to 60°C overnight was easily achieved using their standard equipment. Water heating was turned off once 60°C was reached and allowed to cool. The temperature was found to have dropped to 50°C by morning but the addition of top up water (4000l/day) and bulbs brought the water temperature back down to 44°C at the start of treatment.

At farm 2, raising the tank temperature to 60°C was also achieved without problem and the opportunity was taken to dip some bulbs at this raised temperature. Two varieties (Finland and St Patrick's Day) were used for these short dips as they were found to have significant base rot and so of little commercial value. The bulbs were either control (no dip), dipped for 5 minutes at 60 °C or for 11.5 minutes at 60°C. During these dips the core temperature of a sample of the bulbs was measured. Dipping for 5 and 11.5 minutes raised the core temperature to 51 and 56°C respectively.

The bulk of the bulbs were planted as normal but two subsamples of bulbs were cut open to assess the level rot. The first straight after dipping and the second after being incubated for 28 days at 25°C in moist conditions to encourage rots to develop. Post-dipping, the levels of rot across both varieties were fairly consistent with 20-25% of Finland and 25-35% of St Patrick's Day bulbs showing dry rot (Figure 15).

Following incubation the level of dry rot had increased in all test groups (Figure 16). For Finland the increases in rots for the control and 5 minute dip groups were modest (25% to 30% and 20% to 27.5% respectively) however the increase in the bulbs dipped for 11.5 minutes was more significant (25% to 67.5%). This suggests that for the Finland bulbs a 5 minute dip at 60°C was either neutral or slightly beneficial but the longer 11.5 minute dip actually increased the level of dry rot in the bulbs.

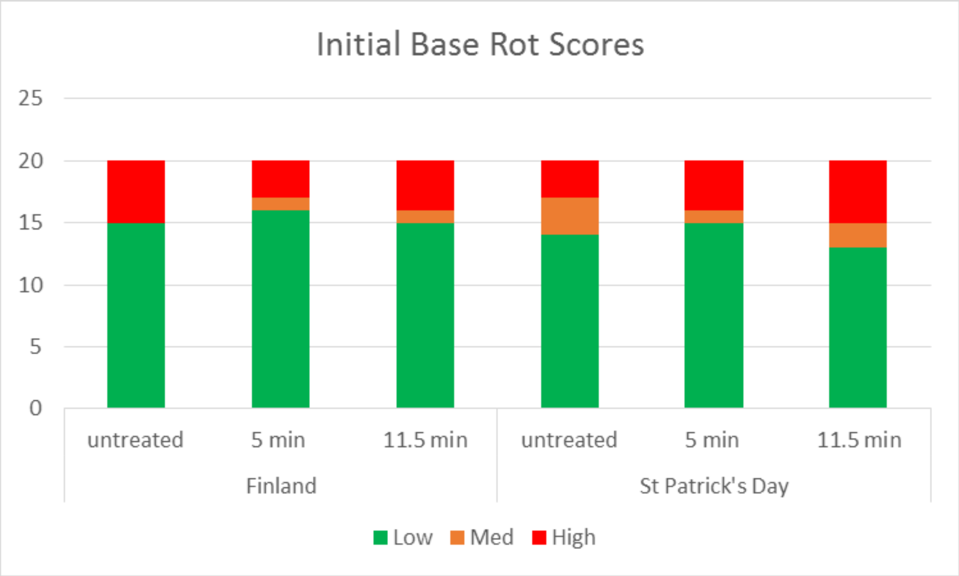


Figure 15. Initial base rot scores for commercial thermal treatment

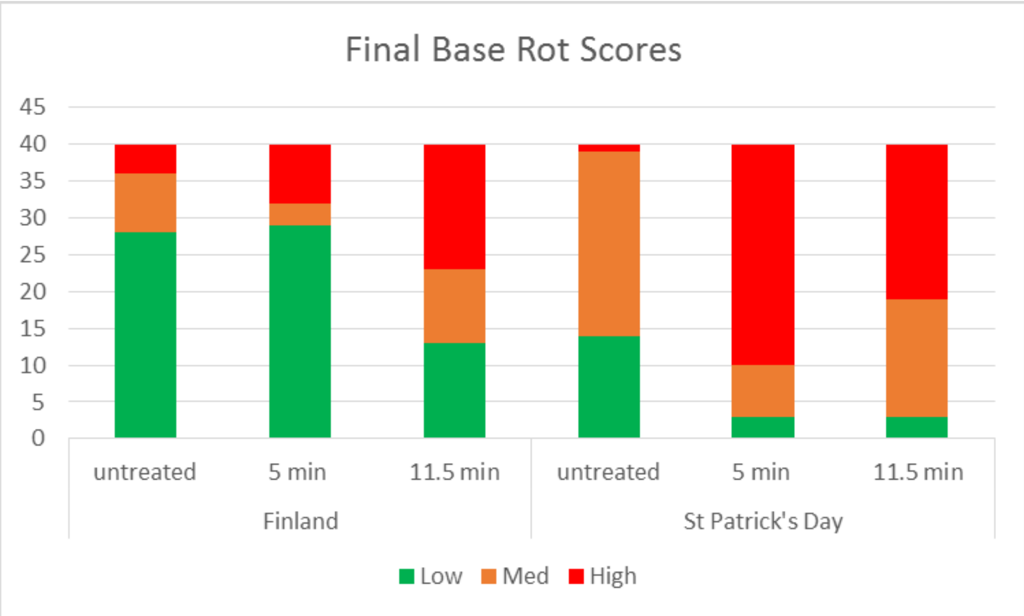


Figure 16. Final base rot scores for commercial thermal treatment

For the St Patrick's Day bulbs, which had shown slightly higher levels of dry rot in the initial observations, the levels observed after incubation were consistently high. Control bulbs showed 65% damaged up from 30% initially, although most of these showed moderate rather than the most severe damage. The 5 minute and 11.5 minute dipped bulbs both showed 92.5% damage (up from 25% and 35% respectively).

The dipping of bulbs for short periods at 60°C, whilst successful from a technical point of view, gave mixed results. In the case of the Finland bulbs those dipped for 5 minutes at 60°C showed the lowest levels of dry rot both initially and after incubation. However these results were only slightly lower than for the control bulbs. The 11.5 minute dip actually increased the levels of dry rot damage after hot boxing. The longer dip at high temperature would be expected to reduce the level of viable fusarium, however it would seem that any such effect was more than counteracted by some secondary effect, most likely a softening or damage to the bulb tissue allowing a more rapid spread of surviving spores.

In the case of the St Patrick's Day bulbs the levels of dry rot present, particularly after incubation, were so high that it is hard to draw any conclusions beyond the fact that neither of the dips appeared to have any beneficial effect.

Following dipping, the remaining bulbs were replanted at Carwin Farm for assessment in spring 2018 and 2019.

Flower assessments

The trial plots were assessed on 26th March 2018. The number of flowers were counted in a representative 1.5m length of each of the two rows for each treatment (Figure 17 and Table 5). The results show that dipping bulbs at 60°C for five minutes had a slightly negative effect in comparison to not dipping the bulbs but the difference was not significant. However, dipping for 11 minutes did have a significant negative effect, suggesting that a dip of this length is damaging to the bulbs.

Table 5. Flower numbers along a 1.5m length for bulbs dipped for 0, 5 and 11 minutes.

	Row 1 flower count	Row 2 flower count	Mean flower count
Finland, Control	24	35	29.5
Finland, 5 minutes	22	26	24
Finland, 11 minutes	0	4	2
St Patrick, control	27	22	24.5
St Patrick, 5 minutes	19	21	20
St Patrick, 11 minutes	1	0	0.5

Work in year 1 showed that dipping for three minutes at 60°C provide effective control of (surface) Fusarium, so while it is too early to recommend this approach, in situations where pest control is not a priority, short dipping is likely to be an effective approach to surface sterilisation of bulbs without affecting bulb physiology or flower yield. However, it is recognised that the logistical difficulties of short dipping are considerable, especially with drive-in front-loading HWT systems. It is hoped to repeat the trials in 2018 and to assess the year two flowers on the existing plots.

Figure 17 Assessment of year 1 flowers for heat treated bulbs. a-c Finland, a – control, b – 5 minute dip, c - 11 minute dip. d-f Variety St Patrick's Day d – control, e – 5 minute dip, f – 11 minute dip.

Although thermal treatment of bulbs was not part of the project remit, there was some interest from growers as to whether it would be a viable approach, so further testing was undertaken at Warwick Crop Centre as part of a student project. The cost was covered by the University of Warwick, not AHDB, but the results are reported here as a matter of convenience.

Batches of 10 bulbs were placed in a water bath at temperatures from 60°C to 70°for durations from 3 to 10 minutes (Treatment are shown in table 5). The interior temperature of these bulbs was tested using a thermal probe at 10mm depth, 20mm depth and bulb centre. This was used to determine the likelihood of damage to the bulbs.

Table 5. Thermal treatment at Warwick Crop Centre

Treatment number	Duration (minutes)	Temperature (°C)
T0	-	-
T1	3	60
T2	5	60
T3	8	60
T4	10	60
T5	3	65
T6	4	65
T7	5	65
T8	10	65
T9	3	70
T10	5	70
T11	10	70

The results show, as expected, that with an increased water temperature or with longer dipping times the core temperature of the bulbs increases (Figure 18). It is however interesting to note that the data obtained at Farm 2 shows a much higher core temperature than the equivalent data obtained at Warwick. This effect was more noticeable at the shorter dipping times. It may be that this is due to the addition of bulbs to the small tanks at Warwick causing a drop in water temperature, which is likely to be less of an issue in commercial 6000l tanks.

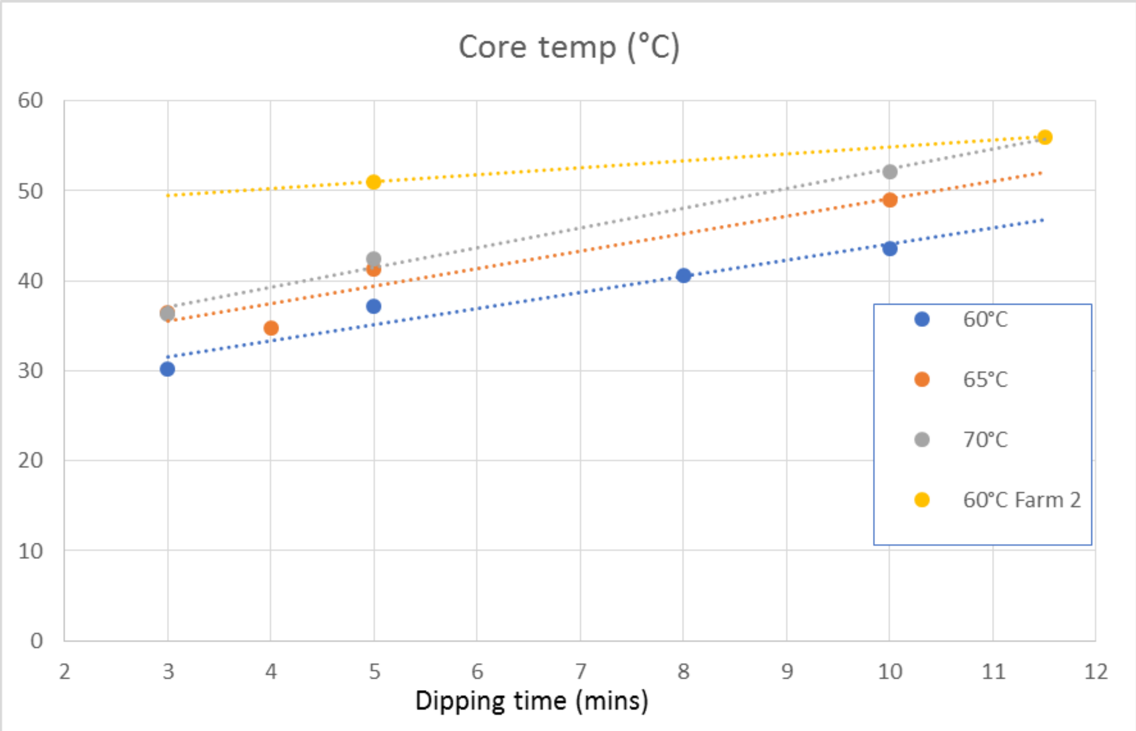


Figure 18. Bulb core temperature at different dipping temperatures and durations

The water bath treatments were then repeated using batches of 20 bulbs. Once treated these bulbs were placed in an incubator at 25°C. After a month the bulbs were dissected and basal rot was scored from 0 to 10. Scores were then grouped to give low, medium and high basal rot categories as indicated in the scoring method (Figure 19).

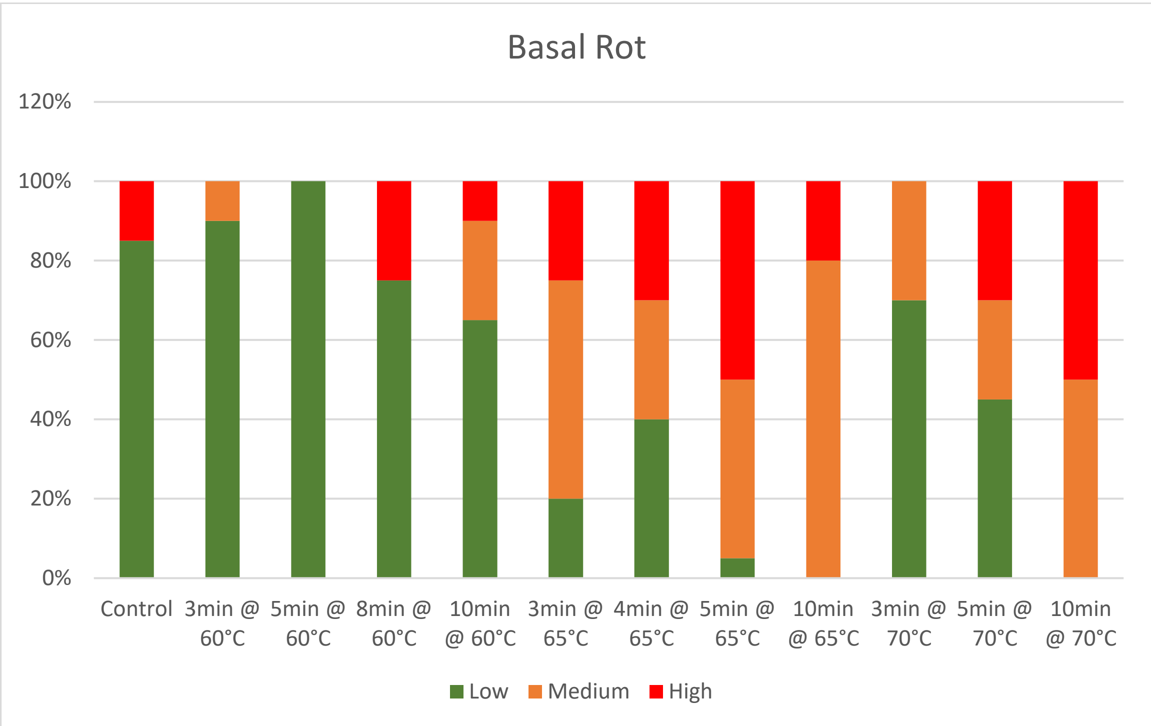


Figure 19. Incidence of basal rot following incubation (Warwick thermal trial)

The water bath treatments were repeated a third time in batches of 20 and these bulbs were planted in pots around 8cm from the surface. Flowers will be assessed in the spring 2018 allowing for quantification of any thermal damage to the bulbs

Chemical Testing, Dosing and stability

Although not part of the original project there was considerable interest in the dosing and stability of fungicide. The opportunity was take to collect water samples from two commercial growers to quantify fungicide levels under different circumstances. Samples were tested for levels of thiabendazole and chlorothalonil by HPLC. The results are presented in Table 6.

Table 6. In-tank fungicide levels at different water status (Farm 1)

Sample number	Water status	Thiabendazole (ppm)	Chlorothalonil (ppm)
1	Full strength chemicals at ambient temperature	533	351
2	After heating water to 60°C	363	55
3	After one load	413	187
4	After 9 loads	510	144

Treatment at 60°C lowered the levels of detectable thiabendazole by approximately 30% and chlorothalonil by approximately 85%. Topping up the tanks after each load recovers some of this activity particularly in the case of thiabendazole. For thiabendazole we can accurately predict the level after 9 loads if we assume that each top up increases the tank concentration by the same amount as the first one (~50ppm) and that heating to 60°C after every 3 loads (nightly) reduces the level by approximately 30%. Perhaps due to the very high level of degradation of chlorothalonil it is not possible to produce a similar model that fits the data for this chemical.

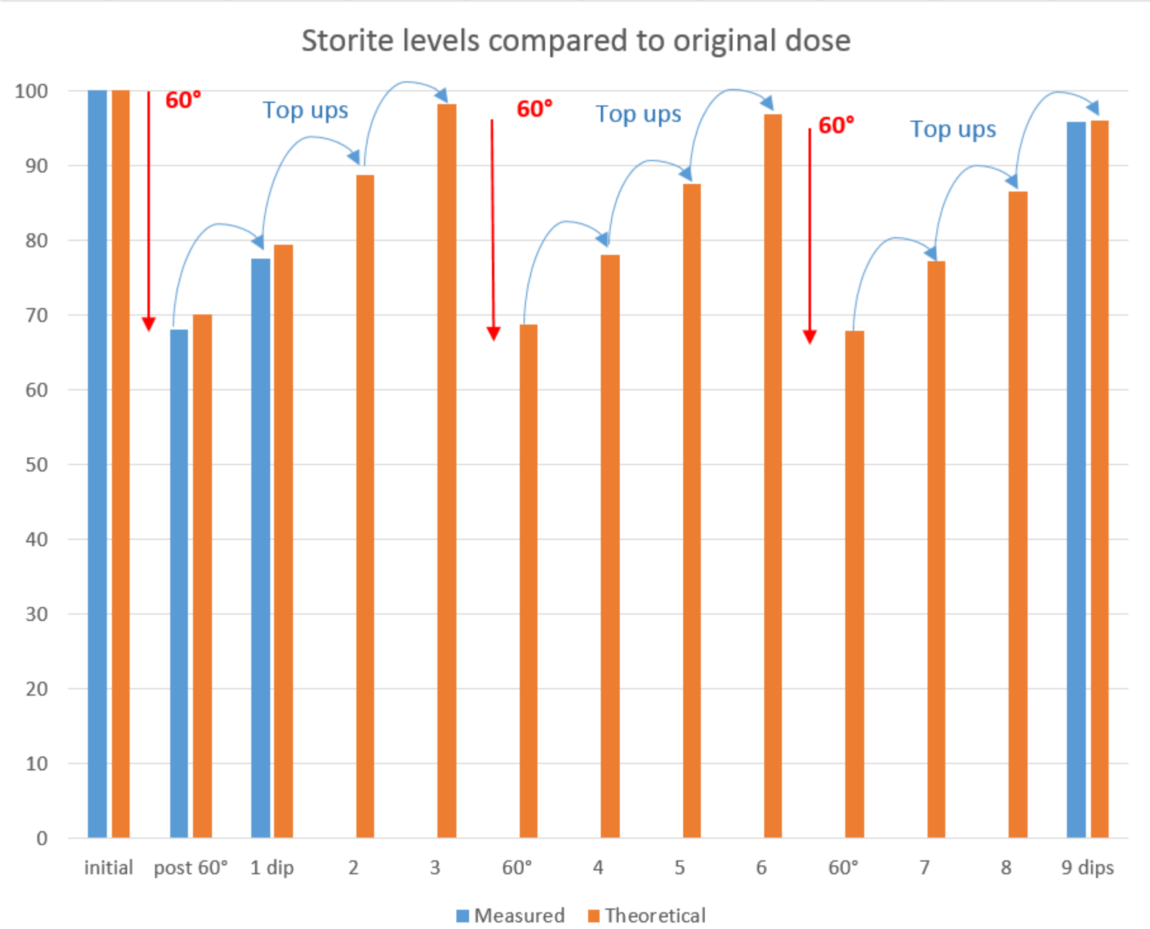


Figure 20. An illustration of how levels of thiabendazole (Storite) vary under temperature and top-up regimes. These theoretical values are based on a 30% loss on heating to 60° and 50ppm increase of concentration for each top up.

At farm 2, water samples were collected at different times and levels of thiabendazole (Storite) and chlorothalonil (Bravo) quantified by HPLC. Levels of thiabendazole decreased fairly uniformly with each run which suggests that it is lost, either through adherence to the bulbs or degraded through heat action, at a greater rate than it was topped up. Levels of chlorothalonil decreased by 14ppm and then 15.ppm for each of two runs. Overall levels were well below the recommended rates (thiabendazole 275ppm and chlorothalonil 500ppm).

This ad-hoc testing of tank fungicide levels revealed some unsettling but perhaps not unsurprising results. Although all growers will strive to ensure that levels of fungicides are at the recommended levels at the start of the dipping season, and will estimate how much topping up is required during the season, they have no way of monitoring levels and therefore no way of knowing the amount of active ingredients that are lost through adherence to bulbs or degraded through thermal action. The results of this very limited work suggest that active ingredients are typically below recommended levels but the effects of this are mostly unknown. Given the importance of this topic to growers, it is hoped to undertake a more thorough survey in summer 2018.

Conclusions

The second year of the project moved the work from laboratory to farm. The conclusions of the work so far are:

- Laboratory testing of UV radiation showed that it could provide effective control of Fusarium spores but that continuous treatment (rather than batch dosing with chemical biocides) did not provide complete control. UV was tested on farm in summer 2017.
- Chlorine dioxide also provided effective control under laboratory conditions although it proved difficult to quantify tank concentrations under on-farm conditions; as it is broken down very quickly once added to tanks. A commercial continuous dosing system will be trialed in 2018.
- Thermal treatment of tank water was shown to provide complete control of Fusarium spores in laboratory conditions and a number of different temperatures and durations were tested under both controlled and commercial conditions.
- Filtration proved difficult to make work effectively under commercial conditions and the approach requires further consideration. Filtration is still likely to be a positive addition to HWT but equally it is important that bulbs are as clean as possible when they enter the HWT.
- Short thermal treatments of bulbs was tested under laboratory and commercial conditions with bulbs planted out for later evaluation.
- Field trials were established in Cornwall to evaluate the effect of UV and thermal treatments. This will be evaluated in 2018 and 2019.

Programme of work in 2018

- Filtration and UV approaches will be tested under commercial conditions in Lincolnshire.
- A commercial automated chlorine dioxide dosing system will be tested in Lincolnshire and/or Cornwall.
- Field trials in Cornwall will be evaluated
- Sampling of commercial tank water will be undertaken to gain better knowledge of biocide and fungicide concentrations.

Knowledge and Technology Transfer

Grower events. Details of the project and its objectives were presented at two grower meetings organised by AHBD:

- Spalding, Lincolnshire 25th May 2017)
- Redruth, Cornwall (17th May 2017)

Visits were made to four growers during the year. Discussions were held with Lin Secker of Secker Engineering regarding filtration.

An article is being prepared to the RHS Daffodil, Snowdrop and Tulip Yearbook 2018

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