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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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Grower Summary

Headline

- High light levels (PAR), vapour pressure deficit (VPD, >3 kPa) and temperature (>35ºC) have previously been linked to the expression of Pansy mottle syndrome (PaMS) symptoms. However, environmental monitoring during 2019 proved inconclusive
- While root development has not been linked directly with PaMS symptoms, poor root development may contribute to plant stress under challenging environmental conditions.
- Gravimetric techniques successfully managed irrigation at plug stage and promoted healthy root development.
- The poor irrigation management regime called 'Extreme Wet' regime, promoted poor root growth.
- Healthy root development can be promoted by irrigation regimes, supported by nutrient monitoring, that:
	- o Match irrigation application to water use.
	- o Allow growing media to dry back prior to irrigation.

Background

Previous environmental monitoring work (PO 016 and PO 016a) suggested that high temperature (>35°C), high vapour pressure deficit (VPD) (>4.5) and high light levels may be potential triggers for Pansy Mottle Syndrome (PaMS). The purpose of this work was to carry out monitoring of Pansy crops to further our understanding of the triggers of PaMS, and to develop recommendations for the mitigation of plant stress events that may contribute to symptom expression. Two irrigation demonstration events, hosted on grower holdings, were designed to present techniques to quantify the water volume applied to Pansy crops at plug stage, and to demonstrate the impact of a number of irrigation regimes on plant and root quality, and how they may help reduce PaMS.

Summary

WP1. Environmental monitoring

Objective: To monitor the environmental factors (light intensity, leaf temperature, air temperature, relative humidity and growing media moisture) *in-situ* on three commercial nurseries during propagation and post-transplant production phases.

Environmental monitoring took place on three commercial nurseries; Nursery A (propagation and pack production), Nursery B (pack production) and Nursery C (propagation). Equipment was delivered to the nurseries in week 30 (Nursery A) and week 31 (Nursery B and Nursery C) and set-up within new batches of Pansy crops.

At Nursery A, the environment was monitored in both the propagation and pack production areas, with batches of plants moving from one area to the other. Plants were both propagated and grown-on under glass. Plants were gapped prior to marketing.

At Nursery C, approximately one week after sowing, plants were moved from the germination room into a fogging area for a further week, and monitored until dispatch to Nursery B. As plants were dispatched from Nursery C to multiple nurseries for finishing, it was decided to monitor a single cultivar that was included in all deliveries to Nursery B. Plants tended to be moved between areas on this nursery, including for gapping.

Monitored batches from Nursery C were transported to Nursery B using refrigerated lorries, with the environment monitored during transit using Tinytag data loggers (temperature and humidity). Plug plants were then transplanted and the growing environment monitored, so that plants were monitored from sowing (Nursery C) through to marketing (Nursery B).

Environmental monitoring was carried out using Tinytag loggers and 30MHz equipment, all of which were set to record at five minute intervals [\(Figure 2\)](#page-17-0). A list of the equipment used at each site is found in **[Table 2](#page-16-0)**; this was supplemented with nursery-owned 30MHz equipment to increase coverage. ADAS and nursery-owned 30MHz equipment were calibrated against each other.

Environmental monitoring equipment was deployed at each site as follows: Tinytag data loggers (4 loggers; temperature, humidity, dew point); 30MHz multi-sensors (2 sensors; temperature, humidity, leaf temperature), light probes (1 probe; photosynthetically active radiation, PAR), and growing media moisture sensors (1 sensor; volumetric water content, VWC). Note, though, that moisture sensors were not used during the propagation phase of production at either Nursery A or Nursery C as the plug size is too small to accept the probes. The equipment were set to record at five minute intervals. Sowing, transport and transplant dates for monitored batches are detailed in **[Appendix 2](#page-50-0)**. Pansy crops were monitored by growers on a weekly basis, recording Pansy mottle and distortion symptoms and the proportion of the crop affected

Summary of results

PaMS was reported on all three monitored sites during 2019. Symptoms included mottle, distortion and loss of growing point.

- At Nursery A (propagation and pack production), there was low incidence of PaMS in all transplant weeks, both in pot and pack throughout the season. However, the symptoms were only observed post-transplant.
- At Nursery C (propagation) the worst symptoms were reported in weeks 31 33, and were predominately leaf mottling and distortion. Plants with symptoms were removed during production and at marketing, and non-symptomatic plants were then transported to Nursery B.
- At Nursery B (pack production) symptoms were present on arrival at the nursery (from Nursery C), and included leaf and flower mottling and distortion, and loss of growing points. Symptom severity was greatest in weeks 33 and 34, and included mottling and distortion of leaves and flowers and loss of growing point.

Symptoms tend to become apparent within crops over a period of days. The course of symptom development appears to be that one or two plants are affected initially but symptoms are expressed in more plants, and more fully, over the course of at least 2-3 days. This can make it difficult to identify the date of first symptoms in a large batch of pansies. In the scenario of nurseries B and C, where plug plants are produced by a young plant producer and then distributed to finishing nurseries, symptoms may be triggered at the propagation nursery, where any plants with visible symptoms are removed from the batch prior to dispatch, and more symptoms are present on arrival at the finishing nursery.

As the precise cause of symptoms is not known, and there is no differentiation between different symptoms (e.g. mottling, leaf distortion, stunting) in the data, it is only possible to identify potential plant stresses that may or may not cause the symptoms to arise. In addition, the extent of any delay between triggers (if they exist) and displaying of symptoms is also not known and it is therefore possible that the display of symptoms could be due to an accumulation of stresses over a long period of time, or conversely triggered by a single event.

Conclusions

The environmental monitoring carried out in 2019 did not identify triggers for PaMS. Previous work had suggested that, high temperature, VPD and PAR could be potential triggers, but these could not be correlated to symptom occurrence by the data for the batches of Pansies monitored in 2019. It is not clear if the symptoms considered to be part of the PaMS complex (mottling, distortion, lost growing points) are caused by a single trigger, different triggers or cumulative triggers. More detailed recording of symptoms including the precise date and time of first symptom, and the proportion of each symptom expressed (mottling, distortion and lost growing point) would enable these distinctions to be statistically analysed.

WP2. Demonstration of optimisation of irrigation practices

Objective: To demonstrate the effect of optimum and sub-optimum irrigation regimes (up to five) on Pansy growth and development during propagation

Seeds of two Pansy cultivars (anonymised at suppliers' request) were sown into 360-cell trays, (peat-based growing media) at Bordon Hill Nurseries, Warwickshire, in week 34 (21 August 2019) and placed into a temperature controlled germination room (15°C \pm 1°C) for five days. They were grown under glass until they reached cotyledon stage, then transferred to ADAS Boxworth, Cambridgeshire, in week 36 (4 September 2019) where they were placed on benches within an unheated polytunnel for the duration of the trial. Temperature and humidity were monitored throughout the trial using TinyTag data loggers.

The trays were monitored, weighed and irrigated according to the irrigation treatments (see below) on a daily basis for three weeks. Treatments ended in week 39 (23 September 2019), when the plugs had reached 3-4 true leaves, and an assessment was completed on the plugs.

Irrigation treatments

The irrigation treatments were based on the gravimetric method described in AHDB Factsheet 18/17 ('Methods and equipment for matching irrigation supply to demand in container-grown crops'). The gravimetric method uses the weight of water lost or taken up by the plant to calibrate the level of irrigation needed for a particular combination of plant, growing media, container size and plant growth stage. This was then used to determine the 'Working Water Capacity' (WWC) required to re-wet the crop to container capacity from the 'Need to Irrigate' stage without applying excess.

The process to determine the WWC was to irrigate the containers (a sample size of at least eight pots or trays) to full capacity and allow to drain for 30 minutes. Each container was weighed after 30 minutes, and left to dry back to the stage at which irrigation was judged necessary. Once the containers reached the 'Need to Irrigate' stage they were re-weighed. The difference in weight between the container capacity and the 'Need to Irrigate' stage was the WWC.

On 5 September 2019, all trays were irrigated to full capacity, allowed to drain for 30 minutes, and then weighed. The trays were weighed again after a further 2.5 hours, and again 2 hours after that, to gain an understanding of how quickly the plug trays would dry back. The trays were then divided into the five irrigation treatments, so that there were three trays per cultivar, per treatment. Irrigation treatments began on 10 September 2019, once the 'Need to Irrigate' stage had been established. The amount of water applied to each tray was dependent on the weight of the tray. Irrigation treatments were as follows:

- **T1 'Extreme Wet'** water twice per day (am and pm) to full capacity regardless of weight.
- **T2 'Extreme Dry' –** water 1 day or more after tray reaches 'Need to Irrigate' stage (tray weighs 700 g or less). Apply 700 g per tray.
- **T3 'Little and Often' –** water applied when weight lost from tray is < or near to 30% of WWC weight (tray weighs approx. 1190 g). Apply 210 g per tray.
- **T4 'Matched to Water Loss'** water applied when weight lost from tray is < or near to 60% of WWC (tray weighs approx. 980 g). Apply 420 g per tray.
- **T5 'Long Dry Down'** water is applied when weight lost from tray is >95% of WWC weight (tray weighs approx. 735 g). Apply 700 g per tray.

Because T1 was irrigated twice per day to full capacity regardless of water loss, this treatment was not weighed (**[Table 1](#page-8-0)**).

Table 1. Total water weight applied to each treatment, and number of applications from 10 September 2019.

Treatment	Total water applied	Number of watering events	
	(g)		
T1 'Extreme Wet'	To field capacity, twice per day	26	
T2 'Extreme Dry'	3500	5	
T ₃ 'Little and Often'	3570	17	
T4 'Matched to Water Loss'	2940	7	
T5 'Long Dry Down'	2800	4	

Summary of Results

There were clear differences between treatments, with effects noticeable both in terms of plant growth and root development. There were no signs of PaMS developing in the plug tray throughout the irrigation trial, likely as a result of the moderate prevailing environmental conditions.

'Extreme Wet' treatment. Plants of both cultivars achieved the highest plant quality scores in the 'Extreme Wet' (T1) treatment. Plants were darker green with poorer root development with fewer root hairs and many more water roots. While the top growth of the plants in this treatment appeared strong, the smaller proportion of roots present with root hairs would limit the plant's capacity to take up water under drier conditions.

'Extreme Dry' treatment. 'Extreme Dry' (T2) treatment plants were smaller and paler green, but with high root quality scores, with rooting in up to 75% of the plug. There were no ""water roots" and many more plants with root hairs.

'Little and Often' treatment. Plants were generally good quality if slightly pale and taller in this treatment. Root development was reasonable, although there were some "water roots" present. Fewer ""water roots"" may have developed had the growing media been allowed to dry back further before water was applied. This could be a useful regime with slight adjustments to the parameter for applying water (in this demonstration < or near to 30% of WWC) and / or the weight of water applied.

'Matched to Water Loss' treatment. The 'Matched to Water Loss' (T4) treatment produced good quality plug plants, although slightly smaller than in other treatments, with very good root development.

'Long Dry Down' treatment. Plants in the 'Long Dry Down' (T5), were similar to those in the 'Extreme Dry' (T2) treatment. Plant height was reduced, but root development was good, with roots throughout the plug, and plenty of root hairs. However, for plug production this treatment may be insufficiently forgiving, with little margin for error.

The irrigation regime impacted on root quality in two ways:

- **"Water roots"**. Allowing the growing media to dry back further between water applications, as in the 'Extreme Dry' (T2) and 'Long Dry Down' (T5) treatments appears to have prevented "water roots" from developing (**[Table 1](#page-8-0)**). The 'Little and Often' (T3) and 'Matched to Water Loss' (T4) treatments also allowed the growing media to dry back between irrigation applications and again, fewer "water roots" were produced.
- **Water quantity**. A greater volume of water was applied to plants in the 'Extreme Dry' (T2) and 'Little and Often' (T3) treatments overall compared with the 'Matched to Water Loss' (T4) and 'Long Dry Down' (T5) treatments. The highest root quality score was achieved by T4 in terms of root spread through the plug for both cultivars. This suggests that it isn't the volume of water *per se* that is critical to good root development, rather it is the period of time allowed for the growing media to dry back between applications. However, during cool conditions, where large water volumes are applied, it will take longer for the growing media to dry back, risking water root development.

Conclusions

For plug production, the aim is to achieve a balance between providing sufficient water to maintain growth while producing plants with well-developed roots; a difficult balance to achieve for small plugs. Plants develop stronger root systems when they are not overly wet, and are forced to search for water and nutrients.

The key factor for the success of any irrigation regime determined using the gravimetric method is correct judgement of when the 'Need to Irrigate' point has been reached. If it's judged that plugs need to be irrigated before they have dried back sufficiently, the growing media may always be too wet, particularly when using 'Little and Often' and 'Match to Water Loss' regimes. The 'Need to Irrigate' point will vary depending on plug size, growing media formulation, plant species and prevailing temperature; in-house trials would help to establish the parameters for when to irrigate.

"Water roots" have few or no root hairs, and have a 'glassy' appearance. They are produced in response to overwatering, when the substrate can be saturated for prolonged periods. With an abundance of "water roots", plants struggle to take up water as moisture levels reduce and would be less able to respond to increased demand for water and nutrients under high temperature, vapour pressure deficit (VPD) or light conditions. However, where "water roots" are present, if the growing media was allowed to dry back, the plants would produce new roots and develop root hairs, in response to their search for water and nutrients, producing plants more resilient to extreme changes in environment post-transplant.

For the most part, treatments T2-T5 may all be suitable for plug production, but with some adjustments to allow the growing media to dry back sufficiently between irrigation applications to minimise the development of "water roots". Consideration should also be given to the practicalities of the various irrigation regimes, for example while the number of irrigation events undertaken for the 'Little and Often' (T3) treatment may be easily managed in nurseries with boom irrigation, they may be less practical where crops are hand irrigated.

Irrigation of plants at plug stage is difficult to monitor closely as moisture probes are too large for the cell size, particularly those used in Pansy production. However, environmental monitoring systems that include wireless scales to measure plug tray weight that will help to automate the process are being developed. Use of gravimetric techniques to determine when to irrigate, linked to manually lifting trays, is a useful aid to setting irrigation parameters and training staff to irrigate to the correct level for healthy root development.

Financial benefits

Published statistics (Defra, 2014) estimate Pansy production in England and Wales at 9.4 million plants with a farm gate value of £2.1 million in 2014 (21p/plant). It is difficult to quantify plant losses due to PaMS for several reasons (the intermittent and variable nature of PaMS, growers rogueing distorted plants, unreported incidence, incidence identified as PaMS), however, reports have been received of 5-20% of batches on individual nurseries being affected. Based on Defra data, this would to equate to losses of £21,000 (1% of crop affected), £105,000 (5% of crop affected) or £420,000 (20% of crop affected). Additional costs are also incurred by nurseries in refilling plug trays or packs once affected plants have been discarded.

Action points

WP1. Environmental monitoring

Growers should take measures to monitor environmental conditions, and reduce plant stress:

- Monitor temperature, VPD, growing media moisture and nutrition.
- Ensure that during periods where extreme high temperatures are predicted, measures are taken to reduce plant stress by providing shade, maximum ventilation appropriate to prevailing weather conditions and adequate irrigation. High VPD may be reduced by increasing relative humidity by, for example, path damping and use of mist irrigation where available.

WP2. Demonstration of optimisation of irrigation practices

- Refer to AHDB Factsheet 18/17 'Methods and equipment for matching irrigation supply to demand in container-grown crops' for further details on the gravimetric technique.
- Gravimetric techniques for managing irrigation should be used in combination with monitoring of other factors including nutrition to determine plant and root quality.
- Calibrate the 'Working Water Capacity' (WWC) for each different combination of plant, growing media, plug / container size and growth stage used.
- Determine the WWC across a sample of at least eight trays / containers to obtain a robust value.
- Recalibrate the 'Need to Irrigate' point as the crop grows, basing decisions on the amount of time between water applications without impacting on final plant quality.
- Implement trials to determine the most suitable irrigation regime for your nursery production system.
- While the number of irrigation events undertaken for the 'Little and Often' (T3) treatment may be easily managed in nurseries with boom irrigation, they may be less practical where crops are hand irrigated.
- Extreme Wet conditions do not produce plants with well-developed root systems to support plant growth.

Science Section

Introduction

Pansy mottle syndrome (PaMS) has been reported (though not understood) since the 1960s, and is recognised as a measureable or visible change in plant growth and function (physiological response). Typical symptoms include leaf distortion, mottling, leaf bleaching, stunting and apical blindness (**Figure 1**). The extent of PaMS may vary from year to year on nurseries; bedding plant species including *Antirrhinum*, *Gerbera*, marigold, *Petunia*, *Primula*, stocks, sweet pea and *Verbena* can display similar symptoms. Determination of the cause is complicated by the transient and intermittent nature of plant response, difficulty in replicating the symptoms and linking the cause with effect (McPherson, 2010). Incidence of PaMS has increased in recent years, particularly in 2017 and 2018, prompting the need for more work to determine the influences on PaMS occurrence within the industry.

Figure 1. Pansy mottle symptoms: mottling and leaf bleaching (left), and leaf distortion (right)

Grower observation suggests that PaMS may be varietal, with incidence occurring in specific seed batches and colours. Blue and orange flowers appear to be affected more than other colours. Outbreaks have also been linked to environmental factors, occurring under humid conditions including warm, wet and windy weather when glasshouse vents are shut, causing humidity to increase within the glasshouse. Plug size (greater risk of PaMS in the larger module tested), growing media, and the plant hormone methyl-salicylate (associated with plant stress) also appear to promote the incidence of PaMS. Symptoms do not appear to be directly increased by fungicide, adjuvant or plant growth regulator application, the light or irrigation regimes tested, virus (tests proved negative), low irrigation or boron/calcium (levels confirmed adequate by plant tissue analysis) (McPherson, 2010). Other research has linked growth distortion with boron deficiency under high relative humidity conditions (100%); these conditions decrease water loss via transpiration, resulting in reduced boron uptake and movement from the roots to the shoot (Krug *et al*, 2013). The precise trigger for the expression of PaMS symptoms however, remains unknown.

Factors that may influences PaMS expression

A number of observations have been made not only from the results of this research programme but also from grower and researcher experience, that may be linked to the expression of PaMS symptoms

Environment

- Plant quality is improved when plants are kept actively transpiring, enabling water and nutrient uptake, and aiding internal temperature regulation. However, a number of occasions have been reported where weather events have caused glasshouse vents to close, upsetting the balance of the glasshouse environment, and PaMS symptoms have subsequently developed that have been attributed to this one event.
- The combination of the speed of environmental change and how extreme the environment parameter may be critical factors.
- High temperature (>35°C) combined with low humidity, resulting in high VPD (>4.5 kPa) combined with high light levels were indicated as triggers for symptom expression during previous research, although the number of incidents within monitored Pansy batches was low and therefore more data was needed to confirm these findings (AHDB PO 016/ 016a).
- These findings were supported by another trial (not designed to investigate Pansy mottle) where symptoms developed when temperature was >30°C almost daily, with VPD >3.0 kPa and a VPD spike of 4.5 kPa, 3 weeks post-transplant. The symptoms occurred in treatments where the growing media had become over wet, and remained rather wet throughout the trial.
- More recent observations saw symptoms develop during peaks in temperature (>35 °C) and VPD (2.4 kPa). Vents automatically closed under windy conditions, causing air temperature and humidity to rise sharply (personal communication).

Water relations

- The small plug size used during Pansy propagation makes water management difficult; root balls dry out quickly and plants are easily overwatered.
- Inconsistent plug quality, where batches are often watered to the requirements of the larger plant. Smaller plants are then overwatered and develop "water roots" and fail to thrive.
- Overwatering leads to "water roots" that have a 'glassy' appearance with few, if any, root hairs. This limits their ability to take up sufficient water and nutrients, particularly when challenged by extreme environmental conditions.
- Planting depth may be a further inconsistency which leads to variation in symptom expression within Pansy packs.
- Growing media structure can contribute to difficulties with water relations if too open or too dense, or if the growing media is changed and adjustments to water management are required.
- While such observations concerning water relations have not been linked causally with PaMS expression, over application of water may limit root development; healthy root development through careful management of water application should help to reduce plant stress.

Previous work

Previous work (PO 016 and PO 016a) included monitoring inputs and the environment (temperature, humidity, irrigation and light levels) of Pansy crops from seed sowing to marketing, with the objective of identifying the trigger(s) for symptom expression. Monitoring suggested that symptoms occur following episodes of high temperature (>35°C) combined with high vapour pressure deficit (VPD) (>4.5) and high light levels, although symptoms occurred in only two monitored batches. Evidence from subsequent studies where the environment has been monitored appears to support these results.

Vapour pressure deficit describes the drying effect of air; high VPD occurs under high temperature, low humidity conditions, where high VPD is greater than 2.0 kPa (dry air) and low VPD is less than 0.2 kPa (humid air). Most plants grow well in the middle of this range (0.5 kPa-0.95 kPa), with pansies performing well around 0.6-0.7 kPa.

It is clear that the outcomes of previous work have not defined the trigger(s) for PaMS and more work is required to further our understanding of the triggers of PaMS and enable us to develop recommendations for how growers can avoid or mitigate specific stress events that may cause symptom expression.

The purpose of the environmental monitoring work carried out in 2019 was to carry out further monitoring of Pansy crops to further our understanding of the triggers of PaMS, and to develop recommendations for the mitigation of plant stress events that may contribute to symptom expression (**WP1**).

In a second work package, two irrigation events were hosted at grower nurseries. These were designed to present techniques to quantify the water volume applied to Pansy crops, to demonstrate the impact of a number of irrigation regimes on root development. While root development has not been linked directly with PaMS symptoms, poor root development may

contribute to plant stress (and therefore PaMS development) under challenging environmental conditions.

Although this work focusses on pansies, the work will have wider benefits. Elements of this work may be expanded in future years to include other crops, and be adapted to resolve other physiological problems. Improving water relations throughout production, but particularly at the vulnerable plug stage, reviewing and updating management techniques to reduce plant stress will improve plant quality and reduce wastage.

Aims and objectives

To identify the environmental conditions that trigger the onset of Pansy Mottle Syndrome (PaMS) in-situ on a commercial nursery and to deliver the results and recommendations to industry in a practical format

WP1. Environmental monitoring

Objective 1: To monitor the environmental factors (light intensity, leaf temperature, air temperature, relative humidity and growing media moisture) in-situ on three commercial nurseries during propagation and post-transplant production phases.

WP2. Demonstration of optimisation of irrigation practices

Objective 2: To demonstrate the effect of optimum and sub-optimum irrigation regimes (up to five) on Pansy growth and development during propagation.

Methods and materials

WP1. Environmental monitoring

Site and crop production details

Environmental monitoring took place on three commercial nurseries; Nursery A (propagation and pack production), Nursery B (pack production) and Nursery C (propagation). Equipment was delivered to the nurseries in week 30 (Nursery A) and week 31 (Nursery B and Nursery C) and set-up within new batches of Pansy crops.

At Nursery A, the environment was monitored in both the propagation and pack production areas, with batches of plants moving from one area to the other. Plants were both propagated and grown-on under glass. Plants were gapped prior to marketing.

At Nursery C, approximately one week after sowing, plants were moved from the germination room into a fogging area for a further week, and monitored until dispatch to Nursery B. As plants were dispatched from Nursery C to multiple nurseries for finishing, it was decided to monitor a single Pansy cultivar that was included in all deliveries to Nursery B. Plants tended to be moved between areas on this nursery, including for gapping.

Monitored batches from Nursery C were transported to Nursery B using refrigerated lorries, with the environment monitored during transit using 2 x Tinytag data loggers (temperature and humidity). Plug plants were then transplanted and the growing environment monitored, so that plants were monitored from sowing (Nursery C) through to marketing (Nursery B).

Environmental monitoring was carried out using Tinytag loggers and 30MHz equipment, all of which were set to record at five minute intervals (**[Figure 2](#page-17-0)**). A list of the equipment used at each site is found in **[Table 2](#page-16-0)**; this was supplemented with nursery-owned 30MHz equipment to increase coverage. ADAS and nursery-owned 30MHz equipment were calibrated against each other. Moisture sensors were not used during the propagation phase at either Nursery A or Nursery C as the plug size is too small to accept the probes. Sowing, transport and transplant dates for monitored batches are detailed in **[Appendix 1](#page-48-0)**.

Site	Equipment	
Nursery A - propagation	4 x Tinytag (temperature, humidity, dew point)* 2 x 30MHz multi-sensor (temperature, humidity, leaf temperature) 1 x Light probe	
Nursery A - production	4 x Tinytag (temperature, humidity, dew point) 2 x 30MHz multi-sensor (temperature, humidity, leaf temperature) 1 x Light probe 1 x Moisture sensor	
Nursery B - production	4 x Tinytag (temperature, humidity, dew point) 2 x Tinytag (temperature, humidity, dew point) for use during transportation 2 x 30MHz multi-sensor (temperature, humidity, leaf temperature) 1 x Light probe 1 x Moisture sensor	

Table 2. Environmental monitoring equipment used at each site, 2019. * One Tinytag logger was lost on the nursery.

Figure 2. Environmental monitoring equipment: 30MHz light sensor (top left), soil moisture sensor (bottom left) and multi-sensor (centre), and Tinytag data logger (right)

Assessments

Pansy crops were monitored by growers on a weekly basis, recording Pansy mottle and distortion symptoms and the proportion of the crop affected

Data analysis process

The 30MHz data, stored in the cloud on servers belonging to Zensie, was extracted through the Zensie Application Programming Interface (API) for accessing the servers remotely, using a bespoke computer program written by ADAS. Data was extracted using a five-minute interval, to ensure consistency with the Tinytag loggers, and stored on ADAS servers.

Nursery A

For the analysis, the time series data from the Tinytag and 30MHz data loggers were converted into batch data based on the key dates during the production process. The Tinytag data was averaged across the four Tinytag loggers (in all production stages) as the logged data was consistent across the four loggers. The data for each batch was then assigned a time unit based on the date of transplanting (i.e. 5 minutes after midnight on the date of transplanting was + 1 time unit) so that all data were expressed relative to the date of transplanting. This was done as it was after transplanting that symptoms were first seen, and

using a fixed point in the production process to define the time allows comparison across batches, making it potentially easier to spot environmental triggers that occur at similar times in the production process. Due to the nature of the production process at Nursery A, the environmental monitoring data for the different batches is effectively a shift in the monitoring data by the number of days between the transplanting dates of the different batches (e.g. the monitoring data on day 15 in A_W32 is the same as day 29 in A_W30).

Nurseries B and C

As with Nursery A, the environmental monitoring data was arranged into batches, extracting the data from the appropriate sensors to provide a time series of data for each batch. Where multiple Tinytags covered the same batch for the same production stage, then the data were averaged across the number of Tinytags. The data for each batch was then assigned a time unit based on the data of transplanting so that all data were expressed relative to the date of transplanting, as it was after transplanting that symptoms were first seen.

WP2. Demonstration of optimisation of irrigation practices

Site and crop production details

Seeds of two Pansy cultivars (kept anonymous at suppliers' request) were sown into 360-cell trays (peat-based growing media) at Bordon Hill Nurseries, Warwickshire, in week 34 (21 August 2019) and placed into a temperature controlled germination room (15˚C ±1˚C) for five days. They were grown under glass until they reached cotyledon stage, then transferred to ADAS Boxworth, Cambridgeshire, in week 36 (4 September 2019) where they were placed on benches within an unheated polytunnel for the duration of the trial. Temperature and humidity were monitored throughout the trial using TinyTag data loggers (*Appendix 2*).

The trays were monitored, weighed and irrigated according to the irrigation treatments (see below) on a daily basis for three weeks. Treatments ended in week 39 (23 September 2019), when the plugs had reached 3-4 true leaves, and an assessment was completed on the plugs.

Trial design and statistical analysis

Each irrigation treatment was set-out on separate bench sections, with three replicate trays per cultivar within each treatment (six trays per treatment in total). Irrigation treatments were not replicated and therefore there was no statistical analysis.

Irrigation treatments

The irrigation treatments were based on the gravimetric method (Burgess, 2018). The gravimetric method uses the weight of water lost or taken up by the plant to calibrate the level of irrigation needed for a particular combination of plant, growing media, plug / container size and plant growth stage. This is then used to determine the 'Working Water Capacity' (WWC) required to re-wet the crop to container capacity from the 'Need to Irrigate' stage without applying excess.

The process to determine the WWC was to irrigate the containers (a sample size of at least eight pots or trays) to full capacity and allow to drain for 30 minutes. Each container was weighed after 30 minutes, and left to dry back to the stage at which irrigation was judged necessary. Once the containers reached the 'Need to Irrigate' stage they were re-weighed. The difference in weight between the container capacity and the 'Need to Irrigate' stage was the WWC.

On 5 September 2019, all trays were irrigated to full capacity, allowed to drain for 30 minutes, and then weighed. The trays were weighed again after a further 2.5 hours, and again 2 hours after that, to gain an understanding of how quickly the plug trays would dry back. The trays were then divided into the five irrigation treatments, so that there were three trays per cultivar, per treatment. Irrigation treatments were applied from 10 September 2019, once the 'Need to Irrigate' stage had been established (**[Table 5](#page-19-0)**). The amount of water applied to each tray was dependent on the weight of the tray.

Table 3. Irrigation treatment list

Details of irrigation volume and tray weights are presented in Error! Reference source not found. and summarised in **[Table 6](#page-19-1)**. As T1 was irrigated twice per day to full capacity regardless of water loss, this treatment was not weighed.

Table 4. Number of irrigation events for each irrigation treatment from 10 September 2019

Assessments

Plugs were assessed at the end of the irrigation period in week 39 for plant height (mm), plant quality (**[Table 7](#page-20-0)**) and plug root development (**[Table 8](#page-20-1)** and **[Figure 3](#page-21-2)**).

Table 5. Plant quality scores

	Score Definition
n	Dead
1	Very poor quality
2	Poor quality
3	Good quality, some damage visible
4	Very good quality, very little damage
5	Excellent quality, no damage visible

Table 6. Root development scores

Figure 3. Root scoring criteria 0-4 used in the Pansy irrigation trial 2019

Results

WP1. Environmental monitoring

PaMS was reported on all three monitored sites during 2019. Symptoms included mottle, distortion and loss of growing point.

At Nursery A (propagation and pack production), there was low incidence of PaMS in all transplant weeks, both in pot and pack throughout the season (**[Figure 4](#page-22-0)**). However, the symptoms were only observed post-transplant. Quantitative data was reported by the nursery as waste records at marketing (**[Table 9](#page-23-0)**).

At Nursery C (propagation) the worst symptoms were reported in weeks 31 - 33, and were predominately leaf mottling and distortion (**[Figure 6](#page-23-1)**). Plants with symptoms were removed during production and at marketing (**[Table 10](#page-23-2)**). Non-symptomatic plants were then transported to Nursery B.

At Nursery B (pack production) symptoms were present on arrival at the nursery (from Nursery C), and included leaf and flower mottling and distortion, and loss of growing points (**[Figure 5](#page-22-1)**). Symptom severity was greatest in weeks 33 and 34, and included mottling and distortion of leaves and flowers and loss of growing point (**[Table 10](#page-23-2)**).

Figure 4. Nursery A. PaMS symptoms, observed post-transplant, included mottling and distortion of leaves

Figure 5. Nursery B. PaMS symptoms included loss of growing point, and mottling and distortion of leaves and flowers

Figure 6. Nursery C. PaMS symptoms at plug stage included mottling and distortion of leaves

Batch no.	Propagation	Production
A W23	None	19.55%
A W25	None	21.87%
A W26	None	14.09%
A W27	None	15.24%
A W28	None	11.35%
A W30	None	19.75%
A W32	None	8.13%

Table 8. Nurseries B and C. Proportion of crop with PaMS symptoms

Nursery A

For Nursery A, the data for a single batch consisted of 20448 rows of data covering eleven different environmental variables. As the equipment was deployed at Nursery A on 25 July 2019 (week 30), it was in place in time to monitor one complete batch through the entire production process (batch A_W32). The timing of the batches through the different stages of production is shown in **Table 3.**

Symptoms were observed in all batches, and these symptoms appeared shortly after transplanting. Based on the overlap with the dates of logging of environmental data, there was therefore potential to identify correlations for a maximum of 4 batches (batches A_W27 to A_W32, depending on whether there was an environmental trigger for the symptoms and how soon before transplanting that trigger occurred.

Nurseries B and C

For Nurseries B and C, a complete set of data for a single batch consisted of 16128 rows of data covering nine different environmental variables. There were fewer environmental variables than for Nursery A, as soil moisture data was incomplete and therefore not used. The monitoring equipment was deployed at Nurseries B and C in week 31. A total of four batches were able to be completely monitored throughout the entire production process (Batches B_W34 to B_W37), due to the timing of the setting up of the logger, but unfortunately there was some missing data for batch B_W35, so a complete set of data for this batch was not available. For all of the other batches, logging was present through some, but not all of the production process. The dates of the key stages in the process are shown in **Table 4**.

Symptoms of Pansy Mottle Syndrome were seen in all batches, with symptoms being seen at both Nursery C and Nursery B. However, after the first two batches, any symptomatic plants at Nursery C were removed during the propagation process and prior to dispatch; however symptoms were apparent on arrival at Nursery B (after the transport stage).

Data analysis

Nursery A

[Figure 7](#page-26-0), **[Figure 8,](#page-27-0) [Figure 9](#page-28-0)**, **[Figure 10](#page-29-0)** and **[Figure 11](#page-30-0)** show the recorded temperature, relative humidity, vapour pressure deficit, photosynthetically active radiation (PAR) and soil volumetric water content for batches A_W28, A_W30 and A_W32. These are the batches with the most overlap between monitoring data and the dates of the production process, with all batches showing PaMS symptoms soon after transplanting.

From **Figures 7-11**, there is no clear consistent spike or dip in any of the environmental data prior to transplanting, or shortly after transplanting that could potentially be linked with the occurrence of PaMS symptoms. Previous work had hinted at high temperature, VPD and PAR as being potential triggers, but there is no consistent evidence of similar conditions in this data for these batches.

In [Figure 7](#page-26-0), there are some spikes in temperature (up to 35°C), but these only occur prior to transplanting for batch A_W32, being after transplanting in the other two batches. There are periods where temperatures exceed 30°C before transplanting in all batches, but they are not consistently around the same time from transplanting. This does not rule these high temperatures out as a potential trigger for the occurrence of PaMS symptoms as the temperatures would have caused stress to the plant and then the effects of this stress may only be seen after transplanting irrespective of when the trigger actually occurred during propagation. **[Figure 11](#page-30-0)**

Figure 11 shows some low growing media moisture content (less than 40%) but only quite late on after transplanting. This does not appear to be coincident with other potential stresses such as high temperature and PAR, but could still contribute to plant stress and the later display of PaMS symptoms.

Figure 7. Mean Temperature Data (°C) for Batches A_W28 (blue), A_W30 (green) and A_W32 (yellow) at Nursery A. Data presented for days pre- and posttransplant; "0" (x-axis) denotes the day of transplant.

Figure 8. Relative Humidity Data (%) for Batches A_W28 (blue), A_W30 (green) and A_W32 (yellow) at Nursery A. Data presented for days pre- and posttransplant; "0" (x-axis) denotes the day of transplant.

Figure 9. Vapour Pressure Deficit Data (kPa) for Batches A_W28 (blue), A_W30 (green) and A_W32 (yellow) at Nursery A. Data presented for days pre- and post-transplant; "0" (x-axis) denotes the day of transplant.

Figure 10. Cumulative Photosynthetically Active Radiation (PAR) data (mol/m²/s) for Batches A_W28 (blue), A_W30 (green) and A_W32 (yellow) at Nursery A. Data presented for days pre- and post-transplant; "0" (x-axis) denotes the day of transplant.

Figure 11. Soil volumetric water content measured at Nursery A for Batch W30. As only one sensor was used, the data covers multiple batches, but with a shift in timing.

Nurseries B and C

Data covering the complete batch up to transplanting, with no missing data, was available for three batches: B_W34, B_W36 and B_W37 and **[Figure 12](#page-33-0)**, **[Figure 13](#page-34-0)**, **[Figure 14](#page-35-0)** and **[Figure](#page-36-0) [15](#page-36-0)** show the mean temperature, relative humidity, vapour pressure deficit and photosynthetically active radiation (PAR) recorded for these batches. These batches were all listed as having symptoms present on arrival at Nursery C, but with the latter two batches having minimal symptoms relative to batch B_W34.

As with Nursery A, there do not appear to be any consistent spikes or dips in the environmental variable data that might indicate a trigger for the occurrence of PaMS symptoms. There are some high temperatures for all three batches in the period up to transport (**[Figure 12](#page-33-0)**), which would potentially have caused stress to the plants during propagation. The most noticeable thing is the temperature during transport, which shows a much lower daily variation. Given that the symptoms often appeared after the transport period, on arrival at Nursery B from Nursery C, this might be considered a potential trigger. However, Nursery C was removing and replacing symptomatic plants prior to transport, so the symptoms were potentially being triggered earlier, during propagation, but the transport may just have acted as an additional stress that caused greater visibility of symptoms.

In **[Figure 13](#page-34-0)**, there are some low relative humidities (<30%) during propagation for batches B W34 and B W37, but these do not occur for batch B W36, Also, batches B W34 and B_W36 experience some higher vapour pressure deficits (**[Figure 14](#page-35-0)**) during propagation. These are events that could have led to plant stress, but they are not consistently apparent across all batches, and the information from the production suggests that all batches experienced some symptoms, and that these were potentially occurring during propagation (with symptomatic plants being replaced).

Given that the information provided by the Nurseries suggested that symptoms became less prevalent as the season progressed, there is little in the data to suggest a similar pattern in the environmental data (e.g. higher temperatures during propagation earlier in the season). The pattern of higher environmental parameter values for the earlier season batches can be seen in the temperature data approximately 3.5 days prior to transplanting, where the temperatures of the early batches are higher than those in later batches [\(Figure 16\)](#page-37-0), and this continues during the transportation of the batches to Nursery B. This may explain why the plant consistently showed symptoms on arrival at Nursery B, after having no symptomatic plants at point of dispatch from Nursery C. However, it does not necessarily help to explain why symptoms were found during propagation on Nursery C, and without information on the date at which symptoms were seen on Nursery C and the symptomatic plants removed, this environmental monitoring data cannot be definitively said to pre-date the occurrence of the symptoms during propagation. If the occurrence of symptoms after transportation was due to conditions during transportation, this does not explain why symptoms were seen at Nursery A, where there was no transportation of the plants prior to transplanting, and the environmental conditions during propagation do not mirror the conditions during transportation.

Figure 12. Temperature data (°C) for nurseries B and C (Batches B_W34 (silver), B_W36 (blue) and B_W37 (green)). Data presented for days pre- and posttransplant; "0" (x-axis) denotes the day of transplant

Figure 13. Relative humidity data (%) for Nurseries B and C (Batches B_W34 (silver), B_W36 (blue) and B_W37 (green)). Data presented for days pre- and post-transplant; "0" (x-axis) denotes the day of transplant

Figure 14. Vapour pressure deficit data (kPa) for Nurseries B and C (Batches B_W34 (silver), B_W36 (blue) and B_W37 (green)). Data presented for days preand post-transplant; "0" (x-axis) denotes the day of transplant

Figure 15. Cumulative Photosynthetically Active Radiation PAR) data (mol/m2/s) for Nurseries B and C. (Batches B_W34 (silver), B_W36 (blue) and B_W37 (green)). Data presented for days pre- and post-transplant; "0" (x-axis) denotes the day of transplant.

Figure 16. Mean temperature data (°C) for all batches at approximately 3.5 Days prior to transplanting. Red circles indicate where early batches (B_W32 (dark blue), B_W33 (orange)) experienced higher temperatures than later batches (B_W34 (grey), B_W35 (yellow), B_W36 (light blue) & B_W37 (green))

WP2. Demonstration of optimisation of irrigation practices

'Working Water Capacity' (WWC)

The average tray weight at full capacity was 1400 g, and the average 'Need to irrigate' weight was 700 g. Therefore, the 'Working Water Capacity' was 700 g. Treatments are presented in **[Table 5](#page-19-0)**.

For treatment T1 'Extreme Wet', water was applied twice per day until field capacity was reached, and the weight of water applied was not recorded. The 'Little and Often' (T3) treatment required many more applications than all other treatments except for (T1). However, there was little difference in the total weight of water applied to treatments T2 (Extreme Dry) and T3 (Little and Often) (**[Table 11](#page-38-1)**); treatment T2 allowed the growing media to dry back between applications. Treatment T5 'Long Dry Down' resulted in less water being applied over the course of the trial than any other treatment, with fewer applications made.

Table 9. Total water weight applied to each treatment, and number of applications from 10 September 2019.

Plant height

When plants were measured in week 39, there were clear height differences in cultivar A, with plants grown in the 'Extreme Wet' regime (T1; 22.1 mm) larger than the other treatments (**[Figure 17](#page-39-0)**). There was little difference in plant height between the remaining treatments (T2 – T5), with the shortest plants produced in the 'Extreme Dry' (T2) treatment (19.0 mm).

With cultivar B, plant height ranged from 17.9 mm in the 'Long Dry Down' (T5) treatment to 21.9 mm in the 'Little and Often' (T3) treatment. Plants grown in the 'Extreme Wet' (T1) regime measured 19.2 mm on average.

Figure 17. Plant height measured at the end of the irrigation treatments, week 39 2019

Plant quality

For both cultivars, plant quality was better in the 'Extreme Wet' (T1) treatment, with plants scoring 4.0; very good quality, very little damage. These plants were darker green and were also developing into larger plug plants. All other treatments scored 3.0; good quality, some damage visible, for both cultivars. These plants were slightly paler, with some crinkling to the leaves, more so in the 'Extreme Dry' (T2) treatment. There was no evidence of PaMS in any of the treatments for either cultivar throughout the trial period. Examples of plants grown in each irrigation treatment can be seen in **[Figure 18](#page-40-0)**.

T1 'Extreme Wet' T2 'Extreme Dry'

T3 'Little and Often' T4 'Matched to Water Loss'

T5 'Long Dry Down'

Figure 18. Example of Pansy plugs grown under each irrigation regime, week 39, 2019

Root development

There were clear differences in root development for both cultivars, with plugs grown under the 'Extreme Wet' (T1) regime showing poorer root development (**[Figure 19](#page-41-0)**). There were also more "water roots" present in treatment (T1) compared with the other treatments (**[Figure 20](#page-42-2)**). Root development was improved in the 'Extreme Dry' (T2) treatment, with no "water roots" and more root hairs. Plants in the 'Little and Often' (T3) treatment had a mix of roots with root hairs and "water roots", while those in the 'Matched to Water Loss' (T4) treatment produced many root hairs, with only a few "water roots". Plants in the 'Long Dry Down' (T5) treatment produced roots throughout the plug, with many root hairs.

Figure 19. Root development assessed at the end of the irrigation treatments, week 39 2019

T1 'Extreme Wet' T2 'Extreme Dry'

T3 'Little and Often' T4 'Matched to Water Loss'

T5 'Long Dry Down'

Figure 20. Example of root development in Pansy plugs grown under each irrigation regime, week 39 2019

Discussion

WP1. Environmental monitoring

The intention was to use a range of statistical techniques for the data analysis, but these could not be used due to inconsistent / incomplete environmental data, and differences between the nursery production systems that meant that the data from the different nurseries couldn't be compared.

Due to the fact that symptomatic plants were removed and replaced during propagation in one set of data, it is not statistically valid to combine the datasets for analysis. In addition, even if the data could have been combined across the nurseries, there would still have been only four datasets where the plants were followed throughout the entire production cycle, and no more than ten datasets that could have been used to analyse conditions from sowing up to transplanting.

Symptoms tend to become apparent within crops over a period of days. The course of symptom development appears to be that one or two plants may be affected initially but symptoms are expressed in more plants, and more fully, over the course of at least 2-3 days. This can make it difficult to identify the date of first symptoms in a large batch of pansies. In the scenario of nurseries B and C, where plug plants are produced by a young plant producer and then distributed to finishing nurseries, symptoms may be triggered at the propagation

nursery, where any plants with visible symptoms are removed from the batch prior to dispatch, and more symptoms are present on arrival at the finishing nursery.

It was not possible for the nurseries to provide precise dates of first occurrence of symptoms for the various batches, or quantitative data on the prevalence of different symptom types, which means that it is not possible to identify statistical correlations between environmental variables and distinct PaMS symptoms. The analysis completed, as detailed, identifies any anomalous readings that could have stressed the plants and therefore may (or may not) be linked to PaMS symptom expression. This would not have been sufficient to allow more detailed statistical tests to be used given the uncertainty about causality and types of symptoms for PaMS.

The precise cause of symptoms is not known, and there is no differentiation between different symptoms (e.g. mottling, leaf distortion, stunting) in the data, therefore it is only possible to identify potential plant stresses that may or may not cause the symptoms to arise. In addition, the extent of any delay between triggers (if they exist) and displaying of symptoms is also not known and it is therefore possible that the display of symptoms could be due to an accumulation of stresses over a long period of time, or conversely triggered by a single event. Further knowledge or larger datasets would allow more detailed statistical analysis to be carried out than the current data allows. For example, if data on at least 20 batches could be collected, along with the prevalence of symptoms at one or more key points in the production process (e.g. transplanting) then it should be possible to use a window pane analysis to identify potential correlations between environmental variables and the prevalence of symptoms.

WP2. Demonstration of optimisation of irrigation practices

There were clear differences between treatments, with effects noticeable both in terms of plant growth and root development. There were no signs of PaMS developing in the plug tray throughout the irrigation trial, likely as a result of the moderate prevailing environmental conditions.

Plants of both cultivars achieved the highest plant quality scores in the 'Extreme Wet' (T1) treatment. They were darker green and were also developing into a larger plug plant. For cultivar A, this treatment also produced the tallest plug plants. However, root development was poorer for both cultivars in this irrigation treatment, with rooting in 26-50% of the plug on average, and these roots produced fewer root hairs, with many more "water roots" present. While the top growth of the plants in this treatment appeared strong, the smaller proportion of roots present with root hairs would limit the plant's capacity to take up water under drier conditions.

In the 'Extreme Dry' (T2) treatment, plants were paler, with some crinkling to the leaves where they had been too dry. This treatment also produced the smallest plants in cultivar A. However, they achieved higher root quality scores, with rooting in up to 75% of the plug, on average. There were no "water roots" in this treatment, and many more plants with root hairs.

The 'Little and Often' (T3) approach gave generally good quality plants, although slightly pale, and taller in this treatment. Root development was reasonable in this treatment, but with a mix of "water roots" and root hairs, suggesting that they could struggle to take up sufficient water in dry conditions. Fewer "water roots" may have developed had the growing media been allowed to dry back further before water was applied. This could be a useful regime with slight adjustments to the parameter for applying water (in this demonstration < or near to 30% of WWC) and / or the weight of water applied.

The 'Matched to Water Loss' (T4) treatment produced good quality plug plants although slightly smaller than in other treatments. Root development was very good in this treatment, with rooting in over 75% of the plug on average, few "water roots" and many root hairs. This appeared to be a successful irrigation regime, resulting in both good quality plants with welldeveloped roots.

The final treatment, 'Long Dry Down' (T5), was similar to the 'Extreme Dry' (T2) treatment. There was no leaf crinkling in this treatment, although plant height was reduced, particularly for cultivar B. Again, root development was respectable, with roots throughout the plug, and plenty of root hairs. However, for plug production this treatment may be insufficiently forgiving, with little margin for error.

The treatments appear to have impacted on root development in two ways:

- **"Water root" development**. Allowing the growing media to dry back further between water applications, as in the 'Extreme Dry' (T2) and 'Long Dry Down' (T5) treatments which received five and four water applications respectively appears to have prevented "water roots" from developing (**[Table 11](#page-38-1)**). Plants in the 'Little and Often' (T3) treatment and 'Matched to Water Loss' (T4) treatments received 17 and 7 water applications respectively; the growing media dried back more between applications in treatment T4 than T3 and again, fewer "water roots" were produced.
- **Water quantity**. A greater volume of water was applied to plants in the 'Extreme Dry' (T2) and 'Little and Often' (T3) treatments overall compared with the 'Matched to Water Loss' (T4) and 'Long Dry Down' (T5) treatments. The highest root quality score was achieved by

T4 in terms of root spread through the plug for both cultivars. This suggests that it isn't the volume of water *per se* that is critical to good root development, rather it is the period of time allowed for the growing media to dry back between applications. However, during cool conditions, where large water volumes are applied, it will take longer for the growing media to dry back, risking water root development.

Conclusions

WP1. Environmental monitoring

The environmental monitoring carried out in 2019 did not identify triggers for PaMS. Previous work had suggested that, high temperature, VPD and PAR could be potential triggers, but this was not borne out by the data for the batches of Pansies monitored in 2019, when symptom occurrence could not be correlated with such environmental events. It is not clear if the symptoms that are considered to be part of the PaMS complex (mottling, distortion, lost growing points) are caused by a single trigger, different triggers or cumulative triggers. More detailed recording of symptoms including the precise date and time of first symptom, and the proportion of each symptom expressed (mottling, distortion and lost growing point) would enable these distinctions to be statistically analysed.

Suggested Future Work

Given that the loggers were deployed on the nurseries relatively late in the season, it would seem sensible to undertake further monitoring during the next full growing season, ideally from mid-June. The ideal scenario from a statistical perspective would be to monitor as many batches as possible, with a consistent monitoring approach used across all sites, supported by robust monitoring of symptoms, indicating not only occurrence, but also prevalence of different symptom types. Ideally there should not be any major difference in production methods between the nurseries in relation to replacement of plants with symptoms.

We would suggest that a monitoring protocol is drawn up that attempts to ensure the following:

- All batches are monitored from sowing through to production using environmental logging for the following: temperature, relative humidity, vapour pressure deficit, photosynthetically active radiation, soil volumetric water content (This could be provided as weight of tray prior to watering if sensors not available).
- The date and time when batches are moved between locations (both within and between nurseries) is provided.
- First symptom date (and time) are provided for: mottling, leaf distortion and other symptoms (to be specified).
- The prevalence of the symptoms (% plants affected within batch) is provided on a daily basis for: mottling, leaf distortion, lost growing points.
- Where symptomatic plants are replaced, the number of plants replaced each day is recorded along with the reason for replacement (e.g. mottling, leaf distortion, etc.).

WP2. Demonstration of optimisation of irrigation practices

For plug production, the aim is to achieve a balance between providing sufficient water to maintain growth while producing plants with well-developed roots; a difficult balance to achieve for small plugs. Plants develop stronger root systems when they are not overly wet, and are forced to search for water and nutrients.

The key factor for the success of any irrigation regime determined using the gravimetric method is correct judgement of when the 'Need to Irrigate' point has been reached. If it's judged that plugs need to be irrigated before they have dried back sufficiently, the growing media may always be too wet, particularly when using 'Little and Often' and 'Match to Water Loss' regimes. The 'Need to Irrigate' point will vary depending on plug size, growing media formulation, plant species and prevailing temperature; in-house trials would help to establish the parameters for when to irrigate.

Where there is an abundance of "water roots", plants struggle to take up water as moisture levels reduce and would be less able to respond to increased demand for water and nutrients under high temperature, vapour pressure deficit (VPD) or light conditions. However, where "water roots" are present, if the growing media was allowed to dry back, the plants would produce new roots and develop root hairs, in response to their search for water and nutrients, producing plants more resilient to extreme changes in environment post-transplant.

For the most part, treatments T2-T5 may all be suitable for plug production, but with some adjustments to allow the growing media to dry back sufficiently between irrigation applications to minimise the development of "water roots". Consideration should also be given to the practicalities of the various irrigation regimes, for example while the number of irrigation events undertaken for the 'Little and Often' (T3) treatment may be easily managed in nurseries with boom irrigation, they may be less practical where crops are hand irrigated.

Irrigation of plants at plug stage is difficult to monitor closely as moisture probes are too large for the cell size, particularly those used in Pansy production. However, environmental monitoring systems that include wireless scales to measure plug tray weight that will help to automate the process are being developed. Use of gravimetric techniques to determine when to irrigate linked to manually lifting trays is a useful aid to setting irrigation parameters and training staff to irrigate to the correct level for healthy root development.

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References

Burgess, C. (2018) Methods and equipment for matching irrigation supply to demand in container-grown crops. AHDB Factsheet 18/17. Revision of Factsheet 19/05.

Appendix 1. Sowing, transport and transplant dates

Table 10. Nursery A. Sowing, transport and transplant dates (week commencing) for all of the monitored batches. * denotes processes undertaken prior or post deployment of the monitoring equipment.

Table 11. Nurseries B and C. Sowing, transport and transplant dates (week commencing) for all of the monitored batches. * denotes processes undertaken prior or post deployment of the monitoring equipment.

Appendix 2. Tray weights and irrigation amounts

T2 'Extreme Dry'

Appendix 3. Polytunnel temperature and humidity

