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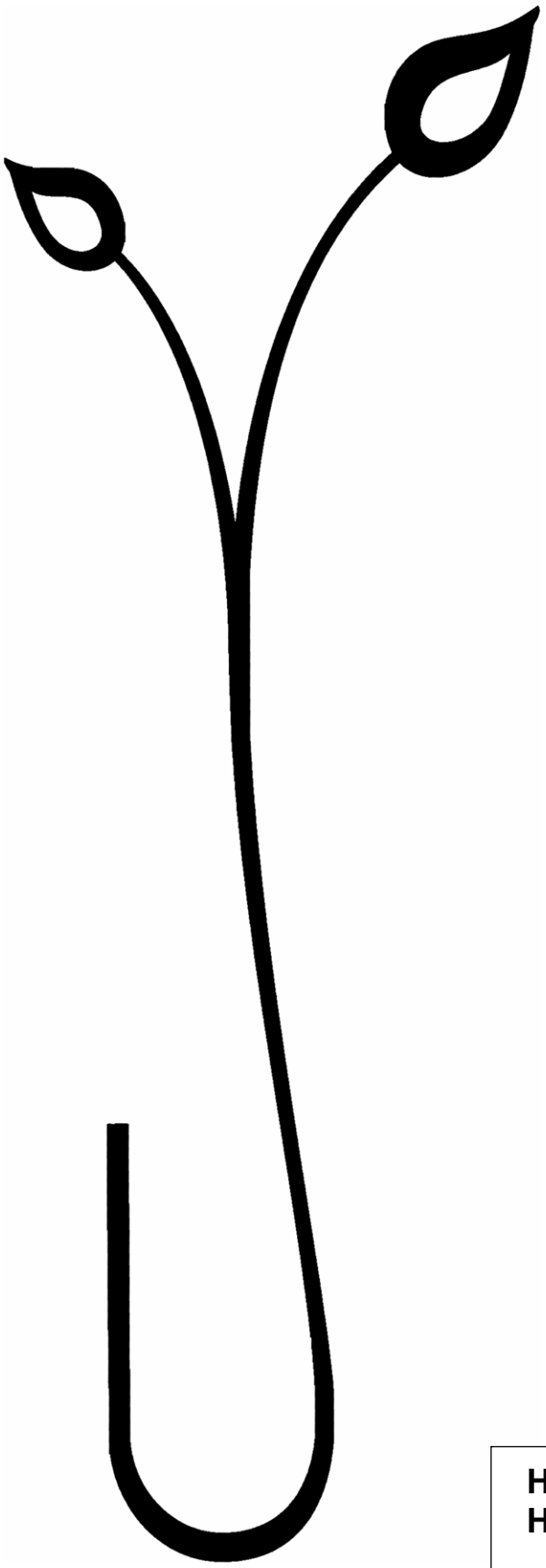

Research
Report

HortLINK project HL0178
HDC Project BOF 63 & CP 36

Integrated control of bulb-scale mite in narcissus

Final Report 2011

Project title:	Integrated control of bulb-scale mite in narcissus
Project number:	BOF 63, BOF 63a, HL0178 (incl. CP 36)
Project leader:	Rosemary Collier Warwick Crop Centre, School of Life Sciences, Wellesbourne Campus, University of Warwick, Wellesbourne, Warwick CV35 9EF
Report:	Final report, August 2011
Previous report:	Annual reports 2007, 2008, 2009
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Location of project:	Warwick Crop Centre, School of Life Sciences, Wellesbourne Campus, University of Warwick, Wellesbourne, Warwick CV35 9EF
Project co-ordinators:	Adrian Jansen
Date project commenced:	1 October 2006
Date project completed:	31 July 2011
Key words:	Bulb-scale mite, narcissus, acaricide, hot-water treatment, warm storage, epidemiology, biological control, integrated pest management, bulb mite

The results and conclusions in this report are based on a series of experiments conducted over a one-year period. The conditions under which the experiments were carried out and the results have been reported in detail and with accuracy. However, because of the biological nature of the work it must be borne in mind that different circumstances and conditions could produce different results. Therefore, care must be taken with interpretation of the results, especially if they are used as the basis for commercial product recommendations.

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

Dr R Collier
Director, Warwick Crop Centre
School of Life Sciences, University of Warwick

Signature ..... Date .....

Report authorised by:

Professor B Thomas
Deputy Head
School of Life Sciences, University of Warwick

Signature ..... Date .....

GROWER SUMMARY

SCIENCE SECTION

Grower Summary

Headline

The project showed that hot-water treatment (HWT) and good hygiene remain key elements in the integrated control of the bulb-scale mite in daffodils. Some methods suggested as techniques for mite control – such as heat-treatment or frosting – appeared to be ineffective, but there were promising results from a trial in which the predatory mite *Amblyseius barkeri* appeared to be effective in killing bulb-scale mites. None of the acaricides tested produced a statistically-significant level of mite control.

Background and expected deliverables

The UK is the world-leader in production of narcissus. Some 4,300 ha of narcissus are field-grown, producing an annual saleable output of about 30,000 tonnes of bulbs and 600 million cut-flowers, of which perhaps 30% of bulbs and 40% of flowers are exported. To be cost-effective, narcissus production has become very intensive, which has increased problems with pest and disease. Of the three major narcissus pests, large narcissus fly, stem nematode and bulb-scale mite, bulb-scale mite has received least attention.

Bulb-scale mite was once regarded as a sporadic pest, but for the last 10 years growers have called for more effective control measures. While bulb-scale mite symptoms rarely cause concern in field-grown bulbs or in storage, the higher temperatures in glasshouses, where bulbs are forced for cut-flowers and grown as pot-plants favour rapid multiplication of the pest, resulting in seriously damaged, distorted leaves and stems, with rejections and serious losses to producers. With the loss of endosulfan there is no approved acaricide to control bulb-scale mite in glasshouse narcissus. Other than HWT, there is no non-chemical means of control. Despite the rigorous use of HWT on bulb stocks before planting, bulb-scale mite problems continue to increase. No acaricide suitable for field application has yet been identified. Standard HWT cannot be used to treat bulbs intended for sale or forcing, since it causes leaf and flower distortion in the year after treatment. The aim of this project was to develop an integrated control strategy for bulb-scale mite based on an understanding of its biology and ecology.

Summary of the project and main conclusions

Objective 1: Define the relationship between temperature and bulb-scale mite development

The original focus of this objective was to monitor development of individuals or populations of bulb-scale mite under different environmental conditions *in vitro.* However, the work was re-aligned at the end of 2008 because the cycling of populations under natural conditions was not fully understood. Some effort had already been devoted to monitoring populations in potted bulbs maintained outdoors, and this effort was transferred to monitoring a stock of Dutch Master, infested with bulb-scale mite, which was planted at Wellesbourne in summer 2008. Samples of bulbs were taken at monthly intervals over a period of two years. The infestation was very high initially, and then declined through winter/spring 2009 before increasing to a small peak in June. This was followed by a decline until March 2010, when numbers increased again, peaking in early July. Adult and immature mites and mite eggs were found inside the bulb, and on the foliage when it was present. However, the neck of the bulb was the favoured location in the summer period. Mites were distributed very unevenly throughout the crop and, for example, in one sample of bulbs taken in November 2008, mite numbers ranged from 5-850 and egg numbers from 0-600 per bulb. The uneven distribution of mites, together with the considerable amount of time required to dissect a single bulb to count them, were two of the major challenges when undertaking the experimental work.

Objective 2: Discover when, where and how bulb-scale mite originates and spreads in field crops and in bulb storage

The use of planting troughs to examine the spread of bulb-scale mites between foliage over distance has indicated that mites may move between bulbs where their leaves touch or where the plant to plant separation distance is 0.5m or less. A field trial to follow this up indicated that movement between bulbs does not occur rapidly.

All of the commercial narcissus fields sampled in south-western and eastern England in 2007 and 2008 and in the south-west in 2009 showed damage due to the presence of bulb-scale mites, although some were at low levels of infestation. In general, infestations were greater in crops from the south-west. There was no evidence that damage was greater or less at the edges of the fields than towards the centre, but there were patches of infestation

in the field. At all sites, *along* rows, high numbers in one sample were correlated with high numbers in adjacent samples (1 m apart) and vice versa. For >1m separations, results were more variable. Results were less clear *between* rows, although there was some evidence of correlation between adjacent rows. This may be a reflection of the way that the bulbs are stored and then planted, or it may indicate the distance over which mites normally disperse from an infested bulb. As described above, leaf 'bridges' may be effective mite routes, but the mites may also move between adjacent bulbs. There is no evidence to date that weeds are a source of bulb-scale mites, but naturalised narcissus (e.g. in field margins or dumps) are a potential source of bulb-scale mite and bulb mite.

The distribution of mite-infested bulbs within crops suggested that infestations might be arising because growers were inadvertently planting infested bulbs, rather than crops being invaded from other sources. This might either be because some mites were surviving HWT, or because hot-water treated bulbs became re-infested with mites prior to planting. To determine whether mite infestations can arise from the exposure of dry bulbs to dust and debris during handling and storage, dust/debris samples were collected from the premises of bulb growers and merchants. Un-infested bulbs were stored in paper bags with this material, which was then tipped into the pots in which the bulbs were subsequently planted. Although some of the symptoms identified in the first year of sampling suggested that mites might have been transferred in this way, more detailed examinations in the second year failed to reveal any mites.

To investigate further whether mites were surviving HWT and (or) bulbs were being re-infested after HWT, growers were asked to provide samples of bulbs taken, first, immediately after HWT, and, secondly, from the same stocks on the date they were planted following intervening storage. These bulbs were dissected to record the numbers of mites and eggs. Some of the samples, both immediately after HWT and at planting, contained dead bulb-scale mites, sometimes in large numbers, but a few also contained live bulb-scale mites. Some bulbs also contained mite eggs, although it was impossible to determine whether these were alive or dead. These findings suggest that either the recommended treatment time and temperature are insufficient to kill all mites or that the temperature varies within treatment tanks. This has lead to some further HDC-funded work (BOF 63b) to determine more precisely the temperatures to which bulbs are exposed during typical commercial HWT.

Objective 3: Design optimal high or low temperature and/or chemical treatments to control bulb-scale mite in bulbs for replanting and for forcing, and ensure all stages in its life-history are killed and that crop quality is unaffected

Current and potential methods of mite control were investigated. These included the storage of dry bulbs at temperatures of 42, 44 and 46°C for periods of 1, 2 or 3 hours (warm storage), exposure of bulbs to low temperatures (-2°C) for up to 72 hours ('frosting'), novel HWT additives and a range of acaricides applied as foliar sprays. Neither warm storage nor 'frosting' were effective ways of controlling bulb-scale mite and a few mites also survived HWT, including HWT with the new additives (FAM 30 with or without Bravo 500). None of the acaricides tested produced a statistically-significant level of mite control.

Objective 4 Design novel biological control strategies and test these as part of an integrated management strategy in commercial crops.

Very small scale tests with biological control agents (predators) suggested that the predatory mite *Amblyseius barkeri* had killed a large proportion of the bulb-scale mites in the infested bulbs with which they were confined. This supports the findings of an earlier Dutch study which indicated that this species might control bulb-scale mite on *Hippeastrum.* This species of predator appeared to be more effective than the three other species that were tried.

Objective 5 Examine the link between bulb-scale mite and smoulder disease.

Experiments in which the inoculation of mite-infested and un-infested bulbs with leaves with active smoulder (*Botrytis narcissicola*) lesions, failed to result in infection with the fungal pathogen, and hence no links between bulb-scale mites and smoulder infection could be demonstrated. However, from the extent of feeding damage caused by bulb-scale mites, and seen on the young growing bulb-scales, it seems very likely that internal fungal infections could be spread to the shoots as they grow out of the neck of the bulb.

Objective 6 Deliver a prototype, improved programme for bulb-scale mite control.

Best practice has been examined and the following are important practices in managing bulb-scale mite in daffodils:

- ► HWT, carried out according to current advice regarding the duration and temperature of the dip (though this does not seem to be fully effective, any surviving mites are generally small in number).
- ► Hygiene, especially the removal of possible sources of re-contamination via dust and debris.

Objective 7 Communicate with the industry

The project and its findings have been presented at a number of grower meetings and in *HDC News*.

Financial benefits

Effective control of bulb-scale mite in forced narcissus would reduce losses by an estimated 15 to 20%. It is expected that the recommendations would be taken up by the bulk of the industry, with a farm-gate value of £2.9m to £3.9m annually. Bulb-scale mite is a prohibited pest for bulbs exported to the USA, with a zero tolerance. Effective control should increase the volume of bulbs eligible for the US market by 7.5 to 10% as the findings are taken up by all growers exporting to the US (these represent about 30% of the UK acreage) – a potential increase of sales by up to £0.5m. However, increasing awareness of bulb-scale mite may lead to stricter tolerances for exports to other countries.

Action points for growers

- Growers should be aware that the debris accumulating in bulb handling and storage facilities may provide a source of mite infestation and that they should implement appropriate hygiene measures, by keeping these areas as free of dust and debris as possible.
- The recommended HWT procedures should be carefully followed.
- Any mite-controlling effects of dry heat or frosting treatments are unreliable.
- Growers should remove volunteer narcissus growing in field margins or close to newly planted crops.

Science Section

Introduction

Steneotarsonemus laticeps (Halbert), the bulb-scale mite, is a pest of many economically important bulb species. In narcissus, feeding mites produce leaves with serrated margins, a degraded waxy surface resulting in bright green leaves and yellow flecking, and severe infestations can result in significant loss of the foliage and damaged flowers, or no flowers at all. Additionally, feeding damage promotes secondary infections, such as attack by the fungal pathogen smoulder (*Botrytis narcissicola* Kleb.), leading to further crop loss. Furthermore, for bulbs for export there may be little to no tolerance of bulb-scale mite infestations. There have been relatively few studies on the bulb-scale mite, as until recently it was considered to be only a sporadic pest. However, it has become increasingly troublesome, although there is little understanding of the reasons behind it.

The aim of this project was to develop an integrated control strategy for bulb-scale mite based on an improved understanding of its biology and ecology.

The project objectives are as follows:

1. Define the relationship between temperature and bulb-scale mite development.
2. Discover when, where and how bulb-scale mite originates and spreads in field crops and in bulb storage.
3. Design optimal high or low temperature and/or chemical treatments to control bulb-scale mite in bulbs for replanting and for forcing, and ensure all stages in its life-history are killed and that crop quality is unaffected.
4. Design novel biological control strategies and test these as part of an integrated management strategy in commercial crops.
5. Examine the link between bulb-scale mite and smoulder disease.
6. Deliver a prototype, improved programme for bulb-scale mite control.
7. Communicate with the industry.

The project milestones were as follows:

Year 1			
1.1	1 Apr 07	6	Initial literature review and grower consultation completed.
1.2	1 Apr 07	6	Field cultures of bulb-scale mite established.
2.1	1 Oct 07	12	Trapping and direct sampling methods developed and review of methods with HortLink PMC
4.1	1 Oct 07	12	Biological and microbial control methods reviewed
Year 2			
3.1	1 Apr 08	18	Effect of low temperatures determined
1.3	1 Oct 08	24	Relationships between environmental conditions and development defined.
2.2	1 Oct 08	24	Bulb distribution and movement in stores investigated
Year 3			
4.2	1 Oct 09	36	Small scale trials to evaluate biological control methods
2.3	1 Oct 09	36	Experiments on dispersal in field completed
4.3	1 Oct 09	36	Effects of acaricides determined
6.1	1 Oct 09	36	Initial commercial evaluation of alternative control techniques
Year 4			
3.2	1 Apr 10	42	HWT and (or) warm storage treatments defined
5.1	1 Jul 10	45	Link with smoulder disease investigated
7.1	1 Jul 10	45	Draft Fact sheet prepared
6.2	1 Oct 10	48	Integrated control strategies tested

Experimental

Preliminary work

Current knowledge of the bulb-scale mite and its management was reviewed earlier as part of this project. Grower members of the consortium from Cornwall and eastern England were interviewed by the PhD student working on the project for her to gain further insight into husbandry practices, current control strategies and theories on the origins of bulb-scale mite infestations. Reports documenting the discussions were compiled for her personal use.

Techniques for dissecting bulbs to assess mite numbers and damage, and for identifying infested bulb stocks, were developed. These involve either making one or more cuts across the bulb to count the number of brown feeding marks or dissecting the whole bulb, or part of the bulb, scale by scale and examining the scales under the microscope to record feeding marks and the numbers of mites and mite eggs. Historically, feeding marks have been used to assess the presence of mite infestations in bulb stocks. However, the data collected indicated that feeding marks in the bulb-scales are not always correlated with the presence of mites: it appeared that darker feeding marks were associated with old infestations and that mites were usually found on scales with only pale marks. Assessing mite infestations by feeding marks is probably an inaccurate measure of the size of current infestations and therefore of the impact of treatments in the short term. Later assessments of treatments, where an estimate of the 'short-term' impact was required, were made either by dissecting whole bulbs and recording the numbers of mites or by sampling bulb shoots i.e. by removing and examining a 10 cm-long length of shoot taken from the tip of the bulb upwards.

Objective 1: Define the relationship between temperature and bulb-scale mite development

Greenhouse and field cultures of bulb-scale mite were established to act as a source of experimental material. Figure 1.1 shows some of the mite-infested bulbs in a glasshouse at the Kirton Research Centre. Subsequently, a well-infested stock of cv Dutch Master was obtained and this was planted in an experimental area (Long Meadow Centre) at Wellesbourne (Figure 1.2).

Figure 1.1: Mite-infested bulbs in a glasshouse at the Kirton Research Centre in February 2008

Figure 1.2: Mite-infested bulbs cv Dutch Master at Wellesbourne

Sampling challenges

It was found that bulb-scale mites were unevenly distributed between plants. For example, Figure 1.3 shows the numbers of mites per bulb for two sets of 10 bulbs sampled in November and December 2008, respectively, from the infested stock of cv Dutch Master. Similarly Figure 1.4 shows the numbers of mites per shoot for 185 bulbs taken from the same stock in summer 2009 and assessed in May-June 2011 (as part of Objective 2). The mean number of bulb-scale mites per shoot was 10.5, but 80 of the shoots had no bulb-scale mites at all. The most infested shoot had 91.

Sampling any part of the bulb for bulb-scale mites is time-consuming, an average rate of 11 shoots or 5-10 bulbs per day being achieved with practice. Therefore, sample size has to be small and the estimated mean infestation level can then be very variable. With pests such as caterpillars, where it is possible to make a count prior to treatment, this is not necessarily a problem, but with bulb-scale mite this would be impossible as sampling has to be destructive. Therefore, many of the trials are based on smaller and more variable sets of data than is desirable for statistical analysis.

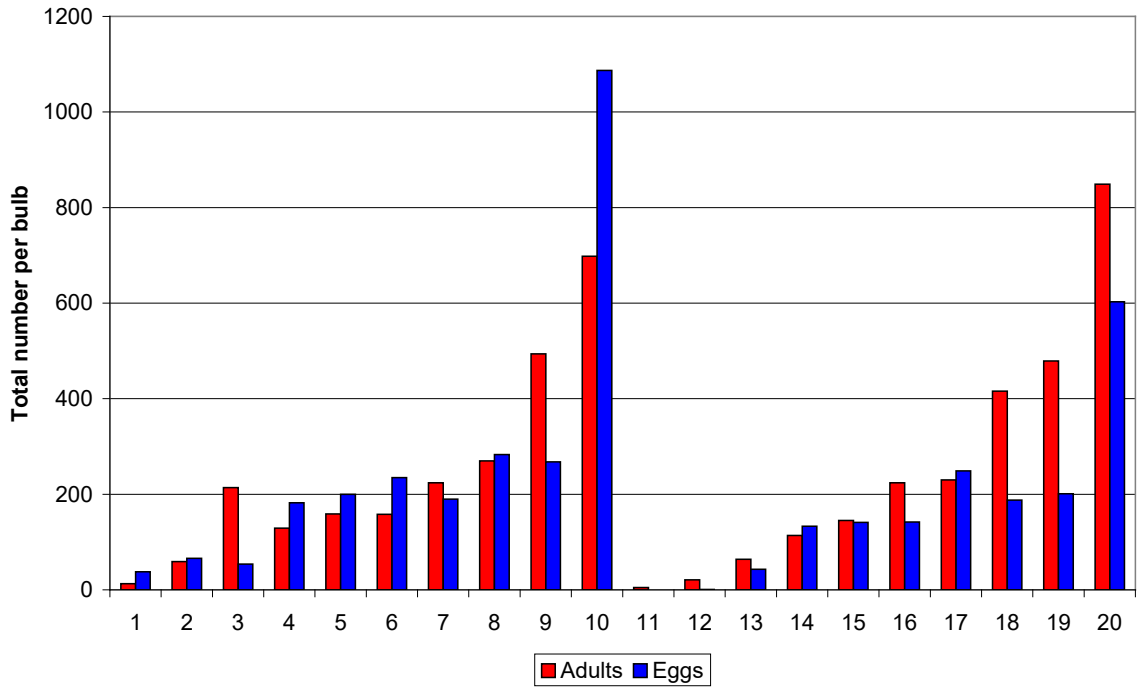


Figure 1.3: Total number of bulb-scale mites and eggs per bulb from 10 bulbs sampled on each of two occasions (1-10 in November 2008 and 11-20 in December 2008) from infested stock of cv Dutch Master at Wellesbourne

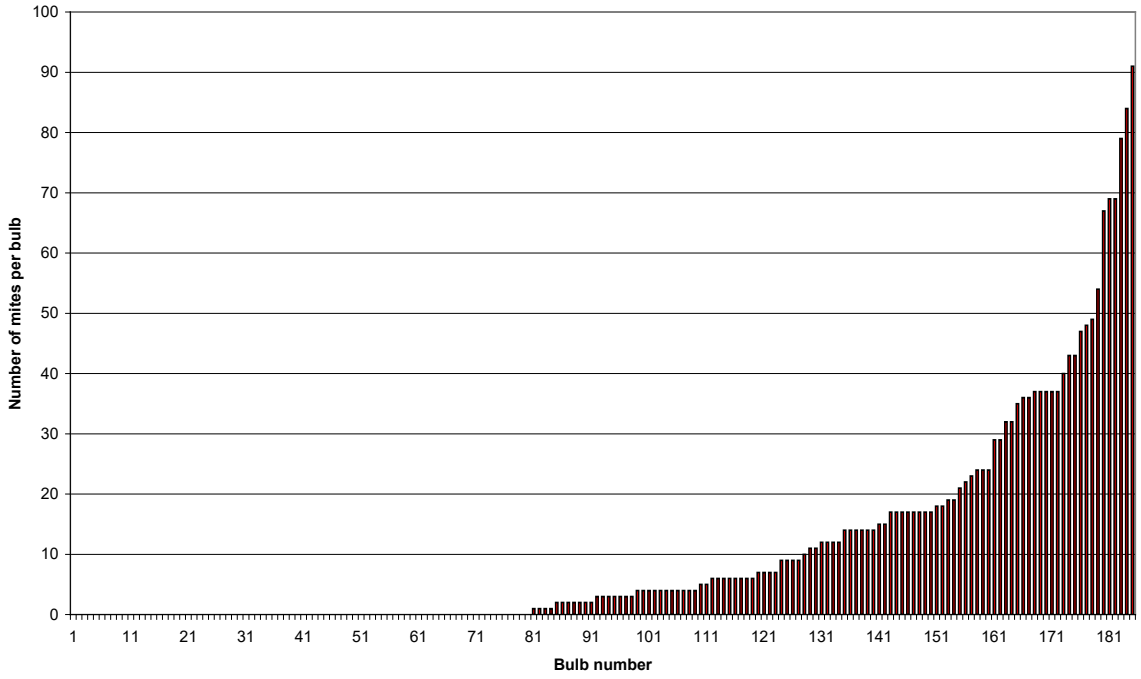


Figure 1.4: Total number of adult mites per bulb shoot – 185 bulbs from infested stock of cv Dutch Master at Wellesbourne, sampled May-June 2011

Laboratory rearing technique for bulb-scale mite

A key early objective was to design and test equipment to contain individuals or populations of bulb-scale mite for *in vitro* rearing purposes. Previous work by the Central Science Laboratory (CSL) (now Food and Environment Research Agency – FERA) developed a preliminary *in vitro* rearing cell, which incorporated live *Narcissus* bulb-scale (Lynch & Bedi, 1994). This rearing cell consisted of two glass microscope slides sandwiching an identically sized, but thicker, black Perspex slide, with a hole drilled in the middle centred over a piece of bulb-scale, the edges of which were sealed with paraffin wax and sealed to the Perspex slide. This cell was replicated and tested, but was found to be unsuccessful, with mites escaping. Therefore the development of a modified form of the CSL apparatus was attempted.

A variety of materials was used to create small cells, with circular holes, creating a sealed arena around a piece of fresh bulb-scale which forms the base of the cell. Glass, perspex and Darvic™ PVC sheet (Weston Hyde Products Ltd., UK) were used in the trials, with sealants for the bulb-scale of paraffin wax (M&B Laboratory Chemicals Ltd., UK), fluoropolymer (Whitford Ltd., UK), Blu-tack™ (Bostik Ltd., UK) and rubber O-rings (RS, UK). Fine-mesh floored cells with circular pieces of bulb-scale placed inside were also tested, but the bulb-scale dried within a few days, even with wax sealing the cut edges, so this was dismissed. Different diameter holes, as well as varying depths to the cell, were also tested. Most success was obtained using 2mm-thick black Darvic™, with a 7mm-diameter hole for optimal viewing of the entire arena under a binocular microscope. The edge between the bulb-scale and Darvic™ was most effectively sealed with moulded Blu-tack™ around the rim of the arena, or with angled sides carved in the Darvic™ so that a 9mm O-ring could be placed beneath the bulb-scale to press up into the arena to create a seal (Figure 1.5)

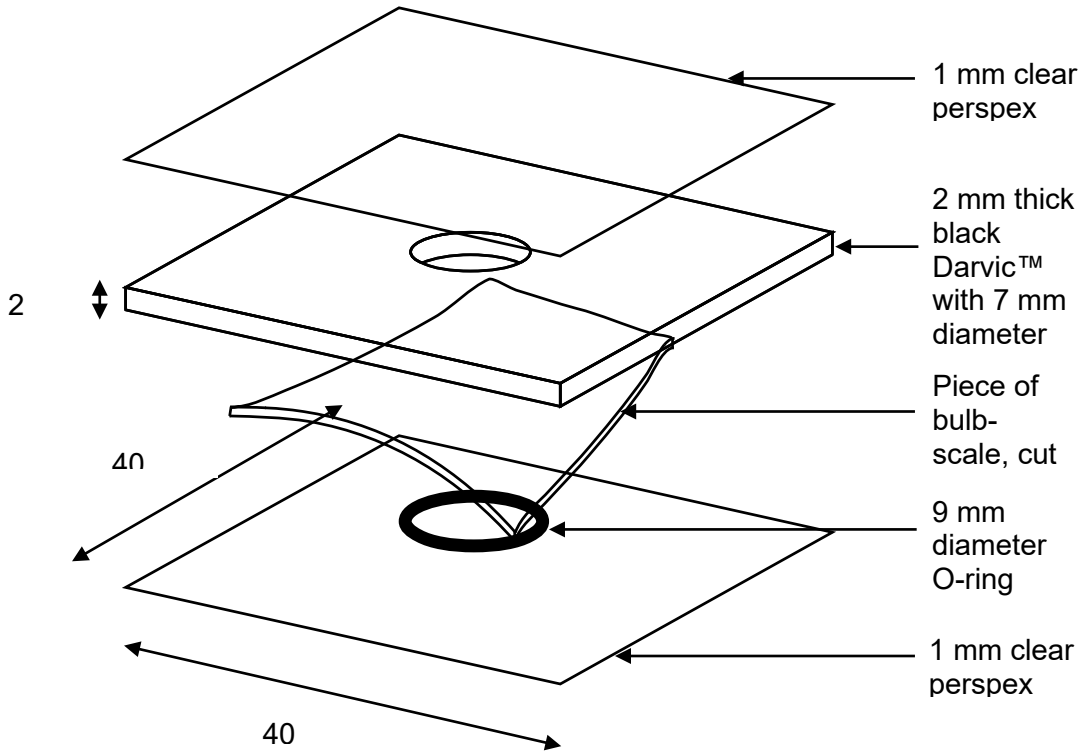


Figure 1.5: Expanded diagram of a cell design used to contain bulb-scale mite for *in vitro* work

For optimal survival of excised bulb-scales, high humidity is necessary to replicate conditions within the bulb. Previous work showed that a relative humidity of ~85% was most effective (Lynch & Bedi, 1994). Cells were therefore placed on metal-grid shelves over aqueous potassium chloride (KCl) solution (160g KCl made up to 400 ml) in sealed perspex containers 155mm x 270mm x 100mm high (Stewart Plastics Ltd., UK), a method developed by De Courcy Williams *et al.* (2004). Humidity and temperature were monitored using a data logger (Squirrel, Grant Instruments (Cambridge) Ltd, UK). The bulb-scale remained fresh for the longest time when the convex side of the scale formed the base of the cell. Problems were encountered with the loss of cells due to fungal growth on the bulb material, and so bulb material was therefore surface-sterilised before use. However, most success was obtained simply by careful selection of healthy material and rinsing it with sterile water prior to use. Mould growth also occurred on the Blu-tack™, meaning that cells could only be used for one week to ten days before risk of fungal contamination. Using O-ring cells resulted in samples of bulb-scale remaining healthy for the longest periods (average of 20 days), although a ledge created by the O-ring hampered observation of the mites.

A considerable amount of time was spent developing this technique, but it was never considered robust enough to collect reliable data on mite development at different temperatures. Therefore, because understanding of the development of mite populations under natural conditions was limited, the project was re-focussed (see below).

Additional studies

Images of all life-stages were taken using a Scanning Electron Microscope (SEM) to aid identification. Male and female adults, and large and small larvae, were critical point dried for examination. Images revealed the presence of folds in the integument of the smaller larvae, the pattern of which could be observed in the larger larvae and adult integument (Figure 1.6). This would suggest small larvae enlarge to stretch out the integument before pupating and moulting to an adult, implying that there is only one larval stage in development. Studies on other Tarsonemid mites have found male larvae to be smaller than female larvae, with a more prominent opisthosoma (Jeppson, Keifer & Baker, 1975). The different sized bulb-scale mite larvae could therefore be different sexes, ranging in size due to age.

In an attempt to increase numbers of mites available for future work, contact was made with a Dutch group working on *Hippeastrum*, to find out how to rear bulb-scale mite on *Hippeastrum*. Some *Hippeastrum* bulbs with a suspected infestation were potted up and infested with bulb-scale mites. These were maintained in a greenhouse at Wellesbourne but because the infested stock of cv Dutch Master was obtained (Figure 1.2), they were never used.

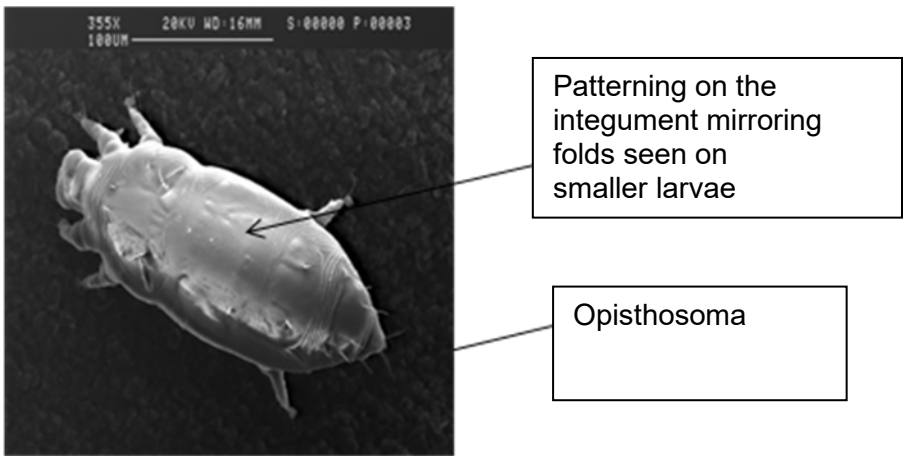


Figure 1.6a: Dorsal view of an adult female, showing patterning in the integument similar to the folds seen in smaller larvae and a much reduced opisthosoma (the hind part of the body behind the legs)

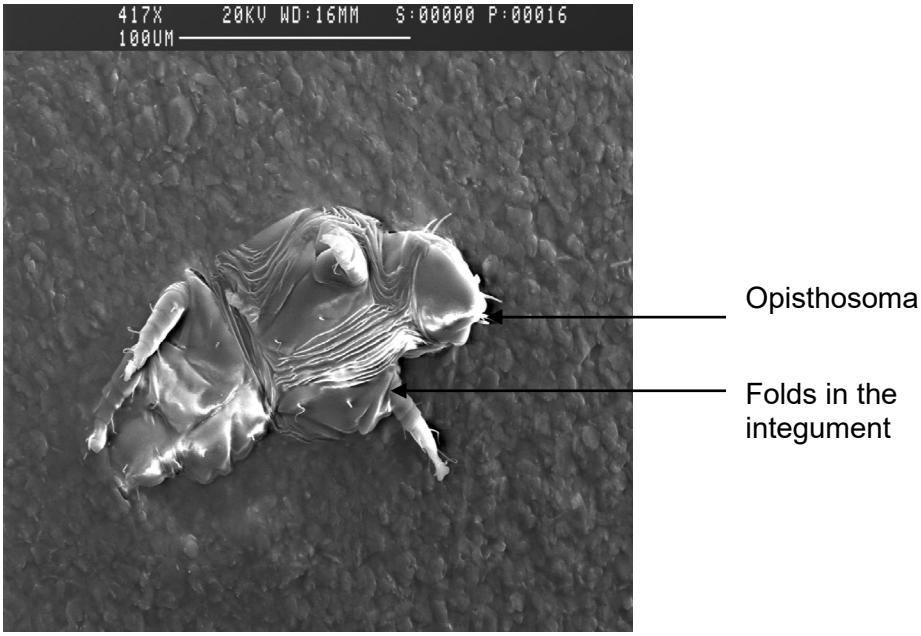


Figure 1.6b: Ventral view of a small larva, showing folds in the integument, particularly between the third leg pairs leading to the pronounced opisthosoma, possibly indicating this is a male (Jeppson *et al.*, 1975)

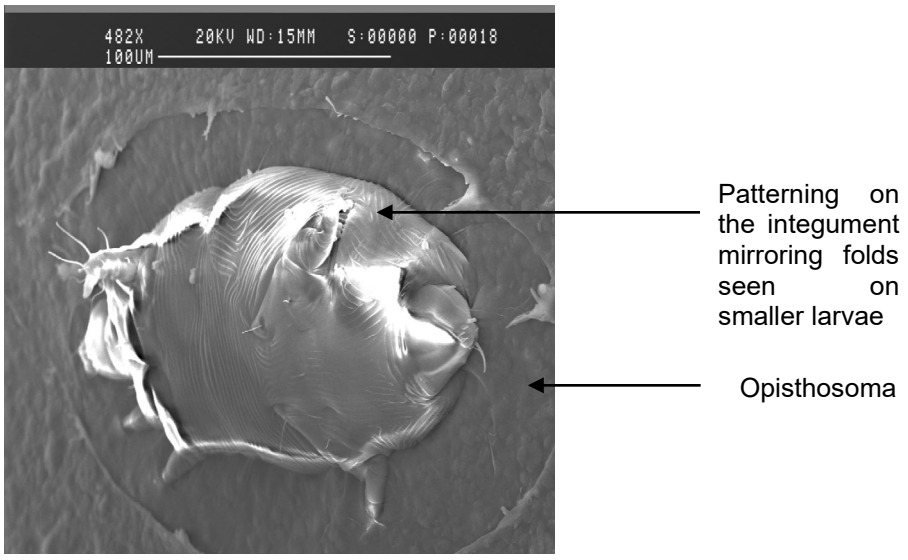


Figure 1.6c: Ventral view of a larger larva, showing patterning on the integument similar to the folds seen in smaller larvae. The less pronounced opisthosoma suggests this is a female (Jeppson *et al.*, 1975)

Development of bulb-scale mite infestations under 'natural' conditions

To increase understanding of the development of bulb-scale mite infestations under natural conditions (see above), experiments were set up to sample bulbs at regular intervals and determine the number and location of the different stages within the bulbs.

The first experiment used potted bulbs that had been used previously for an HDC-funded trial on 'narcissus physiological rust' (BOF 62) and which were known to be infested with bulb-scale mite. These potted bulbs were of mixed varieties with a single variety per pot, and were maintained outside on a gravel standing ground at Wellesbourne. A sample of 30 bulbs (randomised over varieties) per month was examined in detail over a period of several days and the data are summarised by month in Figures 1.7-1.9. The largest numbers of eggs were found on the foliage in January – March, and thereafter egg numbers remained relatively low (less than 0.5 per bulb) (Figures 1.7 and 1.8). The largest numbers of adults were found in June and all of these were inside the bulbs as the foliage had died back by that time (Figure 1.8 and 1.9).

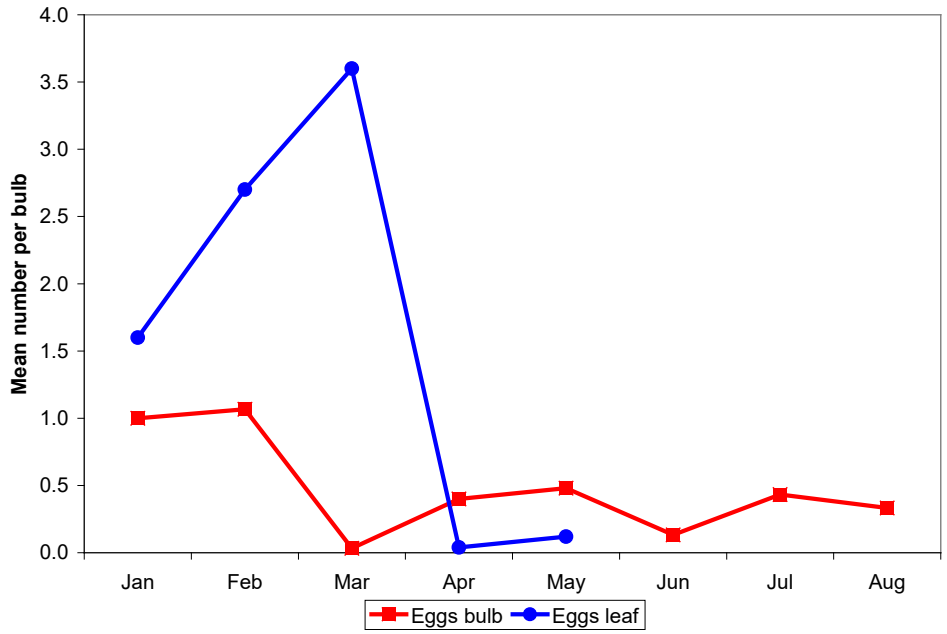


Figure 1.7: Mean number of bulb-scale mite eggs per bulb or leaf in samples taken from potted bulbs stood outdoors at Wellesbourne in 2008

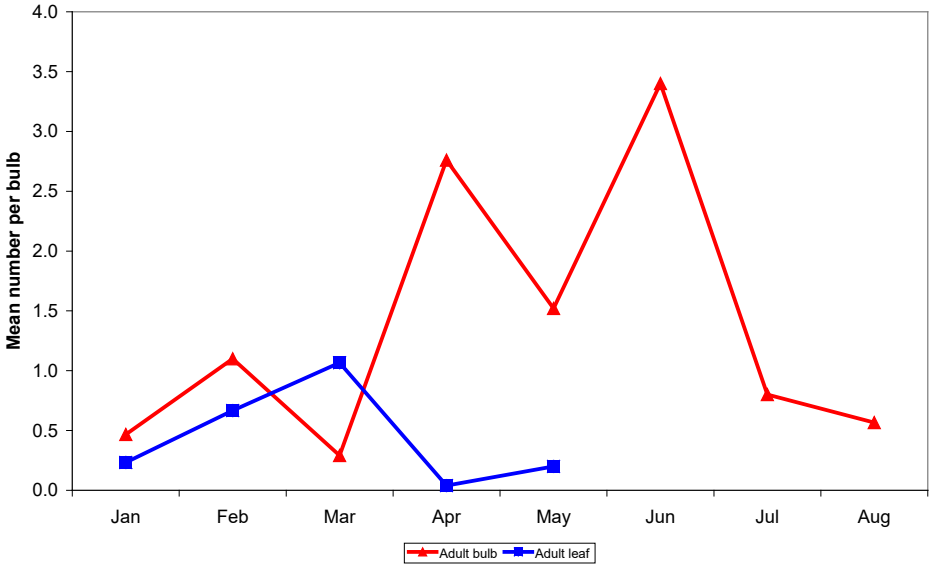


Figure 1.8: Mean number of bulb-scale mite adults per bulb or leaf in samples taken from potted bulbs stood outdoors at Wellesbourne in 2008

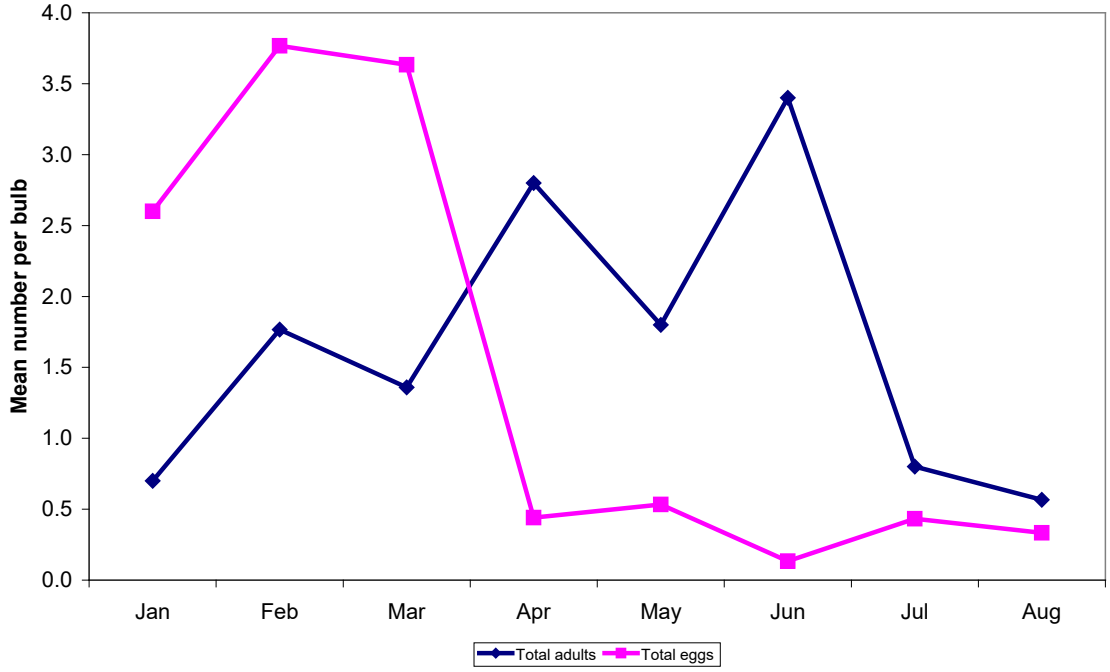


Figure 1.9: Mean number of bulb-scale mite eggs and adults per bulb plus leaf in samples taken from potted bulbs stood outdoors at Wellesbourne in 2008

The larger stock of infested bulbs (cv Dutch Master) was obtained in summer 2008, and the focus of regular sampling was moved to these from November 2008, once they had been planted in the field.

In this instance, a sample of 10 bulbs was removed from the field plot every month as far as possible and the bulbs were assessed in detail. Figure 1.10 shows the mean numbers of adult mites and eggs per bulb during the two year period of assessment. The infestation was very high initially and then declined through winter/spring 2009 before increasing to a small peak in June 2009. This was followed by a decline until March 2010, when numbers increased again, peaking in early July 2010. Figure 1.11 shows the location of the mites (adults and eggs) at different times of the year. The neck of the bulb is the favoured location in the summer period. Figure 1.12 shows the mean feeding mark score throughout the period of assessment (using a scale of 0-5) and Figure 1.12 shows the location of feeding marks (in old, current or new bulb-scales, i.e. in the bulb unit that flowered last year, flowers this year, or will flower next year). The pattern is less clear but indicates that damage was usually greatest in the current scales.

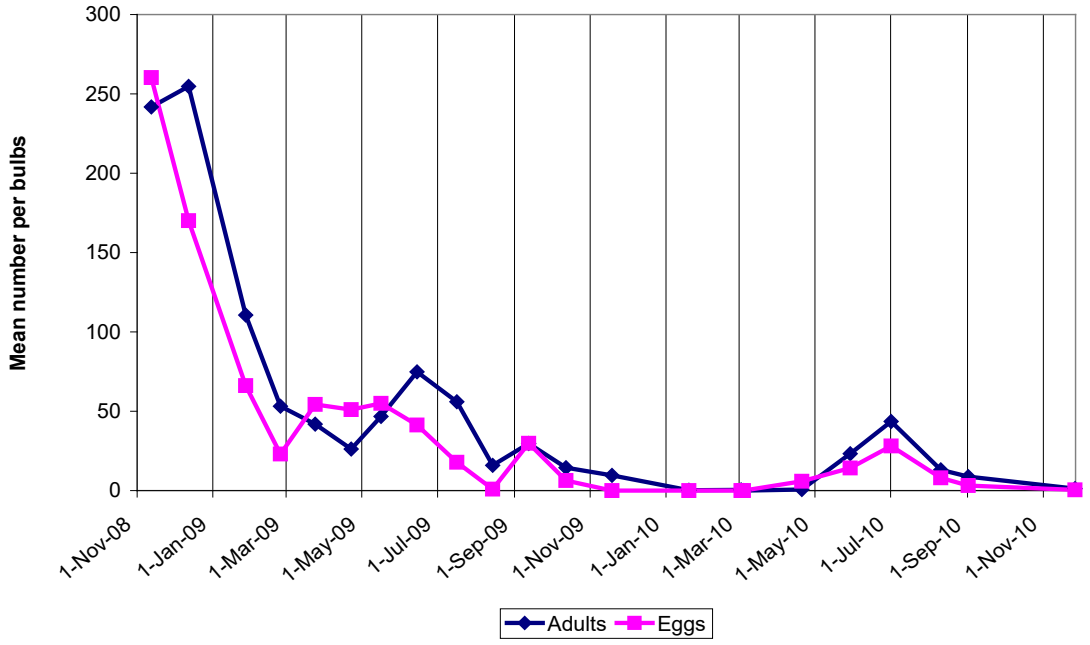


Figure 1.10: Mean numbers of bulb-scale mites and eggs per bulb during the two year period of assessment on cv Dutch Master at Wellesbourne

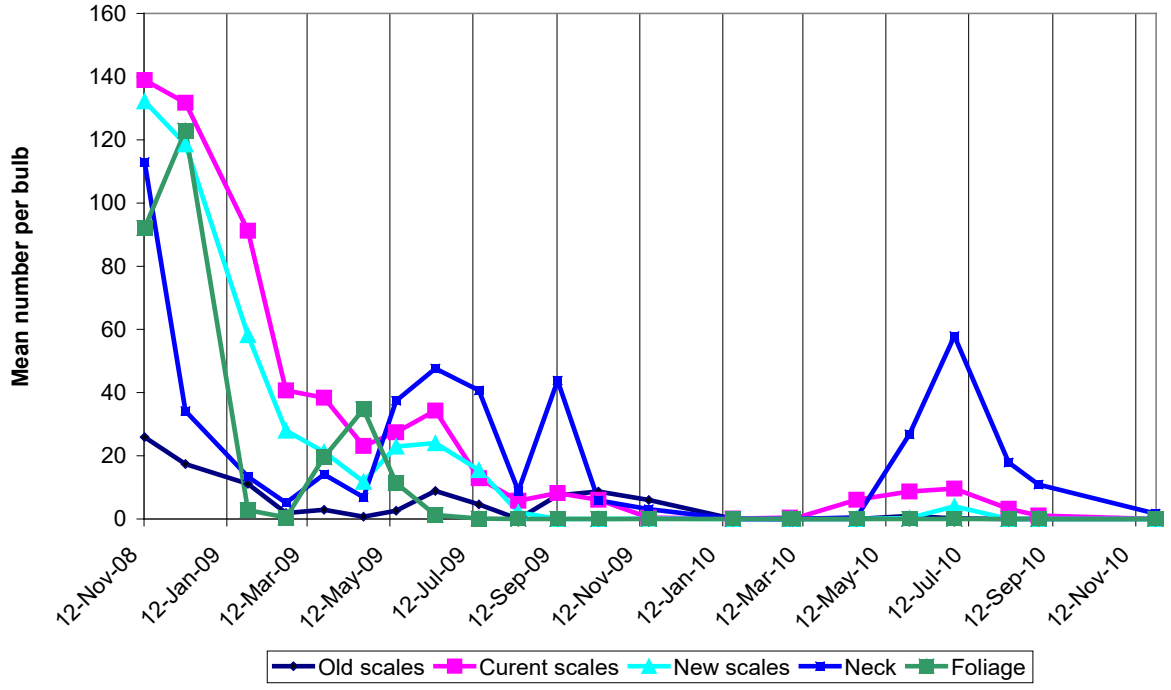


Figure 1.11: Mean numbers of bulb-scale mites (including eggs) per bulb in different locations during the two year period of assessment on cv Dutch Master at Wellesbourne

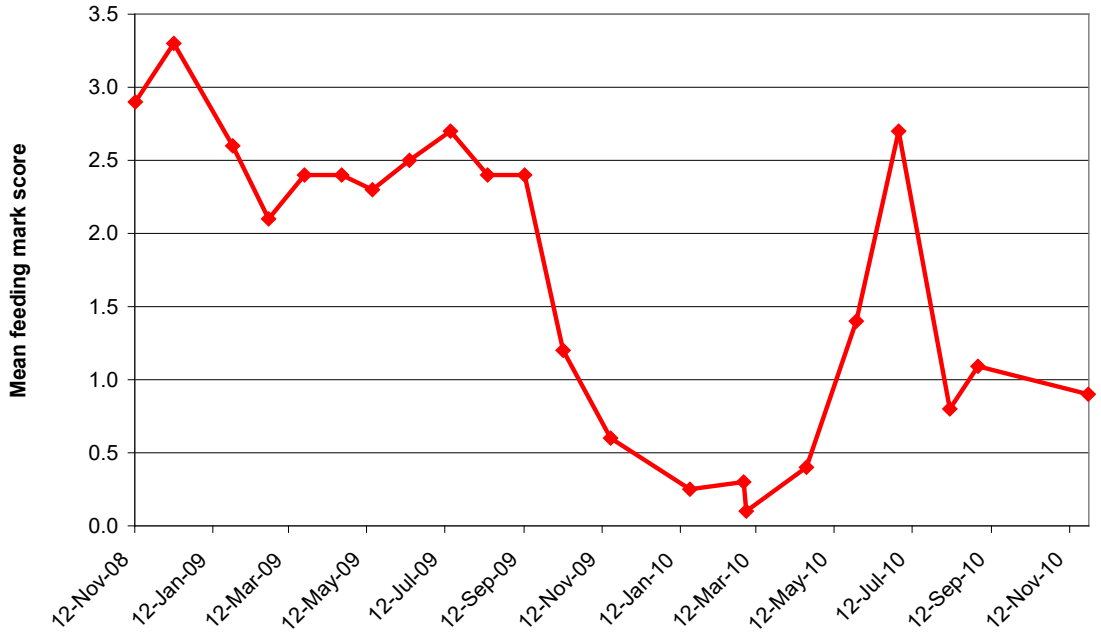


Figure 1.12: Mean feeding mark score (0-5) during the two year period of assessment on cv Dutch Master at Wellesbourne

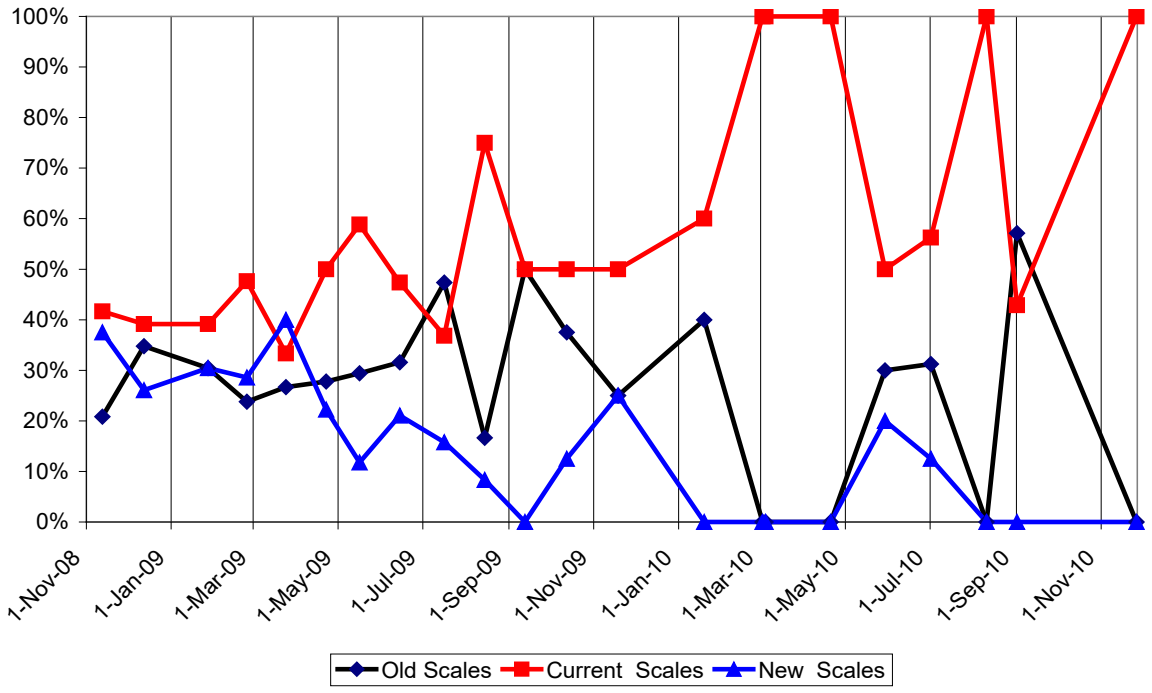


Figure 1.13: Location of feeding marks (old scales, current scales, new scales) as a percentage of the total number of feeding marks during the two year period of assessment on cv Dutch Master at Wellesbourne

Objective 2: Discover when, where and how bulb-scale mite originates and spreads in field crops and in bulb storage

Determine the spatial distribution of BSM in the field by taking samples from commercial crops

2007

Between 5 July and 9 August 2007 five narcissus fields were selected for sampling in each of Cornwall and eastern England (Table 2.1). The fields sampled were typical of the region, had a large block of a single cultivar, and included crops of various ages (crop-years). Ten, 10-bulb samples were dug from each field in the pattern shown in Figure 2.1, which included samples from both sides of the field and from edge, central and intermediate ridges. When digging bulbs, a check was made for bulb-scale mite symptoms (e.g. serrated edges of leaves and stems, curly leaves or bright green foliage) in the immediate vicinity of the sample. Each 10-bulb sample was placed in a labelled nylon net bag and transported to the Kirton Research Centre, where samples were stored in a ventilated location at room temperature and assessed within 7 days.

Table 2.1: Narcissus crops sampled for bulb-scale mite spatial distribution in 2007, including one crop that had been grown for 7 years

Crop number	Region	Location	Cultivar	Crop-year
1	Eastern England	1	Golden Harvest	2
2	Eastern England	2	Carlton	1
3	Eastern England	3	Fortune	2
4	Eastern England	4	Kerensa	2
5	Eastern England	5	Standard Value	1
6	Cornwall	1	Ice Follies	2
7	Cornwall	2	Golden Harvest	3
8	Cornwall	3	Golden Harvest	7
9	Cornwall	4	Golden Harvest	2
10	Cornwall	5	Ice Follies	2

To estimate the amount of bulb-scale mite damage, each bulb was cut transversely two-thirds of the way up. The presence or absence of brown feeding marks was recorded using a 0 to 5 scoring system (0, none; 1, up to 2 small marks; 2, up to two conspicuous marks; 3, up to 5 marks; 4, ≥5 marks; 5, larger areas of damage), and the mean score was calculated for each group of 10 bulbs.

No foliar symptoms were seen. A summary of feeding mark scores is shown in Figure 2.2. There was mite damage in all 10 crops, although the overall level of damage was low (all mean scores were <1). There was no evidence that damage was consistently greater or less at the edges of the fields than towards the centre. Most of the crops sampled were either in their first or second crop-year, and there was no obvious correlation between the age of crops and the incidence of bulb-scale mite symptoms. Even in the one long-term (7-year) crop, the scores were not consistently high. The main finding was that crops in Cornwall generally showed more damage than those from eastern England, though it should be noted that the average age of the crops in Cornwall (2.3 years, if the long-term crop was excluded) was slightly greater than that in the east (1.6 years), this representing the tendency for using longer-term crops in the South-West.

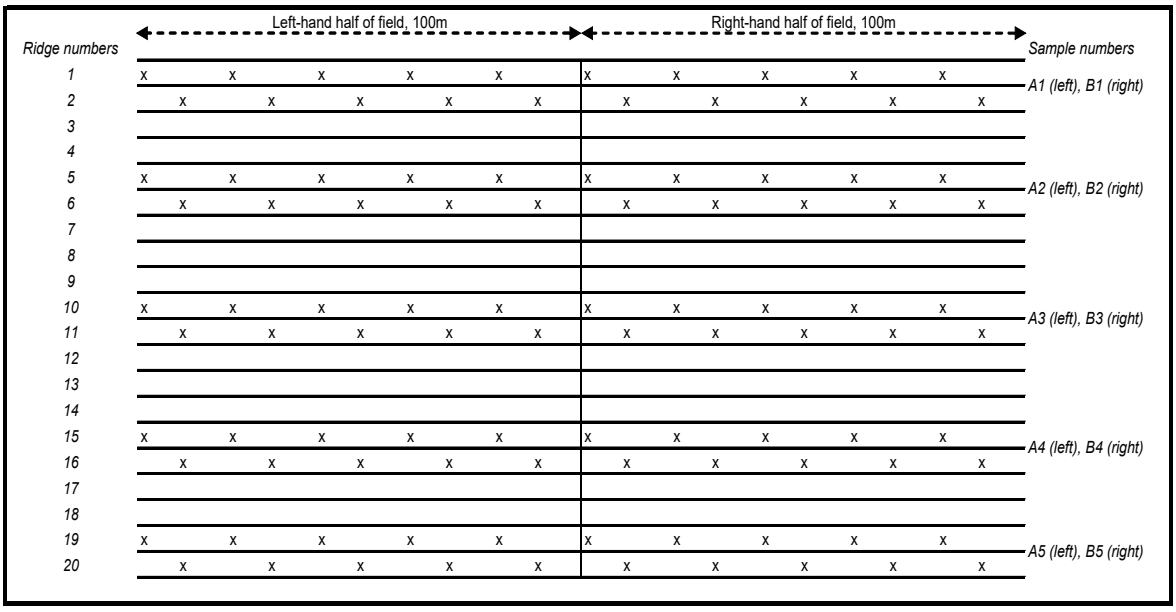


Figure 2.1: Plan used to sample 100 bulbs per field to determine the spatial distribution of mite-infested bulbs. 10 bulb-samples were taken from each of the 10 areas indicated. This illustration shows a field of 20, 200m-long ridges, the plan being adapted to suit actual field dimensions

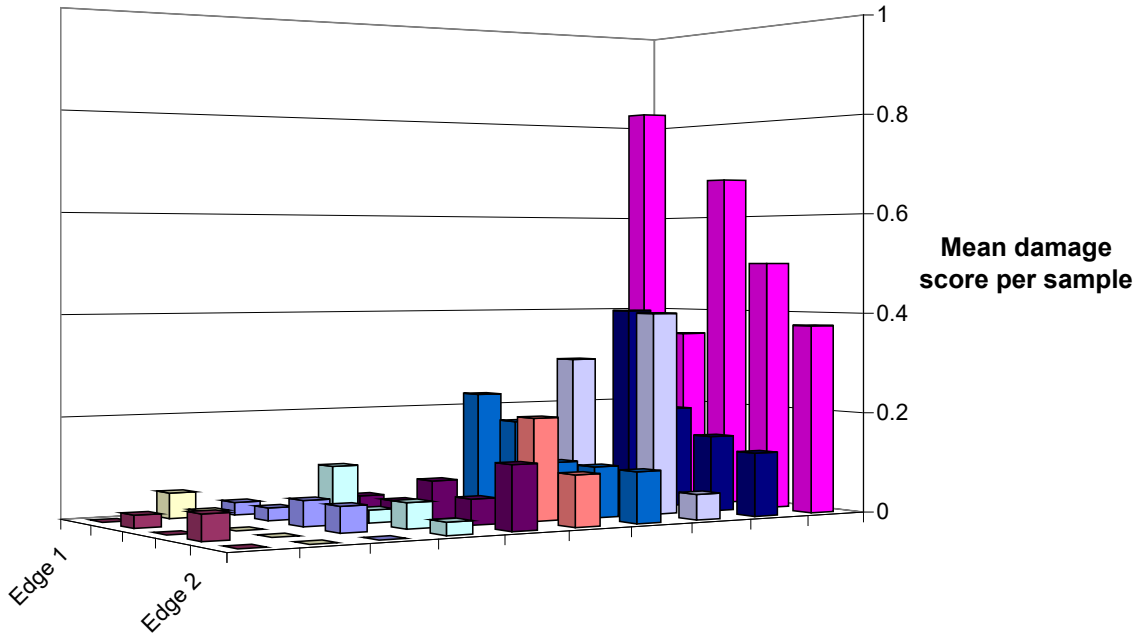


Figure 2.2: The distribution of mite-infested bulbs in ten commercial crops sampled in 2007. X-axis: crops 1 to 10 (left to right) (presented in the order used in Table 2.1 so the first five crops from the left are from Eastern England and the second five crops are from Cornwall); y-axis: ridge position, from edge 1 to edge 2. The mean damage scores are the means of the left- and right-hand replicates

On 5 November 2007 additional samples were taken from one of the more heavily-damaged Cornish crops, the long-term Golden Harvest crop (no. 8 in Table 2.1), to obtain further information about the distribution of mite-damaged bulbs. A 'clump' of ten bulbs was taken from each of 25 locations on a 5 x 5 grid (approximately 5 rows x 5 metres), as shown in Figure 2.3. This sampling was repeated in three locations across the field. The samples were transported to Kirton for storage and assessed as previously described. The incidence of mite damage was measured as the percentage of bulbs in each 10-bulb sample showing feeding marks and the severity of damage (mean damage score). The data were analysed to determine whether there was any evidence of association between the incidence of mite damage in clumps of bulbs at different distances from one another.

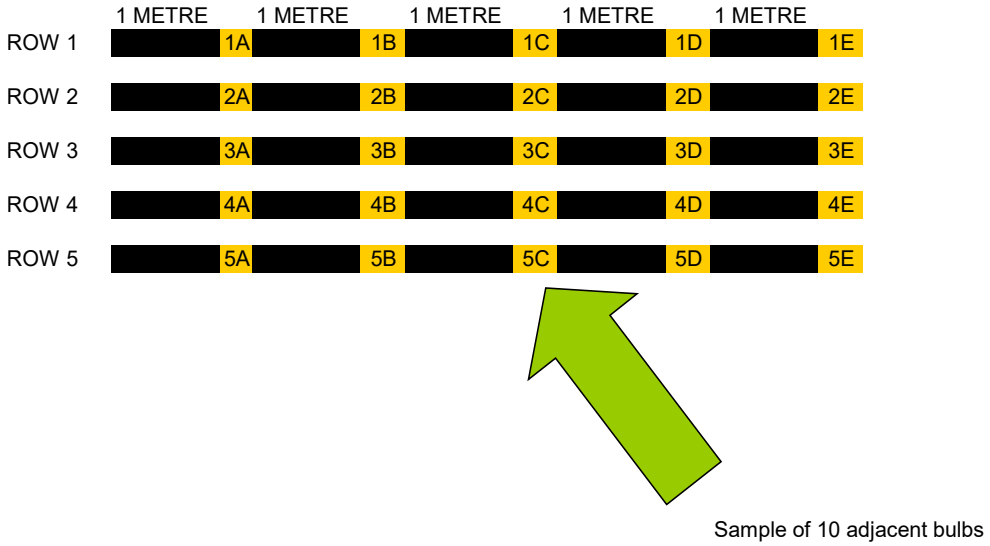


Figure 2.3: Sampling grid used in 2007 to collect a second set of bulb samples to assess spatial distribution of bulb-scale mites

Figure 2.4 shows the percentage of damaged bulbs in one of the 5 x 5 grids, and Figure 2.5 shows the mean damage score for the same grid. These data indicated that infestation occurred in patches in the field. The data were first analysed using a Generalised Linear Model Analysis of the proportion of undamaged bulbs, assessing for differences between the blocks, between ridges within blocks, and between samples along ridges. There was evidence for differences between blocks, but no strong evidence for systematic differences between ridges or between samples.

Moran's I statistic was then calculated for pairs of samples at different distances from one another, to identify any patterns of spatial association between levels of damage. Separate analyses were done based on the mean damage score and the proportion of undamaged bulbs (Table 2.2). The cells shaded in yellow in Table 2.2 indicate significant levels of association at the indicated distances. Values in the row labelled 0 are for different separation distances along the bulb rows (1, 2 and 3m), while values in the other rows also include the separation between ridges (Row 1 indicates one bulb-row apart, Row 2 means two bulb-rows apart, etc). For example, Row 1, Column1 (with a value of 3.778 for Moran's I statistic) represents the samples are one ridge apart and are also separated by 1m along a bulb row. Overall, there was evidence for association both between samples up to 2m apart along a row, and between samples in adjacent ridges.

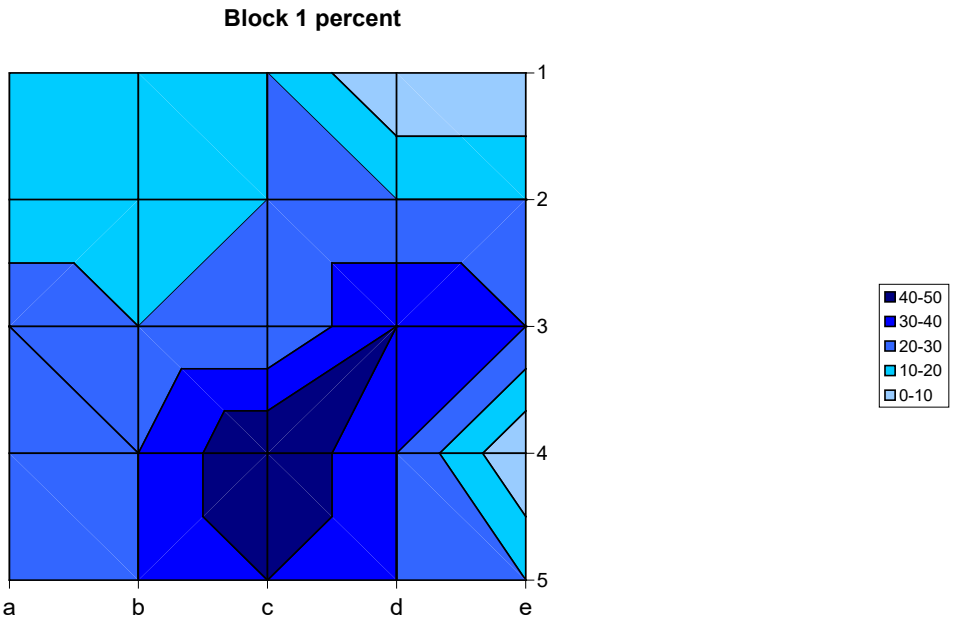


Figure 2.4: The percentage of damaged bulbs in one of the 5 x 5 grids in 2007

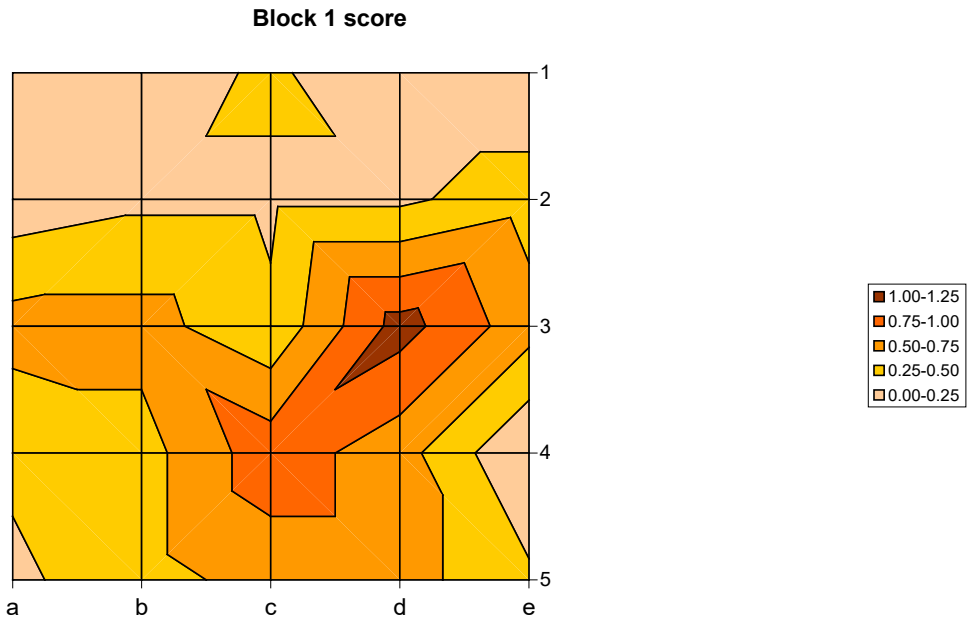


Figure 2.5: The mean damage score for the bulbs in the same grid as Figure 2.4

Table 2.2: Summary of analysis of spatial distribution data using Moran's I statistic (see text for explanation)

a) Mean damage score

Number of ridges apart	Distance between samples (m)			
	0	1	2	3
0	*	4.565	2.165	2.065
1	2.643	3.778	2.866	0.744
2	-0.172	1.386	1.909	0.301
3	-0.321	-0.195	-0.368	0.439

b) Proportion of undamaged bulbs

Number of ridges apart	Distance between samples (m)			
	0	1	2	3
0	*	4.902	1.904	0.403
1	2.544	3.2	2.817	0.998
2	-0.121	0.824	1.96	1.318
3	-0.16	-0.094	-0.378	0.747

2008

Between 17 July and 21 August 2008 further bulb samples were taken from five Cornish and five eastern stocks, where available using the same bulb stocks and fields as in 2007 (Table 2.3). The sampling procedures described above were used, except that the sampling of the 5 x 5 grids was replicated only twice at each site, and the bulb unit (i.e. in the old, current or new bulb unit) in which feeding marks were located was noted.

The summary results for 2008 are given in Table 2.4. Overall the percentage of bulbs with feeding marks, and the feeding mark score, were markedly higher in the Cornish samples than in those from eastern England. From the different stocks, the percentage of bulbs with feeding marks varied between 0.2 and 2.5 (mean 1.4%) in eastern samples and from 0.5 to 25.8 (mean, 11.6%) in Cornish samples (for both sets of samples the data are presented in Table 2.4 as numbers of bulbs with feeding marks out of a total of 500). The average feeding mark scores were 0.13 for eastern samples and 2.21 for Cornish samples. In four out of five samples from the east, the distribution of feeding marks was about equally split between scales of the current and old bulb units, while in four out of five Cornish samples more feeding marks were found in the scales of the old bulb units, implying there had been different infestation rates each year in each region. The average age of crops in Cornwall and the East (excluding the one long-term crop) was similar to that in 2007, 2.5 and 1.4 years, respectively. As previously found, there was no clear correlation between the incidence of feeding marks and the age of the crop, though in 2008 the 8-year-down crop did have the highest feeding mark score. In the three cases where crops were examined in two successive years, there was no obvious increase in bulb-scale mite infestation year-on-year, except in the long-term crop.

The examples in Figure 2.6 show similar distributions to those found in 2007. Several samples evidenced the patch-wise spread of infestations, and, overall, infestations appeared to spread either from the edges or from within crops.

Table 2.3: Narcissus crops sampled for bulb-scale mite spatial distribution in 2008. Where the crop used was the same as in 2007, this is indicated by *

County	Location	Cultivar	Crop-year
Eastern England	1	Spellbinder	1
Eastern England	2	Tamara	1
Eastern England	3	Carlton	1
Eastern England	4	Carlton	2
Eastern England	5	Standard Value	2*
Cornwall	1	Standard Value	3
Cornwall	2	Standard Value	3
Cornwall	3	Golden Harvest	8*
Cornwall	4	Golden Harvest	3*
Cornwall	5	Ice Follies	1

Table 2.4: Summary of bulb-scale mite feeding mark (FM) data for samples of ten narcissus crops in 2008

Region	***Farm***	***No. of bulbs with FM (per 500 bulbs)***	***For bulbs with FM, percentage in different bulb units***			***FM score (average of 50, 10-bulb samples)***
			New	*Current*	*Old*	
East	1	5	0	83	17	0.12
	2	12	0	75	25	0.28
	3	11	9	45	45	0.22
	4	1	0	0	100	0.01
	5	4	25	50	25	0.04
	Mean	**7**	**7**	**51**	**42**	**0.13**
Cornwall	1	75	0	0	100	3.07
	2	31	3	52	45	0.96
	3	54	0	0	100	2.08
	4	129	0	4	96	4.90
	5	2	0	0	100	0.04
	Mean	**58**	**1**	**11**	**88**	**2.21**

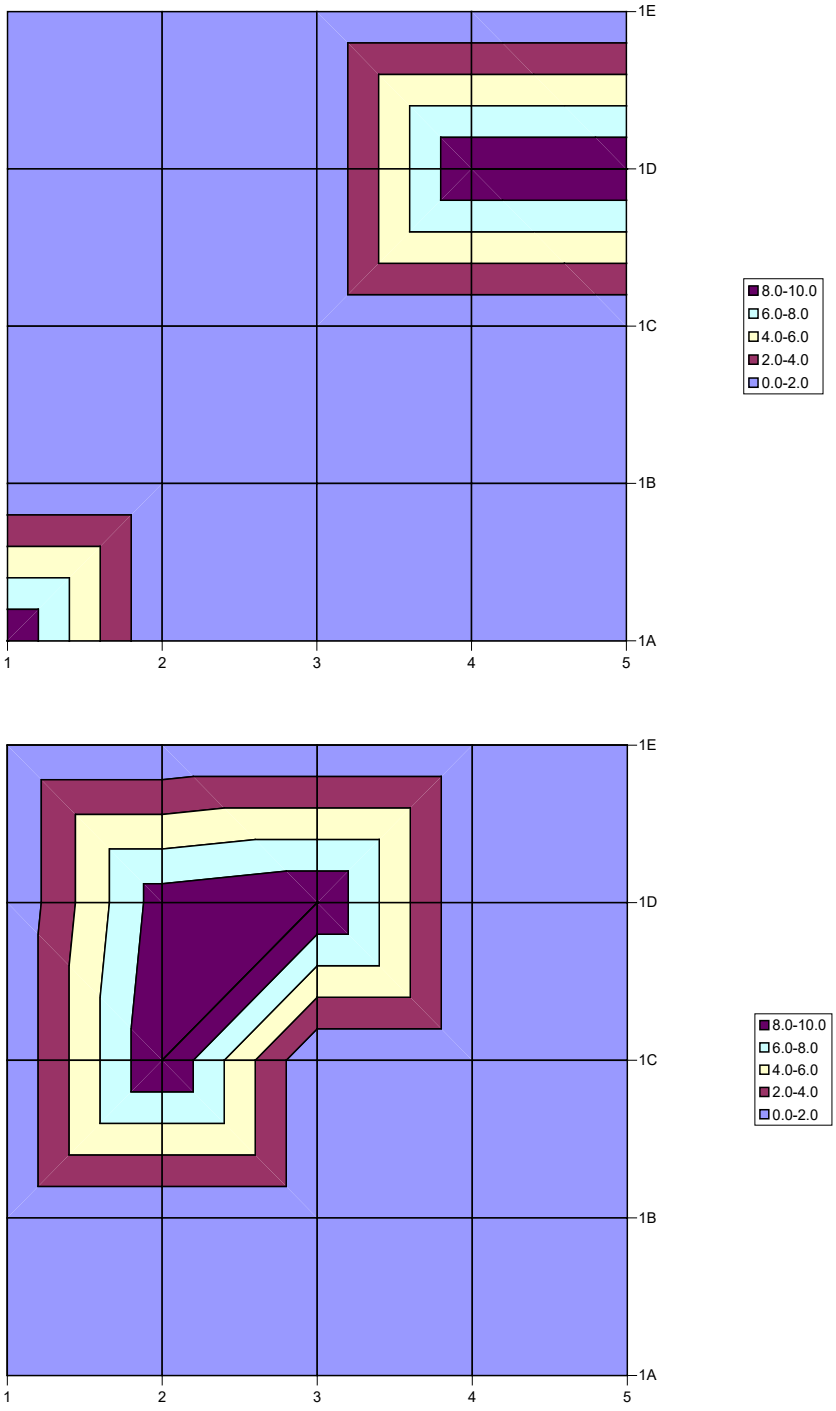


Figure 2.6: Two examples of the distribution of feeding marks (as the percentage of damaged bulbs in 5 x 5 grids) from samples taken in 2008

The data from 2008 were subjected to statistical analysis as described above. All eastern sites and 1 south-western site had damage levels that were too low for analysis. The data from the 4 remaining locations in south-west showed the following:

Along rows

- At all sites high numbers in one sample were correlated with high numbers in adjacent samples (1 m apart) or vice versa
- For >1m separations the results were more variable
- For 3 m separations there were mainly negative correlations suggesting that if damage was high in one spot, then bulbs 3 m away would have lower damage (the reasons for this are unclear)

Between rows

- The results were less clear – there was some evidence of correlation between adjacent rows.

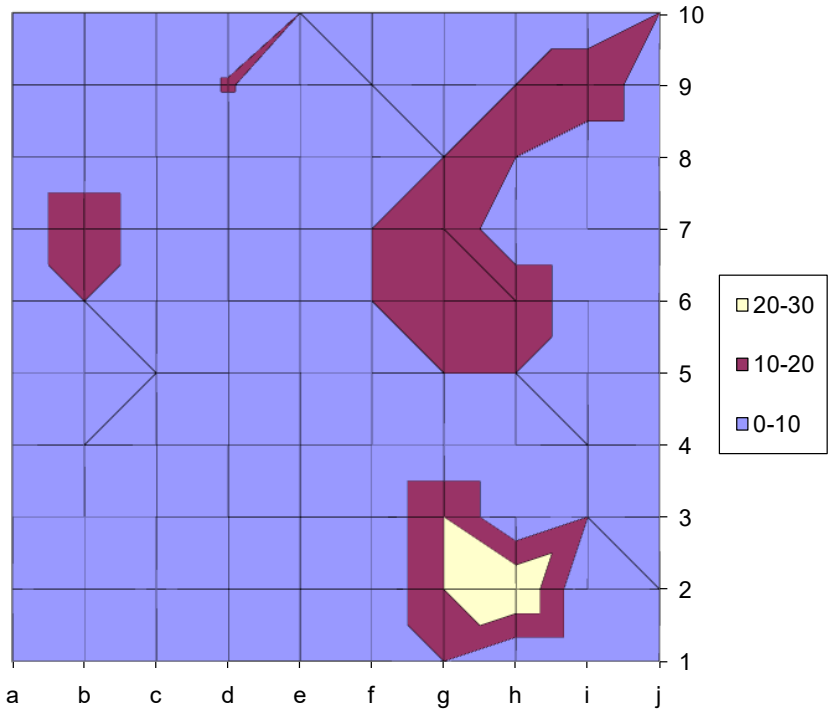
2009

In 2009, five Cornish crops were sampled using a 10 x 10 grid i.e. 10 rows x 10 metres long. Two grid samples were taken in each crop. A sample of 10 bulbs was taken from each sampling point (at metre intervals along each row). These bulbs were assessed for the presence of feeding marks. The mean percentage of damaged bulbs in each grid is shown in Table 2.5. All crops had been damaged by bulb-scale mite but the infestation levels varied considerably between crops. There was a good level of consistency between crops, suggesting that the samples represented the overall level of damage in that planted stock. Diagrams summarising the distribution of damaged bulbs in the 10 x 10 grids are shown in Figures 2.7a-e. The rows are labelled a-j and the sampling points along the rows are labelled 1-10. Most of the diagrams show clear foci of infestation, suggesting that mites have moved out from a central infestation point. The diagrams for Grower D in particular suggest movement along rows as well as some movement across rows. The data were again analysed using Moran's I statistic. There was no overall pattern to the distribution of mite infestation between the sites. However, there were similar patterns in the two grids sampled at each site.

Table 2.5: Mean percentage of damaged bulbs for the 10 x 10 grid samples taken in 2009

Grower	Years down	Grid 1	Grid 2
A	2 years	3.7	2.7
B	13 years	10.7	12.4
C	-	7.5	3.3
D	3 years	15.5	21.1
E	4 years	44.9	39.1

Grower A1



Grower A2

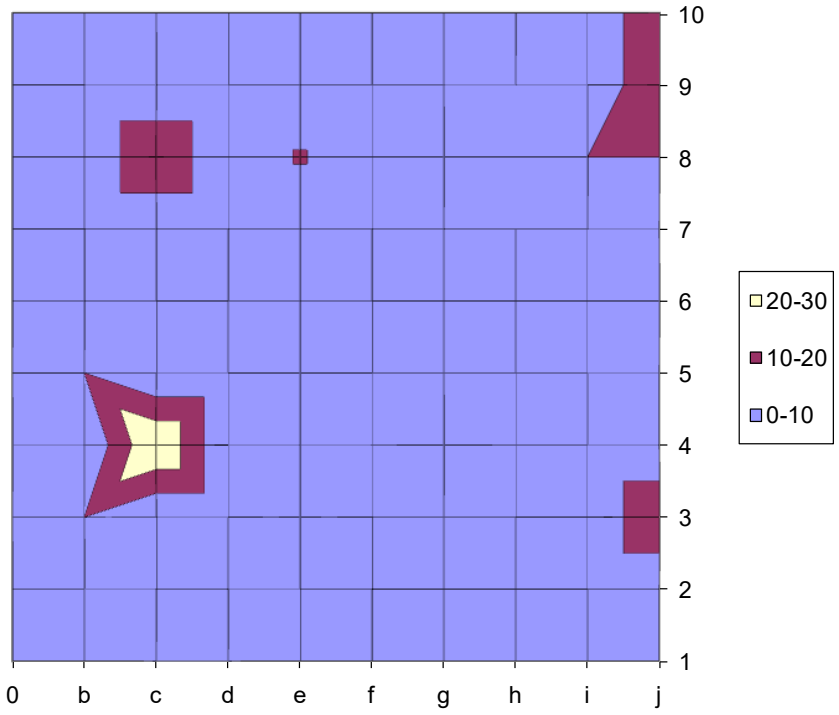
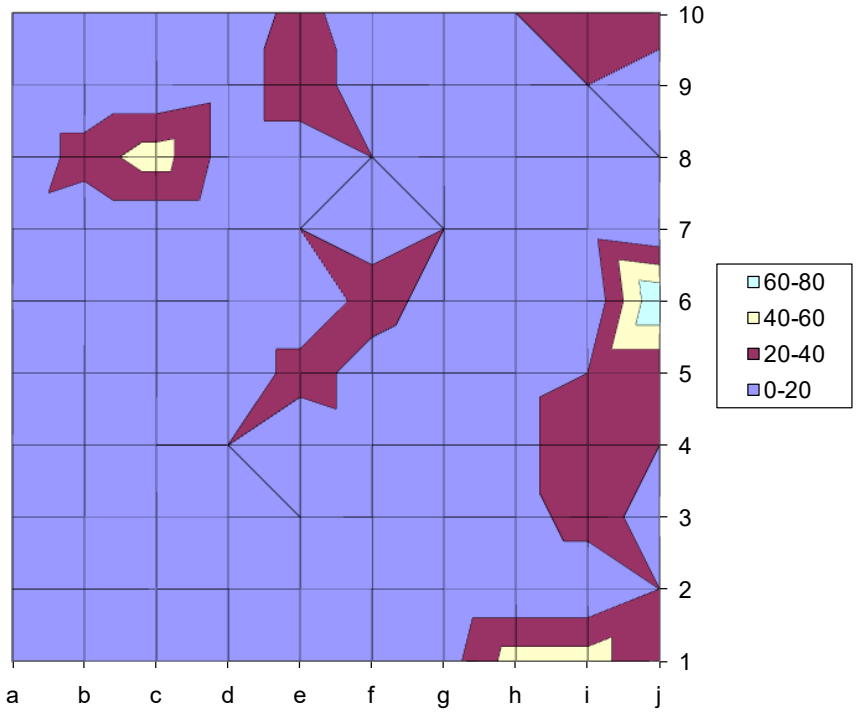


Figure 2.7a: Diagrams of the distribution of feeding marks (as the percentage of damaged bulbs in 10 x 10 grids) from samples taken in 2009. The rows are labelled a-j and the sampling points along the rows are labelled 1-10

Grower B1



Grower B2

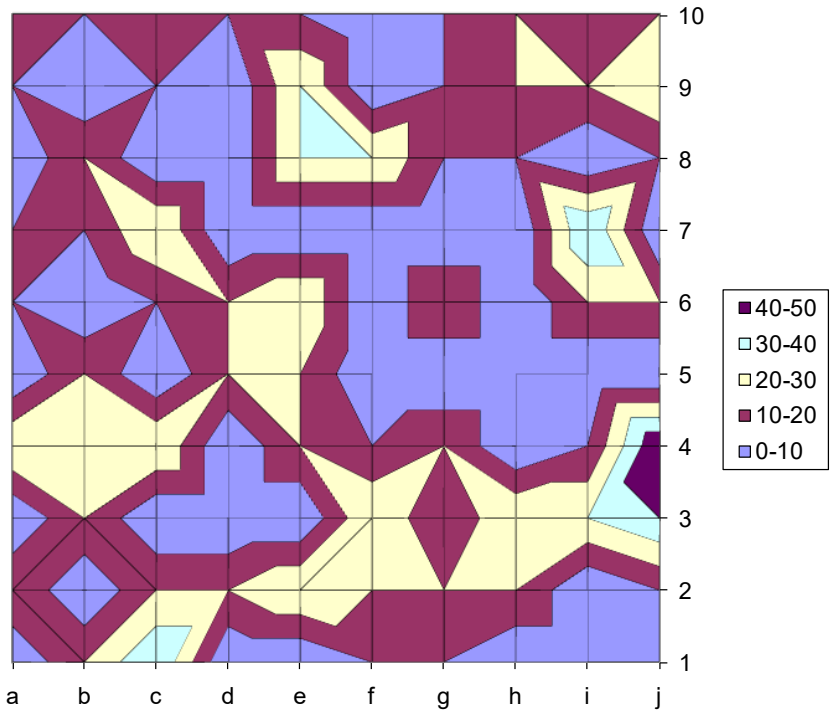
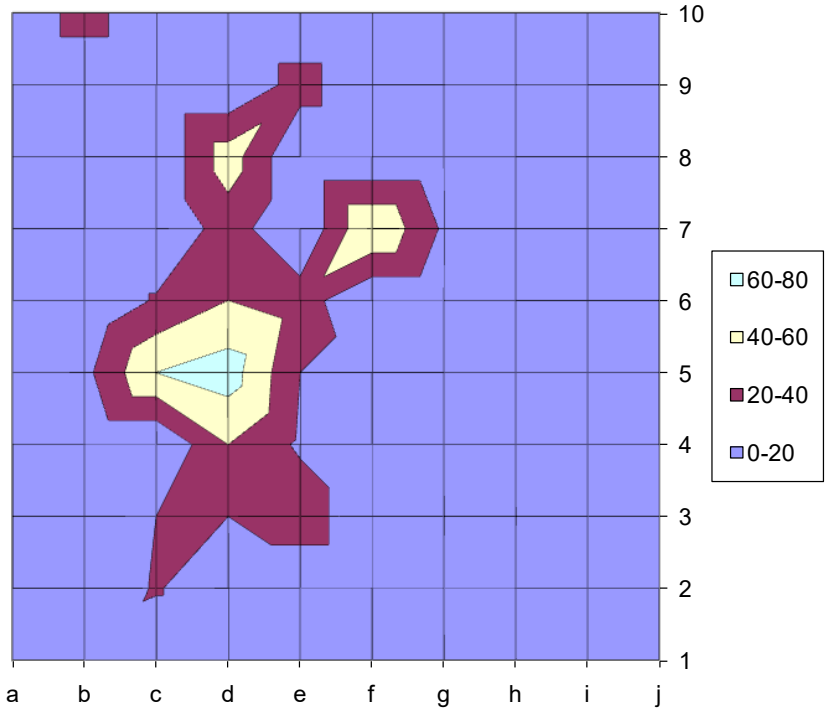


Figure 2.7b: Diagrams of the distribution of feeding marks (as the percentage of damaged bulbs in 10 x 10 grids) from samples taken in 2009. The rows are labelled a-j and the sampling points along the rows are labelled 1-10

Grower C1



Grower C2

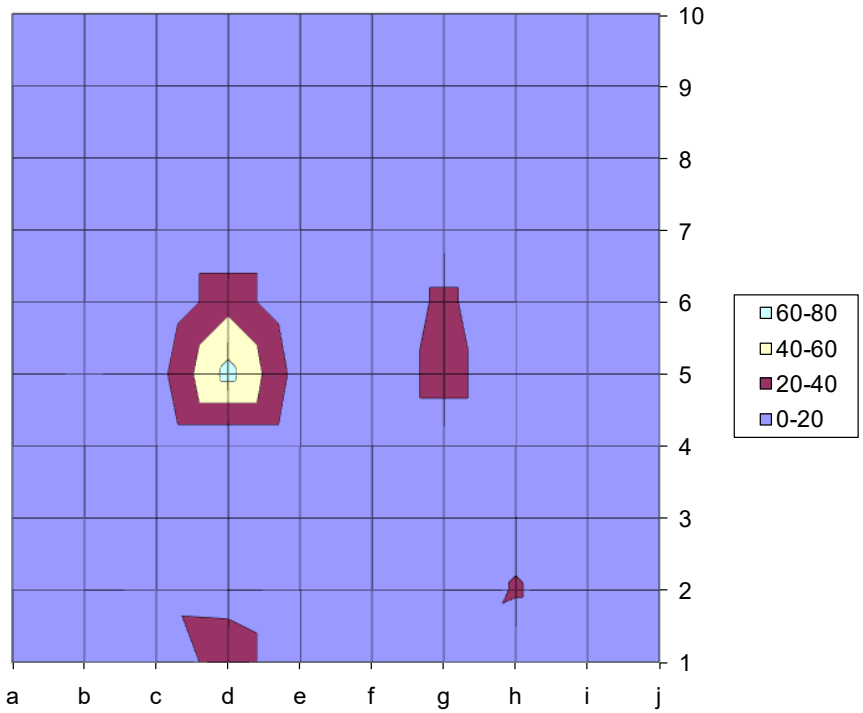
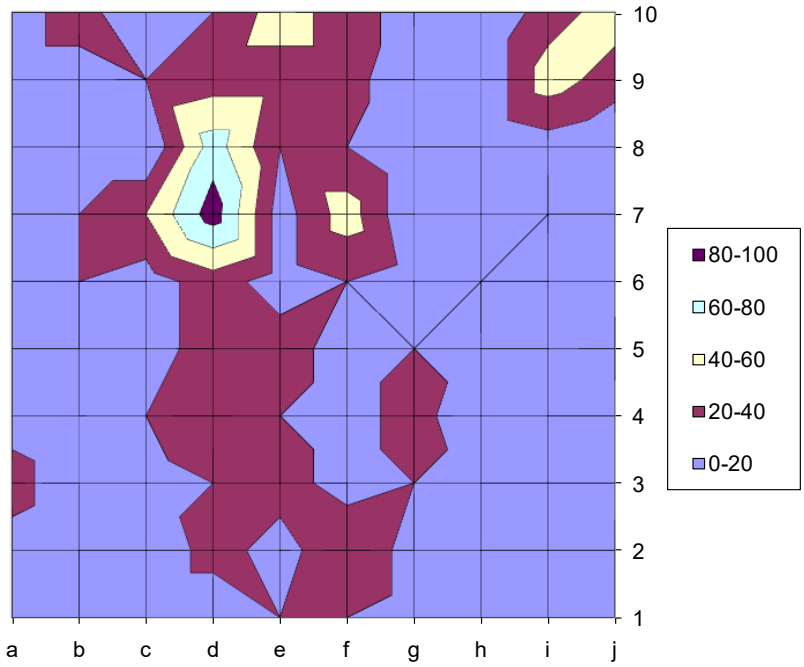


Figure 2.7c: Diagrams of the distribution of feeding marks (as the percentage of damaged bulbs in 10 x 10 grids) from samples taken in 2009. The rows are labelled a-j and the sampling points along the rows are labelled 1-10

Grower D1



Grower D2

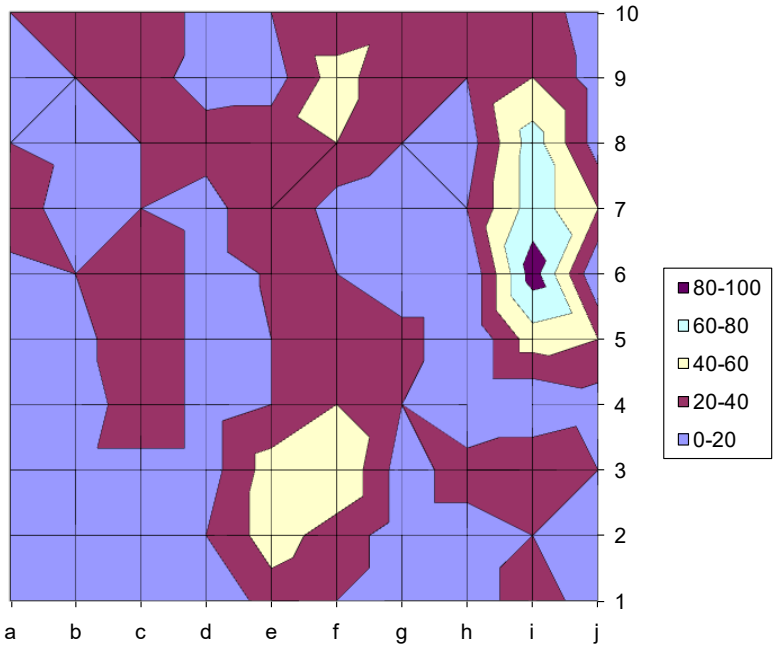
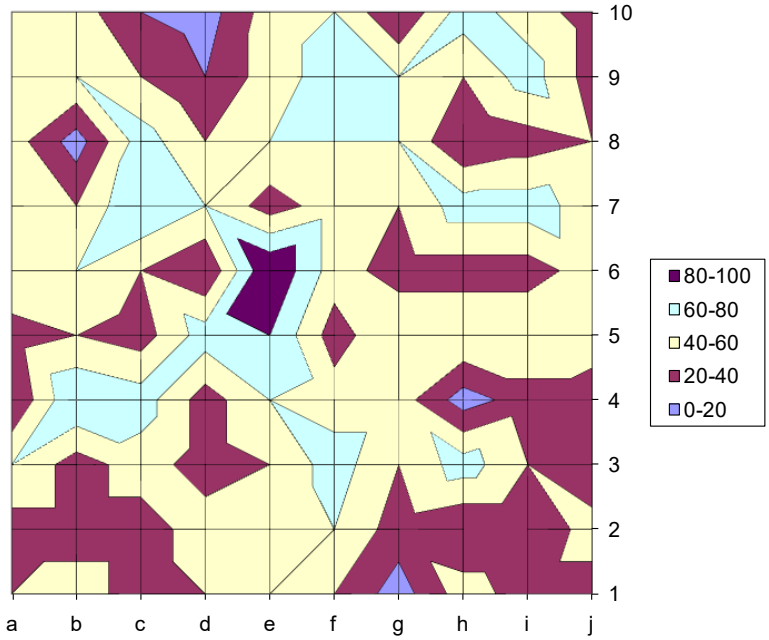


Figure 2.7d: Diagrams of the distribution of feeding marks (as the percentage of damaged bulbs in 10 x 10 grids) from samples taken in 2009. The rows are labelled a-j and the sampling points along the rows are labelled 1-10

Grower E1



Grower E2

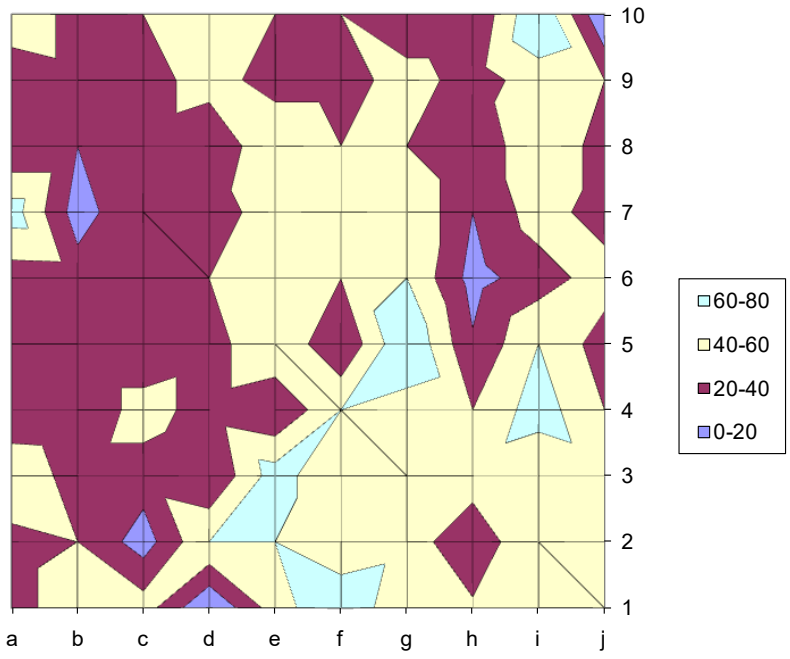


Figure 2.7e: Diagrams of the distribution of feeding marks (as the percentage of damaged bulbs in 10 x 10 grids) from samples taken in 2009. The rows are labelled a-j and the sampling points along the rows are labelled 1-10

Are bulb-scale mites found on common bulb-field weeds?

A preliminary evaluation was carried out on 27 May 2008 on a weedy commercial bulb-field in Lincolnshire. Five plants each of the predominant weeds at the site, rose-bay willow-herb (*Chamaenerion angustifolium*), annual sow-thistle (*Sonchus oleraceus*), groundsel (*Senecio vulgaris*) and potato ground-keepers (*Solanum tuberosum*) were collected (by cutting off at ground level) at random from each section of a 3 x 3 grid across a narcissus cv Golden Ducat crop. The leaf blades and stems, and especially in leaf axils and around leaf bases, were examined under a low-power (LP) binocular microscope. No mites were found.

On 28 May 2008 further checks of weeds were carried out at Kirton Research Centre. Individual weeds, typically groundsel and knotgrass (*Polygonum aviculare* agg.), growing immediately adjacent to narcissus cv Carlton plants that had distinct bulb-scale mite symptoms, were examined. Again, no mites were found.

The approach was tested again at in 2009 at sites in Cornwall and in the large plot of cv Dutch Master at Wellesbourne. No live bulb-scale mites were found on any of the weeds examined.

Are bulb-scale mites present on naturalised bulbs?

Samples of naturalised bulbs were taken in the east and the south-west in 2009. The bulbs (10 per clump) were sent to Wellesbourne where the shoots were examined for mites. Figure 2.8 shows the mean numbers of bulb-scale mites and bulb mites per bulb shoot in each location. No bulb-scale mites were found in the samples from Lincolnshire. Bulb mites were found in both regions.

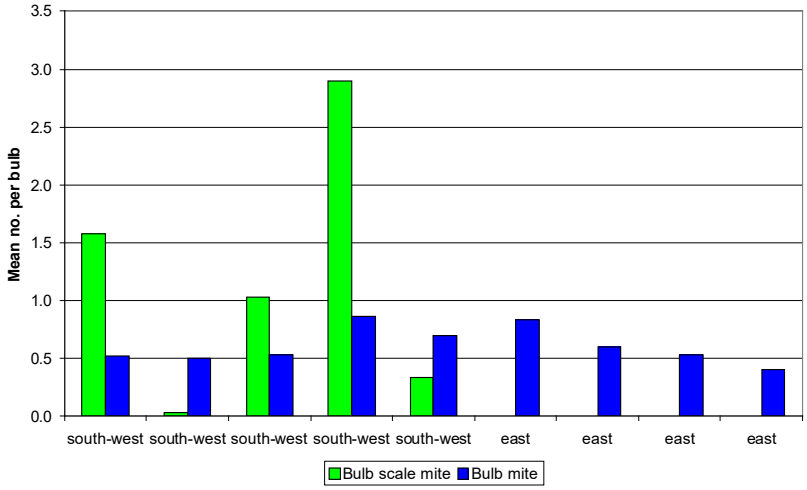


Figure 2.8: The mean numbers of bulb-scale mites and bulb mites per bulb shoot in the south-west and the east in 2009

Sampling was repeated in 2010 at 5 locations in the south-west and one location in Lincolnshire. On this occasion, bulb-scale mites were also found in Lincolnshire (Figure 2.9).

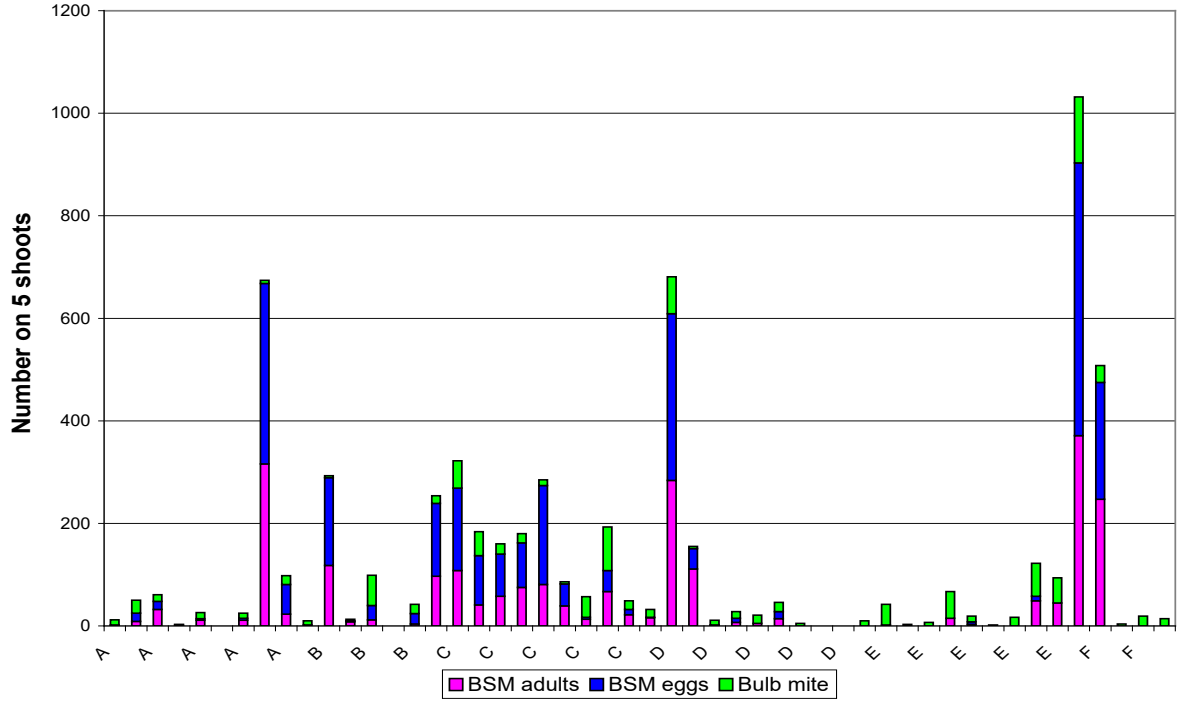


Figure 2.9: Naturalised narcissus - number of bulb-scale mites (BSM), bulb mites or mite eggs on 5 shoots taken from 6 different locations in 2010 (A-E in Cornwall; F in Lincolnshire)

Investigation of movement of bulb-scale mite between growing bulbs

On 6 February 2007, groups of infested bulbs (cv Carlton; 8-10 cm grade) and hot-water-treated narcissus bulbs (cv. Golden Harvest; 12-14cm grade) were planted in 1 m-long plastic planting troughs. Two replicates of the following treatments were set up:

- Control — 8 hot-water-treated bulbs only
- Bulbs touching — 8 hot-water-treated bulbs and 8 infested bulbs
- Foliage touching — 8 hot-water-treated bulbs and 8 infested bulbs – two sets of bulbs separated by a 0.5m gap with foliage tied together in the middle
- 0.5 m apart — 8 hot-water-treated bulbs and 8 infested bulbs – foliage of two sets not touching
- 1 m apart — 8 hot-water-treated bulbs and 8 infested bulbs – foliage of two sets not touching

The troughs were stood initially in a polytunnel at Wellesbourne but when the weather became warmer they were moved outside to the Dutch Lights area (23 April 2007). A further two replicates were set up on 13 May 2007 and these were also stood outside in the Dutch Lights area. They were again provided with shelter in a polytunnel during the coldest part of winter 2008-2009 (moved in on 8 January 2009) in case severe frosting might kill the mites. The troughs were then sampled destructively during February and March 2009 (one replicate at a time) and the numbers of mite adults and eggs were recorded. Figure 2.10 shows the percentage of 'clean' bulbs that became infested with mites. This suggests that mites move between bulbs most readily when the bulbs are close to one another. Figure 2.11 shows the mean number of mites per bulb, suggesting again that mites move most readily when bulbs are close. These results may be 'distorted' because mite numbers per bulb can be very variable.

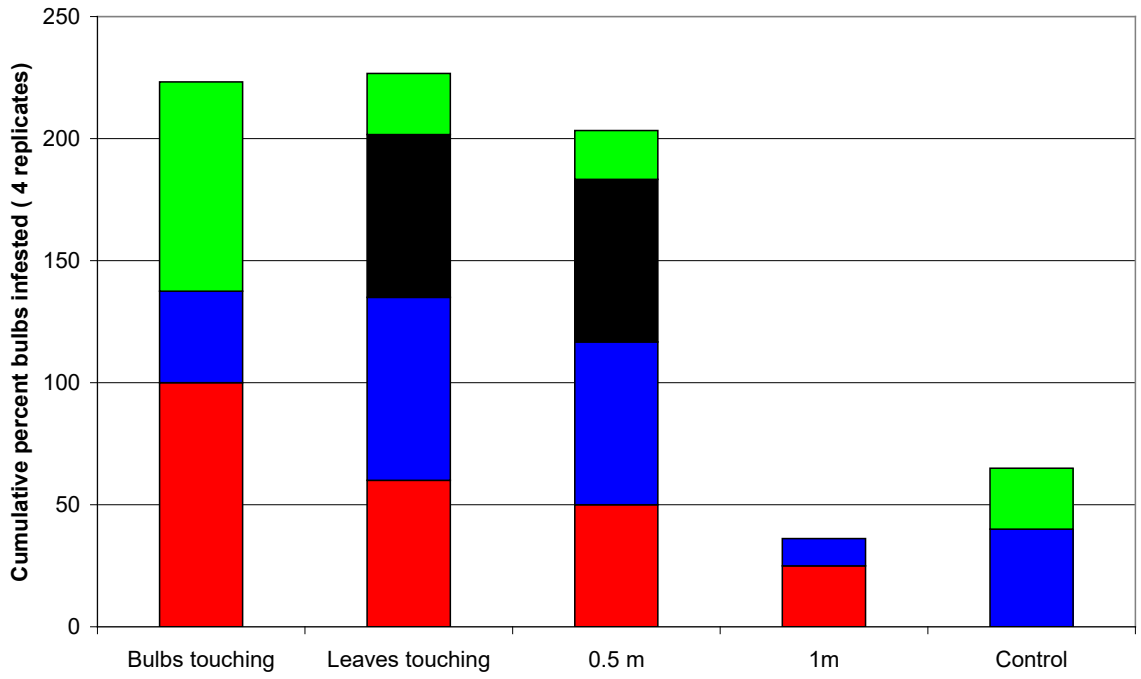


Figure 2.10: The percentage of 'clean' bulbs (cumulative for 4 replicates) that became infested in the trough trial in 2007-8

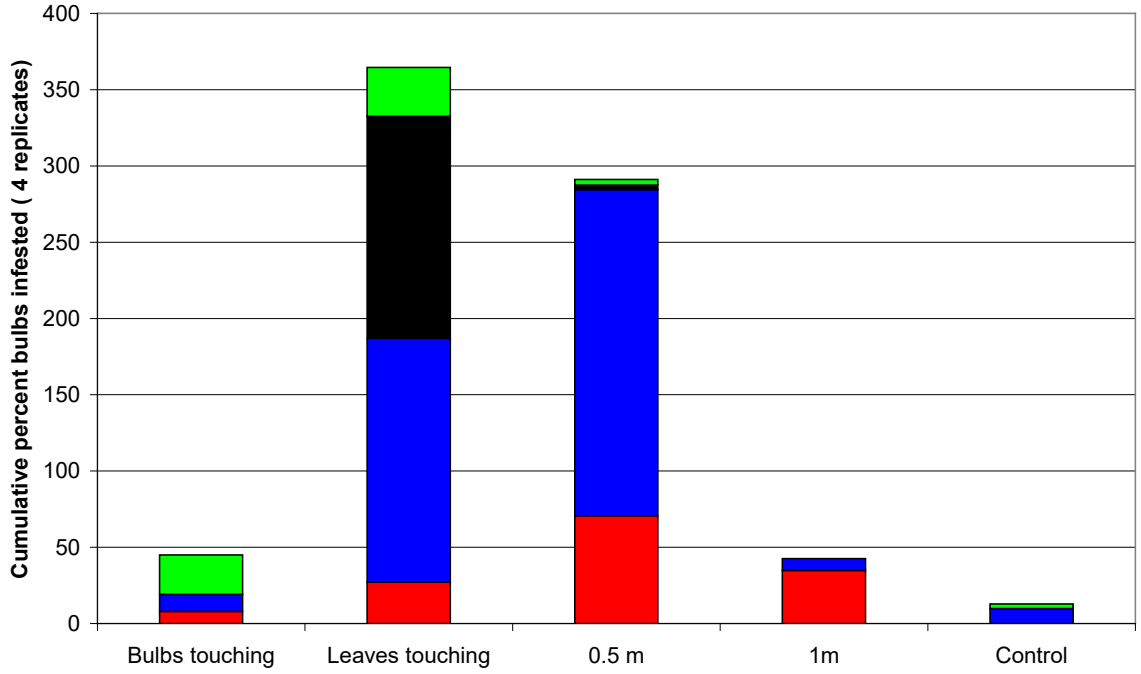


Figure 2.11: The mean number of mites per bulb for 'clean' bulbs that became infested in the trough trial in 2007-8.

To extend studies on mite dispersal, a field trial was planted at Wellesbourne in late summer 2009. Replicated plots of bulbs were planted with un-infested (cv Golden Harvest) and infested bulbs (cv Dutch Master) separated by:

- 0m
- 0.5m
- 1m
- 2m
- 4m

There was also an un-infested control treatment. These bulbs were assessed in spring/early summer 2011 by removing the shoots and counting bulb-scale mites and bulb mites. The data are shown in Figure 2.12 and 2.13.

The infested stock was still infested with bulb-scale mite in the spring/early summer of 2011, indicating that the mite population had survived. Of the initially un-infested bulbs, two were found to be infested in 2011 (Figure 2.12). One of these was immediately adjacent to a row of infested bulbs (0m spacing) and the other was 4m distant from the nearest infested bulbs (4m spacing).

The pattern was similar for bulb mite, although slightly more of the initially un-infested bulbs were infested (Figure 2.13).

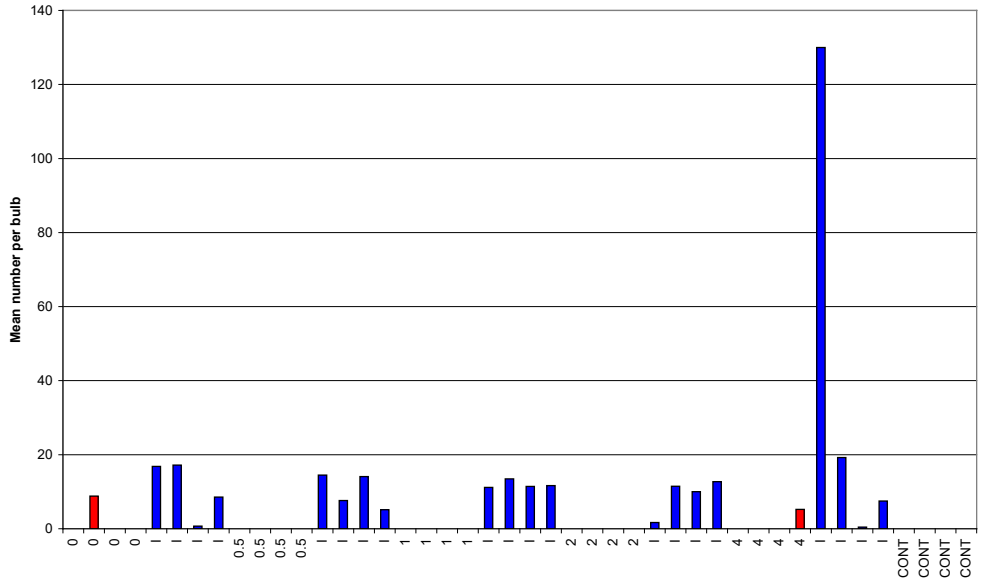


Figure 2.12: Mean number of bulb-scale mites per bulb (shoot) in bulbs planted in rows at different distances from one another. The blue bars indicate bulbs from the infested stock at the start of the experiment and the red bars bulbs from the un-infested stock. For each spacing, the initially infested bulbs are shown to the right of the initially un-infested bulbs. The control bulbs (CONT) were un-infested at the start of the experiment.

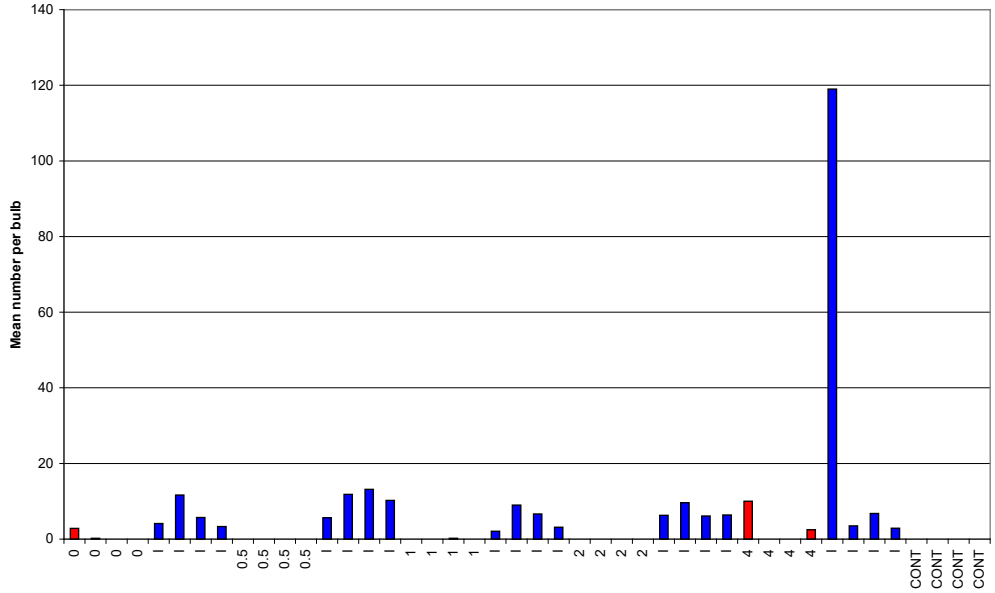


Figure 2.13: Mean number of bulb mites per bulb (shoot) in bulbs planted in rows at different distances from one another. The blue bars indicate bulbs from the infested stock at the start of the experiment and the red bars bulbs from the un-infested stock. For each spacing, the initially infested bulbs are shown to the right of the initially un-infested bulbs. The control bulbs (CONT) were un-infested at the start of the experiment.

Investigate extent of movement of bulb-scale mite between bulbs and around bulb stores and bulb-handling facility

2007

To determine whether mite infestations can arise from the exposure of bulbs to dust and debris during the handling and storage of dry bulbs, dust/debris samples were collected from the premises of seven bulb growers and merchants in August 2007. At each site, samples were taken from up to six locations, representing the whole bulb storage and handling process from the initial bulb cleaning lines and drying walls (or drying floors) through to HWT, grading and pre-planting or pre-sales storage areas (see Table 2.6 for details of locations).

The dust/debris samples were taken to Kirton and, after thoroughly mixing each sample, 50ml samples were mixed with sets of nine test bulbs in paper bags and stored at 20°C for 4 weeks (representing the storage phase when infestation could take place). The test bulbs were cv Golden Harvest that had previously received HWT at the elevated temperature of 46.4°C to ensure they carried no live bulb-scale mites. For each dust sample eight replicate bags of bulbs were set up. As controls, 24 additional bags of nine bulbs each were set up with no added dust.

After 4 weeks the bulbs were planted in 20cm-diameter pots by tipping the contents of the bag (including the dust/debris) onto a peat growing medium, spacing the nine bulbs evenly and topping up the pot with growing medium. The pots were watered, placed outdoors on a standing ground, covered with fleece and kept watered as needed. When the bulbs were ready to force (judged by the development of the bud in the neck of the bulb) they were moved to a glasshouse (heated to 17°C, vented at 20°C).

After 4 weeks in the glasshouse the incidence and severity of bulb-scale mite symptoms (serrated leaf/stem margins, curled leaves and feeding marks on leaves/stems) were recorded (Table 2.7 and Figure 2.14). In the control, there was a low incidence of symptoms of bulb-scale mite damage (about 0.7 per pot of nine bulbs), and this would have been caused by mite activity prior to HWT in 2007. With few exceptions, an elevated incidence of bulb-scale mite symptoms was seen in bulbs that had been mixed with dust samples. Dust from all farms resulted in an increased incidence of symptoms with dust from at least some locations on the farm, but no one location was consistently infective across all farms. Further, the overall incidence of damage varied somewhat between farms. In many instances the incidence of damage rose from the about 0.7 per pot of the controls to 2.0 to

3.0 per pot. The highest incidence of symptoms was seen using dust from the grading line and pre-planting holding areas of one farm, the drying wall and the main bulb storage area of another farm, and the pre-planting storage area of a third. In general, the highest scores were seen in dust samples from drying, grading and storage areas (Tables 2.5 and 2.6).

Table 2.7: Summary of farm locations used for dust samples in 2007, with incidence of bulb-scale mite symptoms. The symptoms seen in the control treatment may reflect a low level of infestation in this 'clean' hot-water-treated stock or may be due to causes other than bulb-scale mite

Farm no.	Location ref.	Location description	Symptoms per pot	Farm no.	Location ref.	Location description	Symptoms per pot
Control			0.7	5	B	Cleaning line	0.8
1	B	Cleaning line	1.7		C	Drying wall	0.4
	C	Drying wall	2.0		D	Grading line	1.7
	D	Grading line	2.2		E	HWT shed	1.0
	E	HWT shed	1.8		F	Storage area	3.0
	F	Storage area	0.9	6	A	Grading line	2.3
2	B	Cleaning line	0.6		B	Grading line	1.4
	C	Drying wall	0.7		C	Drying wall	1.2
	D	Grading line	0.5		D	Drying wall	1.2
	E	HWT shed	1.8		E	Drying wall	1.2
	F	Storage area	1.5		F	Heat store	1.3
3	B	Cleaning line	0.5	7	A	Cleaning line	1.3
	C	Drying wall	1.7		B	Drying wall	1.3
	D	Grading line	2.6		C	Drying wall	2.3
	F	Storage area	3.0		D	Grading line	1.9
4	A	Cleaning line	1.2		E	HWT shed	1.6
	B	Grading line	1.2		F	Storage area	1.7
	C	Cold store	1.8				
	D	Drying wall	2.7				
	E	Storage area	2.5				
	F	Storage area	0.8				
	G	Drying wall	1.7				

Table 2.8: Summary of bulb-scale mite symptoms from dust samples taken at different types of locations in 2007

Location	Number of samples	Minimum score	Maximum score
Control		0.7	
Cleaning line	6	0.5	1.7
Drying wall	11	0.4	2.7
Grading line	8	0.5	2.6
Heat store	1	1.3	1.3
HWT shed	4	1.0	1.8
Storage area	7	0.8	3.0
Cold store	1	1.8	1.8

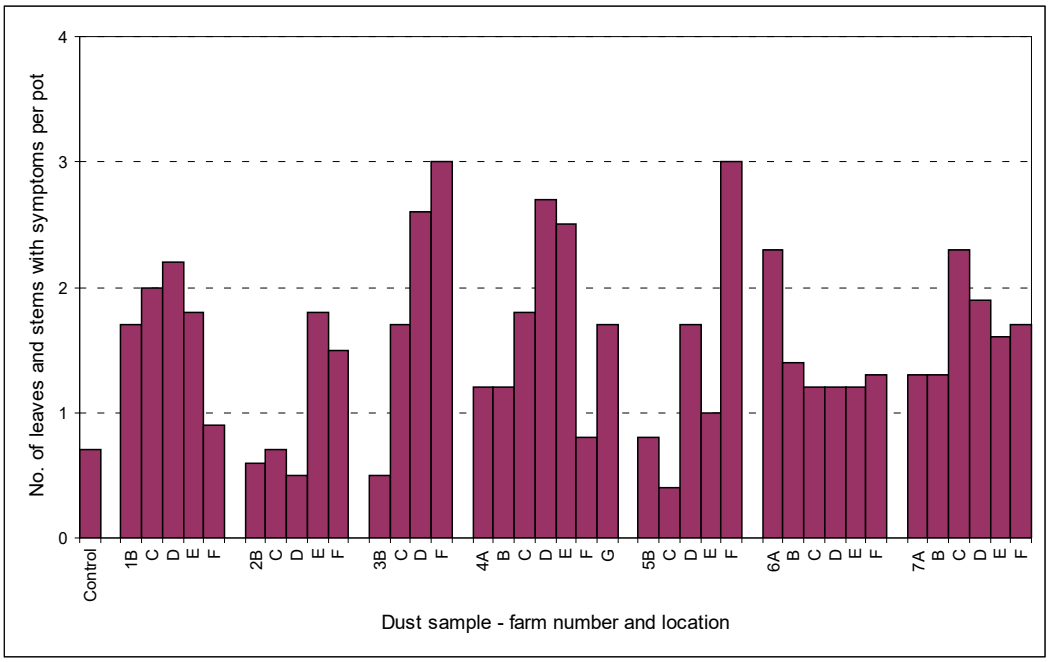


Figure 2.14: The number of leaves and stems with symptoms of bulb-scale mite infestation from dust samples taken in different locations at seven farms (see Table 2.5 for location codes). Data from the 2007-2008 experiment assessed in spring 2008

After foliar senescence the pots were moved to a ventilated, non-heated mesh tunnel and grown-on. Over several days in December 2008 bulbs were recovered from their pots, cut transversely and scored for the presence and severity of feeding marks on the current bulb units (Figure 2.15). While several of the dust treatments produced higher scores than in the controls, there was, however, no correlation between the spring and autumn assessments (Figure 2.16), and the reasons for this need to be investigated. It may be that assessments made in the following autumn are not valid because of the dispersal of mites over the previous spring and summer.

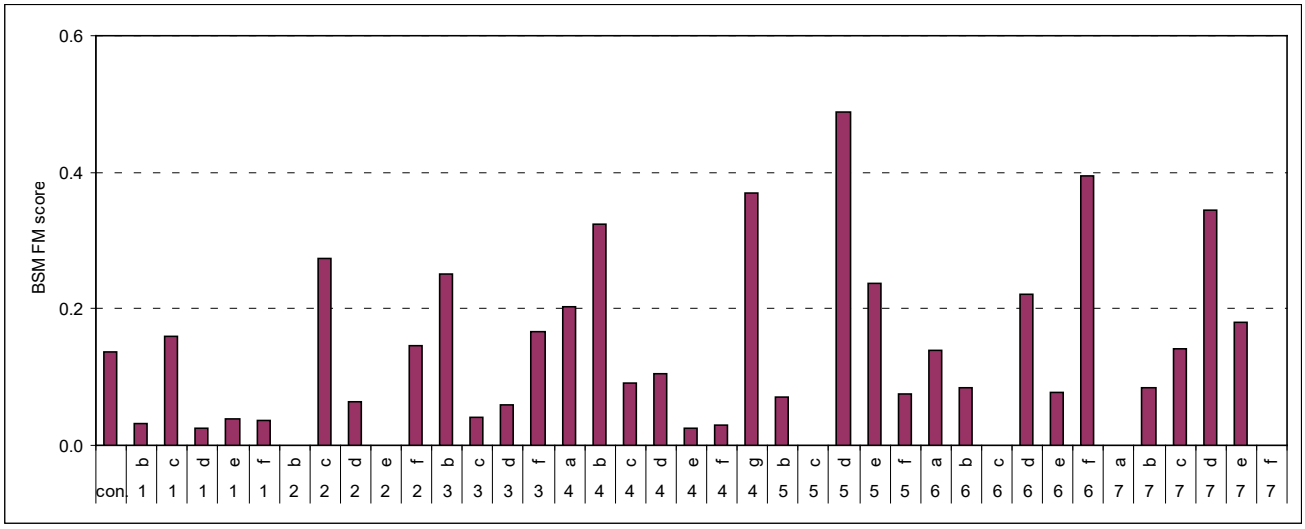


Figure 2.15: Feeding mark scores in bulbs previously treated with dust samples taken in different locations at seven farms. Data from the 2007-2008 experiment assessed in autumn 2008

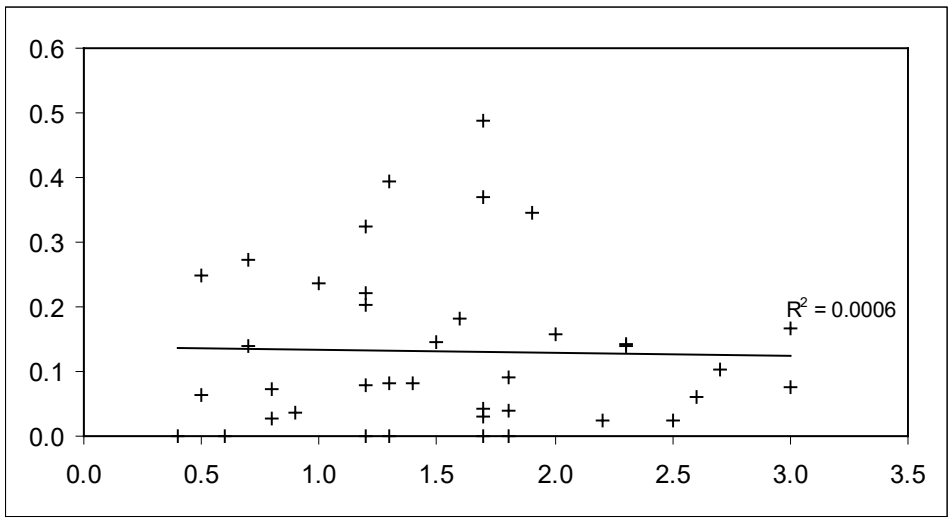


Figure 2.16: Correlation plot of foliage symptoms (in spring 2008) against feeding mark scores (in autumn 2008) in bulbs previously treated with dust samples taken in different locations at seven farms. Data from Figures 2.7 and 2.8

In addition to the main series of dust samples, further samples were collected in late-May 2007 from one Cornish farm. One sample was collected from each of 10 separate drying floors before the company's usual pre-season cleaning of facilities. The volumes collected were variable and the samples contained a proportion of parts of bulbs as well as dust and general debris. After transport to Kirton the samples were placed in calico bags and stored at room temperature until required. These 'pre-season samples' were otherwise treated as

described above. The results (Figure 2.17) showed that several samples had increased incidence of bulb-scale mite symptoms above control levels, though to a variable extent. There were also a few leaves and stems with symptoms in the control treatment.

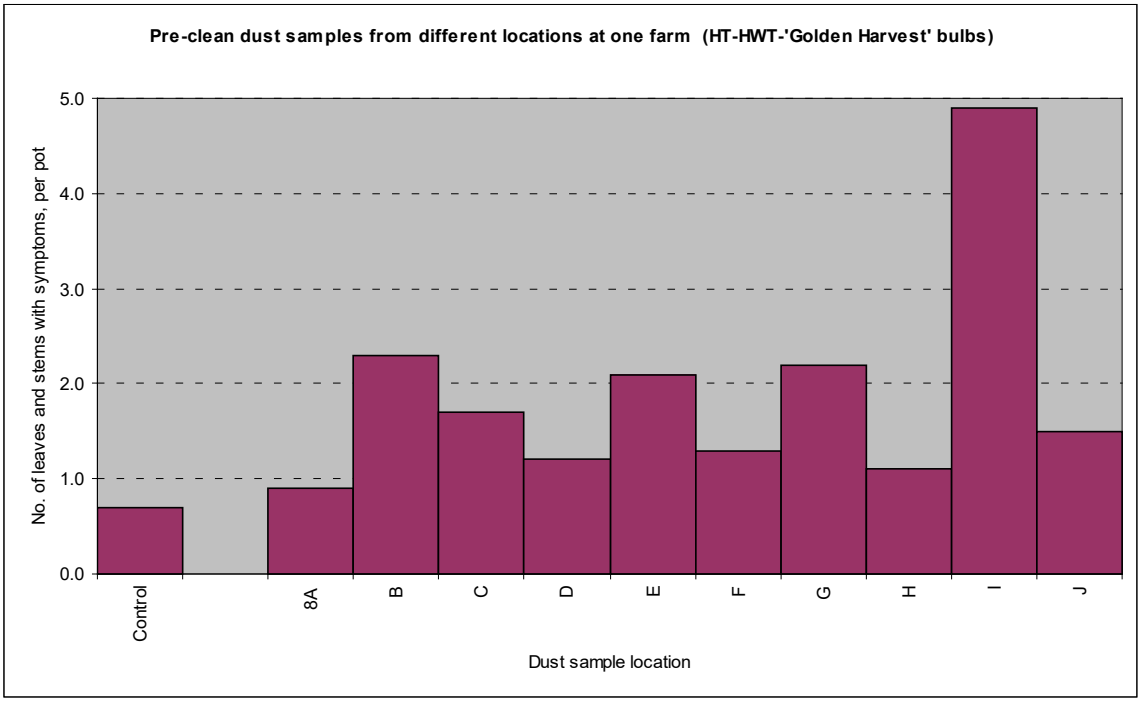


Figure 2.17: The number of leaves and stems with symptoms of bulb-scale mite infestation from dust samples taken from different drying floors prior to seasonal cleaning

2008 and 2009

The transmission of infestations via dust and debris was further investigated in 2008. In August, dust samples were collected from two Lincolnshire and two Cornish sites that had produced infective samples in 2008. At each site samples were collected from four locations at each site, the drying wall, the grading (or mixing) line, the storage area for bulbs awaiting despatch, and the storage area for bulbs for replanting. Each of these locations was divided into three sections, and samples were collected from each section, making 48 samples in all (four farms x four locations x three sub-locations). The samples were tested using similar procedures to those of 2007. The bulbs were planted in pots on 14 October 2008, grown-on in an unheated mesh tunnel, and transported to Wellesbourne in February 2009. The pots were kept in a glasshouse at Wellesbourne and shoots were sampled for the presence of live mites. No mites were found.

In 2009, dust samples were taken from four farms x four locations x three sub-locations, and un-infested bulbs (cv Kerensa) were incubated with the dust as described above.

These were potted up and the bulbs were assessed for live mites in 2011. No bulb-scale mites were found in the bulbs. A few of the bulbs were infested with bulb mite.

Are growers re-planting infested bulbs?

The information on the spatial distribution of mite-damaged bulbs within commercial crops, and the frequency with which eggs and live bulb-scale mites were found in bulbs that had undergone HWT in experiments, suggested that mites were either surviving HWT and (or) bulbs were being re-infested after HWT and prior to planting. In summer 2009 consortium members and other growers in Cornwall and eastern England were asked to provide samples of bulbs taken immediately after HWT, and, secondly, from the same stocks on the date they were planted following any intervening storage.

The growers were provided with a detailed protocol for collecting the bulb samples, and were asked to provide basic details of their HWT and associated procedures (Table 2.9). The sampling protocol supplied to the growers specified that, as each selected load was removed from the HWT tank and only the initial draining-down had taken place, two bulbs should be taken at random from just under the top layer of each of five bulk bins (ten bulbs in total per stock). Each ten-bulb post-HWT sample was to be placed in the labelled mesh bag provided and stored in a clean place away from the immediate vicinity of HWT and bulb storage facilities, while allowing the bulbs to cool, ventilate and surface-dry before despatching to Wellesbourne. When each of the same stocks was planted, a second ten-bulb 'planting sample' was to be taken from each, and the treatment of these samples was to be similar to before.

In all 76 samples were received from five farms in Cornwall and three in Lincolnshire. On arrival at Wellesbourne they were unpacked and stored as received in a site away from obvious contamination and with temperatures not lower than 17°C. Over the period from September 2009 to January 2010 five bulbs from each sample were (a) cut to record the intensity of feeding marks and (b) fully dissected to determine the numbers of bulb-scale mites (eggs, live adults and dead adults) and other mites in the old, current and new bulb units and in the neck of the bulb. The aim was to sample a further five bulbs at a later date, but sampling was so slow a process that this was not feasible. The remaining bulbs were, however, grown-on in pots outdoors, and the numbers of mites and eggs on the shoots in pots of stocks where live mites were found previously were recorded in spring 2010.

In summer 2010 further samples were collected from 13 of the Cornish stocks in which bulb-scale mite adults and (or) eggs had been found at planting in 2009. Samples of five bulbs from each stock were assessed.

Table 2.9: The log sheet provided to growers

Log sheet to be completed by growers
(This information is for research staff use only and no information in it will be connected to individuals or individual firms)

Name:

Business and farm name:

	Stock 1	*Stock 2*	*Stock 3*	*Stock 4*	*Stock 5*
Variety name					
Stock number (if any)					
Week stock was lifted	Week no.:	Week no.:	Week no.:	Week no.:	Week no.:
Any chemical treatment at/after lifting? (give details)					
Drying routine (ambient/high temperature, loose bulk/bins)					
Storage conditions once dry (temperature, loose bulk/bins)					
Date of HWT	___/____/ 2009	___/____/ 2009	___/____/ 2009	___/____/ 2009	___/____/ 2009
Any pre-warming? (give details)					
Any pre-soaking? (give details)					
Temperature of HWT	°C	°C	°C	°C	°C
Duration of HWT	hours	hours	hours	hours	hours
Chemicals used in HWT					
Drying method immediately after HWT					
Drying and (if any) storage conditions subsequently					
Planting date	___/____/ 2009	___/____/ 2009	___/____/ 2009	___/____/ 2009	___/____/ 2009
Any further comments?					

Some of the samples, both immediately after HWT and at planting, contained dead bulb-scale mites, sometimes in large numbers, but the key finding was that a few also contained live bulb-scale mites. Some bulbs also contained mite eggs, although it was impossible to determine whether these were alive or dead. There was considerable variation in mite numbers between different bulbs, even between those from the same batch or treatment. This was not unexpected, considering the experiences of the previous sampling exercises. In addition, there were few bulb-scale mites in samples from Lincolnshire, compared with those from Cornwall, confirming the results of earlier samples

Table 2.10 summarises the overall data for the two regions. Because of the small sample size and the very variable distribution of mites, care must be taken no to over-interpret the data. In the Lincolnshire samples no feeding marks were found, suggesting there had been no history of bulb-scale mite infestation. Adult bulb-scale mites were found only very exceptionally, and only in the neck of the bulb, and no adults were found when shoots were assessed later. Small numbers of eggs were found, but again, only in the bulb necks: there was an average of 0.1 eggs/shoot in samples taken after HWT, and of 0.4 eggs/shoot in samples taken at planting, but it was impossible to determine whether these eggs were viable or not. With the small size of the samples, it is unlikely that the difference between the pre- and post-HWT averages was statistically significant.

In contrast, in the Cornish samples feeding marks were often found, suggesting a history of infestation. Both adults and eggs were found in post-HWT and planting samples; they occurred in both the 'old' and 'current' bulb units and in the bulb neck, but were very rare in the 'new' bulb units. The total average numbers of adults and eggs per bulb in HWT samples were 0.36 and 1.75, respectively; in planting samples the numbers were higher: 0.54 adults and 3.38 eggs per bulb. When shoots were assessed the next spring, no adults were found, but there were averages of 3.40 eggs per shoot in the HWT sample and 43.33 eggs per shoot in the planting sample. Again, with the small sample size, it is unlikely that this difference was statistically significant. It was not possible to identify the eggs to species.

Table 2.10: Average numbers of bulb-scale mites in bulb samples from Lincolnshire and Cornwall taken after HWT or at planting

County	Sampling time	No. of 5-bulb samples	Feeding-marks (score) and bulb-scale mites (no./bulb) by location, autumn 2009											Live bulb-scale mites (no. /shoot), spring 2010 – selected samples	
			Score (0-5)	Live adult bulb-scale mites					Bulb-scale mite eggs						
				BU08	BU09	BU10	Neck	Total	BU08	BU09	BU10	Neck	Total	Adults	Eggs
Lincs	HWT	9	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.13	0.13	0.00	0.00
	Planting	9	0.00	0.00	0.00	0.00	0.01	0.01	0.00	0.00	0.00	0.36	0.36	0.00	0.00
Cornwall	HWT	28	0.26	0.02	0.18	0.00	0.16	0.36	0.23	0.40	0.01	1.11	1.75	0.00	3.40
	Planting	30	0.09	0.34	0.08	0.00	0.12	0.54	1.33	0.75	0.00	1.31	3.38	0.00	43.33

Assessments of bulbs from each of the five Cornish growers are shown in Table 2.11. Feeding marks occurred regularly in bulbs from all farms except Grower A, suggesting a generally widespread occurrence of bulb-scale mite in the county. Adult bulb-scale mites were found in HWT samples from four of the five growers, with numbers per bulb ranging from 0.07 to 0.96; in planting samples mites were found in samples from all five growers, with from 0.02 to 3.67 per bulb (Figures 2.18 and 2.19). There was little here to suggest a

re-introduction of mites into bulbs following HWT. Bulb-scale mite eggs were found in varying numbers in HWT and planting samples from all growers except Grower A. The numbers of mites could rise or fall between HWT and planting (Figures 2.20, 2.21 and 2.22). There was no obvious correlation of adult and egg numbers at the different farms. Adults and eggs occurred in BU08 and BU09 ('old' and 'current' bulb units) and in the bulb necks, but not in BU10 (the 'new' bulb units).

In shoots assessed the following spring, no adult bulb-scale mites were found. No or very few eggs were found, except at one farm (Grower D) where there were 2.80 eggs per shoot in the HWT sample and 26.00 eggs per shoot in the planting sample. It was not possible to identify the eggs to species.

Table 2.11: Average numbers of bulb-scale mites, etc., in bulb samples from five Cornish growers taken after HWT or at planting

Grower	Sampling time	Feeding-marks (score) and bulb-scale mites (no./bulb) by location, autumn 2009												Live bulb-scale mites (no. /shoot), spring 2010 – selected samples		Other mites (no./bulb), summer 2010		
		Score (0-5)	Live adult bulb-scale mites (and additional dead mites – score only)						Bulb-scale mite eggs							Live bulb mites	Live 'hairy mites'	Mite eggs
			BU08	BU09	BU10	Neck	Total	Dead*	BU08	BU09	BU10	Neck	Total	Adults	Eggs			
A	HWT	0.02	0.02	0.94	0.00	0.00	0.96	+	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0	0	0
	Planting	0.00	0.00	0.00	0.00	0.02	0.02	0	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0	0	0
D	HWT	0.52	0.00	0.00	0.00	0.00	0.00	++	1.04	1.84	0.00	3.70	6.58	0.00	2.80	0	0	0
	Planting	0.04	0.00	0.00	0.00	0.04	0.04	++	3.90	1.08	0.00	2.36	7.34	0.00	26.00	0	0	0
E	HWT	0.24	0.00	0.00	0.00	0.07	0.07	+++	0.00	0.00	0.00	0.04	0.04	0.00	0.00	4	0	0
	Planting	0.27	3.13	0.00	0.00	0.53	3.67	+++	0.00	2.00	0.00	0.00	2.00	0.00	0.00	4	0	0
G	HWT	0.27	0.00	0.00	0.00	0.12	0.12	+	0.15	0.05	0.05	0.20	0.45	0.00	0.50	26	0.5	8
	Planting	0.08	0.00	0.02	0.00	0.00	0.02	0	2.90	0.00	0.00	3.03	5.93	0.00	0.00	11	0.5	8
H	HWT	0.26	0.04	0.03	0.00	0.41	0.49	+	0.04	0.20	0.00	1.24	1.49	0.00	0.00	12	0	0
	Planting	0.11	0.07	0.22	0.00	0.15	0.44	+	0.27	1.00	0.00	0.84	2.11	0.00	0.00	11	0	0

* 0, dead bulb-scale mites not seen, or +, ++ and +++, seen in <50%, ≥50% or 100% of samples

The number and distribution of bulb-scale mites varied considerably between the five growers:

- A: low populations and of adults only, found only in HWT samples
- D: some high populations; very few adults but many eggs, and overall similar numbers in the HWT and planting samples, though distribution through the bulbs varied
- E: some high populations, with adults in planting samples only and more eggs in the HWT sample (virtually all in the bulb neck)
- G: very variable populations, with few adults, mostly in the neck of HWT samples, and many more eggs in planting than HWT samples and mostly in the neck and BU08 ('old' bulb units)

- H: generally low populations, with similar numbers of adults in HWT and planting samples but more eggs in the planting samples than HWT samples; all well distributed through the BU08 and BU09 ('old' and 'current' bulb units) and the bulb neck.

The husbandry practices of the Cornish growers revealed some notable differences, though it would be worthwhile seeing if it is possible to correlate farm practice with mite numbers. The main differences in husbandry were as listed here.

- Grower A was unusual in being located near the eastern border of Cornwall, so would experience a somewhat less maritime climate than the other growers (who were all in west Cornwall).
- All five growers pre-warmed their bulbs (a treatment used in Cornwall to reduce the damage to flower buds caused by HWT), but growers E and G did not follow this by the conventional 'pre-soak' before HWT (pre-soaking ensures the bulbs are fully hydrated before HWT).
- Grower D was unusual in including chlorpyrifos (an insecticide to control large narcissus fly, which also has acaricidal properties) in his pre-soak.
- Grower E reported using a harsher HWT regime (4 hours at 47°C, expected to be more effective in controlling pests in the and dip) than the other growers; grower A also used a 4-hour HWT, but at the lower temperature used by the other growers (45 to 46°C).
- Unusually, growers E and G did not generally add any pesticides or biocides to their HWT dip. The other growers included a fungicide, and grower D also added an acidifier (to enhance the effects of thiabendazole fungicide), and they always pre-soaked bulbs before HWT.
- There were very variable intervals between the dates of HWT and planting in the different stocks and at different growers.

Although the sample sizes were small and care is needed in interpreting the results, by comparing differences in practices and the numbers of mites found, it could be suggested that:

- The very low mite numbers in bulbs from grower A could be related to his location, reinforcing the notion that bulb-scale mite is mainly a problem in the warmer, more maritime, South-West of the county
- Growers E and G had some commonality in husbandry, i.e. they did not pre-soak bulbs and did not include pesticides or biocides in HWT; nevertheless, the numbers of mites in their respective samples were dissimilar

- Grower D, who included the insecticide chlorpyrifos in the pre-soak, still had some high populations of mites in bulb samples
- Grower D also added acidifier to his HWT tank, which did not appear to affect mite numbers
- Grower E used a hotter, longer HWT than the other growers, but nevertheless his bulbs still showed some high numbers of mites
- Despite the diverse intervals between the dates of HWT and planting in the different stocks and at different growers (from 1 to 49 days), the change in mite numbers between HWT and planting – a possible period for re-contamination – were mostly slight. This is shown for grower H's bulbs in Figure E.

Bulb mites, as opposed to bulb-scale mites, were found in bulbs from growers E, G and H. Occasional mite eggs (probably bulb mite) and individuals of another mite species ('hairy mite') were also noted in samples from grower G.

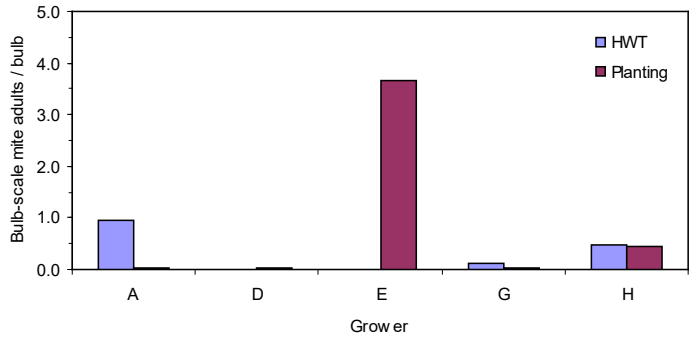


Figure 2.18: Numbers of live bulb-scale mite adults (mean per bulb) at the end of HWT and at planting for bulb stocks from five Cornish growers

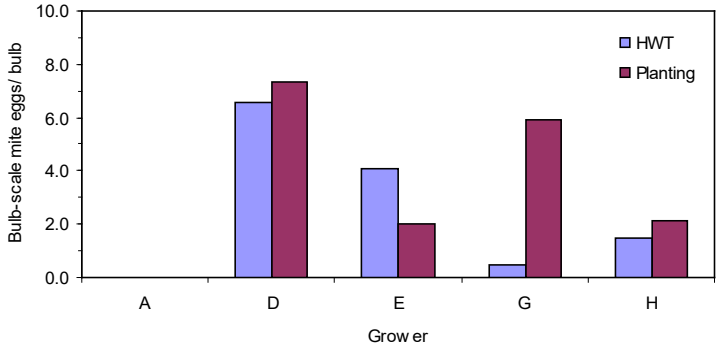


Figure 2.19: Numbers of bulb-scale mite eggs (mean per bulb) at the end of HWT and at planting for bulb stocks from five Cornish growers

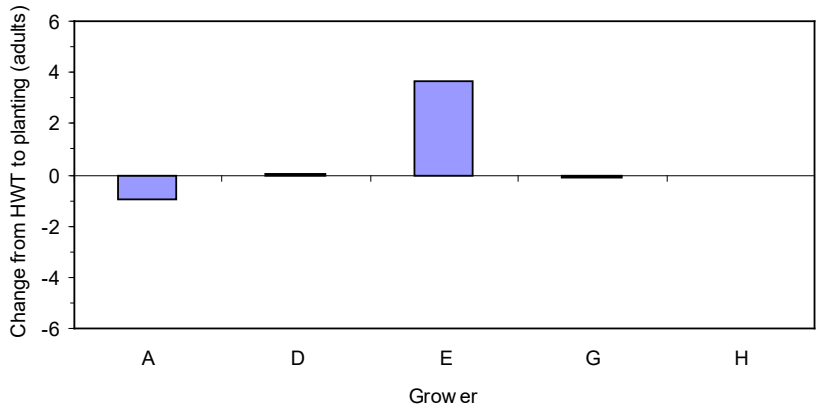


Figure 2.20: The mean change in numbers of live bulb-scale mite adults between HWT and planting for bulb stocks from five Cornish growers

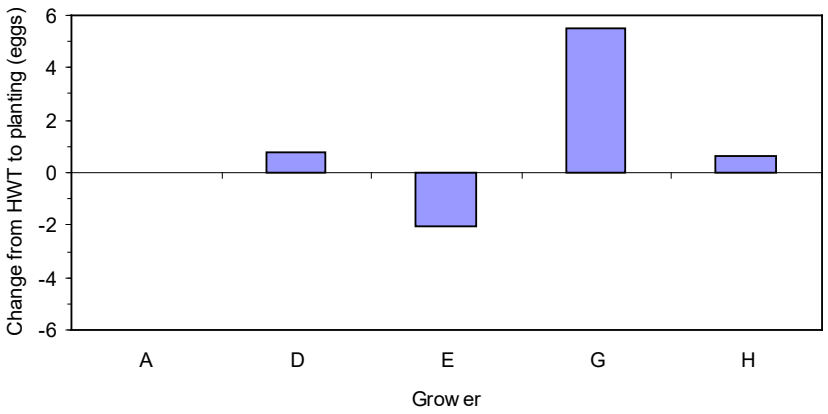


Figure 2.21: The mean change in numbers of bulb-scale mite eggs between HWT and planting for bulb stocks from five Cornish growers

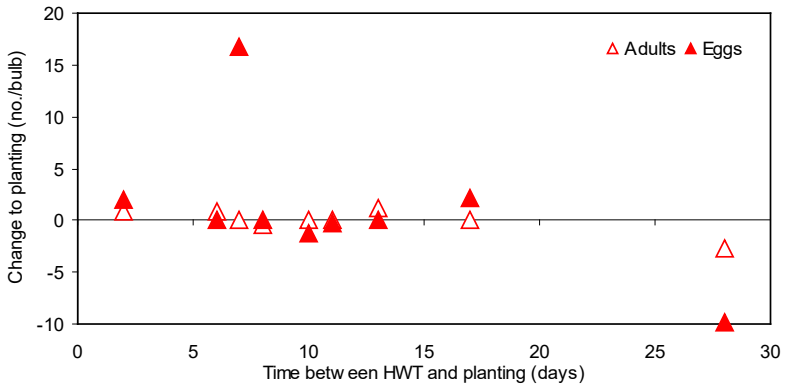


Figure 2.22: The changes in numbers of live bulb-scale mite adults and eggs between HWT and planting for nine stocks from grower H, plotted against the interval between HWT and planting

Despite the 2009 findings, no bulb-scale mites or eggs were found in any of the 2010 samples: however, in nine of the 13 stocks the feeding mark score had increased, though by only 0.3 on average, which could simply reflect continuing tissue damage as a result of earlier mite activity. Vindicating the 'dissect and search' assessment strategy, however, bulb mites (usually with associated eggs) were found in bulbs from all 13 samples, sometimes in high numbers, and 'hairy mites' (and associated eggs) were found in three of the 13 samples.

Determine whether cultivars are differently susceptible to bulb-scale mite and whether bulb-scale mite show cultivar preferences

2007-8

'Golden Harvest' bulbs (from Kirton stocks) and bulbs of cultivars 'Counsellor', 'Dutch Master', 'California', 'St Keverne', 'Golden Ducat', 'Ice Follies' and 'Carlton' (supplied by a Consortium member) were used as test bulbs. This was a mix of popular cultivars including various possible susceptibilities and what was available at the time from the industry partners.

The supplied bulbs were not specially treated, and therefore variable degrees of infestation with bulb-scale mites would be expected. In August 2007, groups of nine test bulbs were placed into brown paper bags, and five mite-infested bulbs were added to half of the bags. The bags were stored at 17°C for 4 weeks (representing the storage phase when infestation could take place). For each cultivar there were eight pots with mite-infested bulbs and eight without infested bulbs. The bulbs were planted in pots of a peat growing medium by tipping out the contents of the bag onto growing medium in a 20 cm-diameter plant-pot, arranging the bulbs evenly, with the mite-infested bulbs in the centre of the pot, and topping-up with growing medium in a standard fashion. The pots were watered well and placed in a cold store at 9°C. When the bulbs of each cultivar had received a sufficiently long cold treatment they were moved to a glasshouse (details as above) for growing-on.

After 4 weeks in the glasshouse the incidence and severity of bulb-scale mite symptoms were recorded, as previously described. Only a low incidence of bulb-scale mite symptoms was seen on any of these plants, with no indication of differences between cultivars and the data are not presented here.

After foliar senescence the pots were moved to a ventilated, non-heated mesh tunnel and grown-on. In November 2008 bulbs were recovered from their pots, and sound (non-rotted)

bulbs of flowering size were cut transversely and scored for the presence and severity of feeding marks in the current bulb units (Figure 2.23). As there were no clear differences in the amount of symptoms for each variety planted with inoculator bulbs and those planted alone, it appeared that most feeding marks were the result of prior infestations, which naturally varied between varieties and stocks. It was seen that the feeding marks were almost always in the scales of the current bulb unit, which indicated that the addition of inoculator bulbs had been totally ineffective.

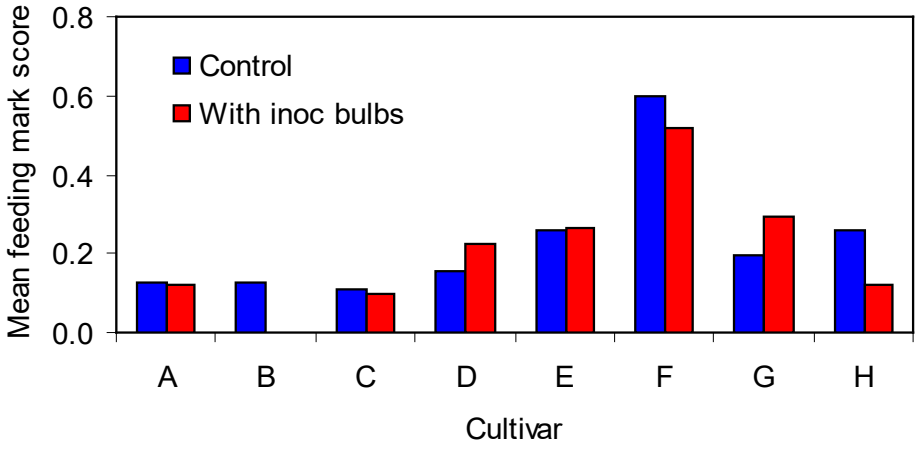


Figure 2.23: Mean feeding mark scores in eight narcissus varieties grown alone ('control') or with added inoculator bulbs ('with inoc bulbs'). Cultivars: A, Carlton; B, Golden Ducat; C, Dutch Master; D, Ice Follies; E, Counsellor; F, California; G, Golden Harvest and H, St Keverne

2008-9

In order to collect further data on varietal differences, and hopefully to explain the result obtained in 2007-8, this experiment was repeated in 2009. The same bulb stocks as before were used, with the addition of cv. Hollywood, but the bulbs of all stocks received standard HWT prior to set-up; in addition, Golden Harvest bulbs were tested following both standard HWT and HT-HWT. The UK standard is 3 hours x 44.4°C and the 'extra hot' treatment was 3 hours x 46.4C (originated from the earlier HWT experiment where a range of temperatures from 44.4 to 48.4°C were tested). The bulbs were planted in pots on 21 October 2008, placed in a 9°C cold store, moved to Wellesbourne in February 2009, and were examined in summer 2011. None of the bulbs contained bulb-scale mites, although several were infested with bulb mites.

Objective 3: Design optimal high or low temperature and/or chemical treatments to control bulb-scale mite in bulbs for replanting and for forcing, and ensure all stages in its life-history are killed and that crop quality is unaffected

Determine what HWT regimes are effective in controlling all stages of the bulb-scale mite life-cycle

2007

In 2007 mite-infested bulbs (cv. 'Carlton', 8-10 cm grade) were used to test the effects of HWT regimes. On 3 September netted groups of nine bulbs each were treated for 2, 3 or 4 hours at 42.4, 44.4 or 46.4°C, with a further group of bulbs remaining untreated as controls. This was repeated with fresh sets of bulbs on 4 and 5 September, giving three 'replicates' of each of the ten treatments. Following then-standard practice, the HWT dip contained formaldehyde (as commercial formalin), a prochloraz fungicide (as Mirage 40EC), non-ionic wetter and anti-foam preparation. To simulate commercial HWT conditions, the netted groups of bulbs were placed in HWT tanks fully loaded with stock bulbs (cv Carlton), and treatments were timed from when the netted bulbs were added to the tank, assuming this small volume of bulbs would warm up rapidly. After HWT the bulbs were removed from the tank, cooled and surface-dried by standing under strong ventilation, and stored at 17°C. The bulbs were planted on 17 September in 20 cm-diameter plant-pots and placed in a 9°C store until judged ready for forcing (11 January 2008), when the pots were moved to a heated glasshouse.

After 4 weeks in the glasshouse the incidence and severity of bulb-scale mite symptoms on leaves and stems were recorded. Very few symptoms were present, and only on control (non-HWT) bulbs (Table 3.1).

After a further 1 week in the glasshouse, two random shoots from each pot were excised and all leaf and stem surfaces were examined under a LP microscope for bulb-scale mites and eggs. There were numerous mites and eggs on all control plants, and a few on one only of the treated plants. After a further 4 weeks additional shoot samples, from the controls and the 3h x 44.4°C treatment only, were examined. The earlier result was confirmed (Table 3.1).

Table 3.1: BSM assessments of forced plants in the glasshouse, following different HWT regimes, 2007-2008 experiment (assessed spring 2008)

HWT	BSM symptoms (mean no. of leaves and stems per pot with symptoms, at week 4)	BSM/eggs on leaves (mean incidence score)	
		Week 5	Week 9
None	0.7	3.7	6.0
2h 42.4°C	0	0	-
3h 42.4°C	0	0.3	-
4h 42.4°C	0	0	-
2h 44.4°C	0	0	-
3h 44.4°C	0	0	0.3
4h 44.4°C	0	0	-
2h 46.4°C	0	0	-
3h 46.4°C	0	0	-
4h 46.4°C	0	0	-

-, not assessed

After foliar senescence the pots were moved to a ventilated, non-heated mesh tunnel and grown-on. Over several days in late-October 2008 bulbs were recovered from their pots. All bulbs were cut transversely and then scored for the presence and severity of feeding marks on the scales, also recording whether feeding marks were on the current, previous or new bulb units. Numerous feeding marks occurred on scales of both the previous and current bulb units (Figure 3.1). As expected, feeding marks were found on previous bulb units in both control and treated bulbs (since HWT cannot eliminate feeding marks made prior to treatment, but should control current mite activity). In the current bulb units the highest feeding mark score occurred on control bulbs, but bulbs from various hot-water treatments also showed some evidence of mite activity, indicating only a partial control of bulb-scale mite by the treatments had been achieved.

In addition, bulbs from three key treatments (the control, the standard HWT of 3h x 44.4°C, and the extreme HWT of 4h x 26.4°C) were fully dissected into individual scale pieces and all pieces were examined under a LP microscope for the presence and numbers of bulb-scale mites and eggs. Table 3.2 shows the distribution of feeding marks and bulb-scale mites to the generations of bulb units. The data confirmed that both current and previous bulb units contained feeding marks, that those in the current scales were fewer where HWT had been given, and that no feeding marks occurred in the new bulb units at this stage. However, in new bulb units bulb-scale mites were sometimes found without accompanying feeding marks; to a lesser extent this effect was also seen in current bulb units. Table 3.3 shows the numbers of scale pieces with feeding marks and mites, and the total numbers of

mites. It confirms that there were more feeding marks in control bulbs than in treated bulbs. However, one of the two HWT treatments examined contained active and inactive mites and mite eggs, like the control, suggesting that new infestations had taken place by the autumn of the year following HWT. In this particular case, the number of mites and eggs was skewed by very high numbers of mites and eggs in one of the replicates.

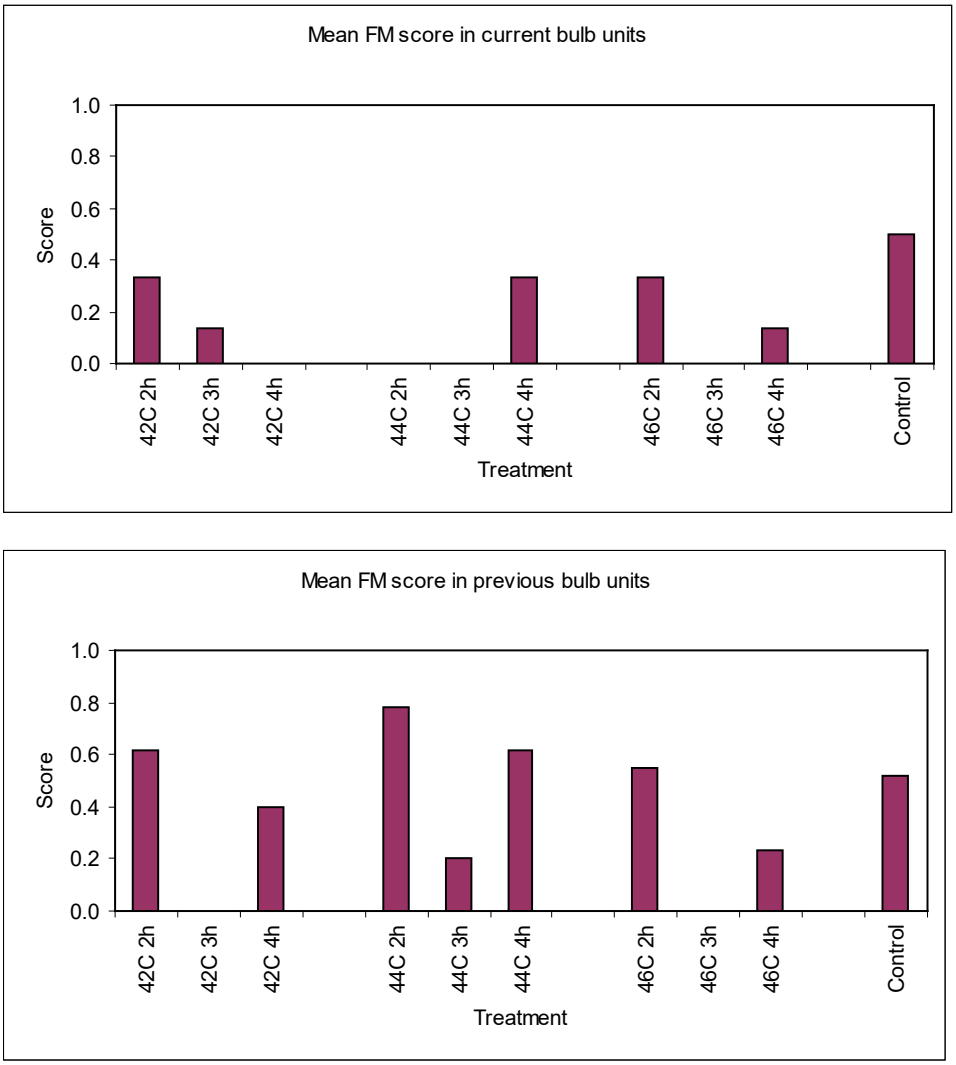


Figure 3.1: Feeding mark scores in current (top) and old bulb units (bottom) of bulbs previously given HWT at the temperatures and durations stated. Data from the 2007-2008 experiment assessed in autumn 2008

Table 3.2: Bulb-scale mite assessments of plants grown-on after glasshouse forcing following key HWT regimes, 2007-2008 experiment (assessed autumn 2008)

No. of scales per bulb with:	Bulb unit	44C 3h	46C 4h	Control
Feeding marks	Previous	0.27	0.73	2.18
	Current	0	0.27	2.62
	New	0	0	0
BSM but no feeding marks	Previous	0	0	0
	Current	0	0.17	0
	New	0	1.33	0.83

Table 3.3: BSM assessments of plants grown-on after glasshouse forcing, following different HWT regimes, 2007-2008 experiment (autumn 2008)

HWT	Number of scale pieces with:				Number of mites		
	Feeding marks	Active BSM	BSM eggs	Inactive BSM	Active BSM	BSM eggs	Inactive BSM
44°C 3h	0.27	0.00	0.00	0.00	0.0	0.0	0.0
46°C 4h	0.93	2.00	0.83	0.17	57.2	11.0	0.2
Control	4.80	1.67	0.83	0.67	16.2	9.0	2.0

2008

The HWT experiment was repeated in 2008, using infested bulbs of cv. Dutch Master. As in 2007, netted groups of bulbs (ten each) were treated for 2, 3 or 4 hours at 42.4, 44.4 or 46.4°C or remained untreated as controls; the three replicates were treated on 28 August, 29 August and 1 September 2008. The bulbs were planted on 12 September, methods as before, placed in a 9°C store until judged ready for forcing (29 December 2008) and then moved to a heated glasshouse.

After 4 weeks in the glasshouse the incidence and severity of bulb-scale mites symptoms on leaves and stems were recorded. Mite symptoms were common on control plants but occurred only rarely on treated bulbs (Table 3.4). These bulbs were re-located to a glasshouse at Wellesbourne on 3 February 2009 and then shoots were removed from each plant (by severing them at the top of the bulb) and examined for mites. The results are summarised in Figures 3.2-3.4. There were no live mites in shoots from hot-water-treated bulbs, although there were live mites in the shoots taken from the untreated bulbs.

Table 3.4: Bulb-scale mite damage symptom assessments of forced plants in the glasshouse, following different HWT regimes, 2008-2009 experiment (assessed spring 2009)

HWT	BSM symptoms (mean no. of leaves and stems per pot with symptoms)
None	6.0
2h 42.4°C	0.0
3h 42.4°C	0.0
4h 42.4°C	0.0
2h 44.4°C	0.0
3h 44.4°C	0.7
4h 44.4°C	0.3
2h 46.4°C	0.0
3h 46.4°C	0.7
4h 46.4°C	0.0

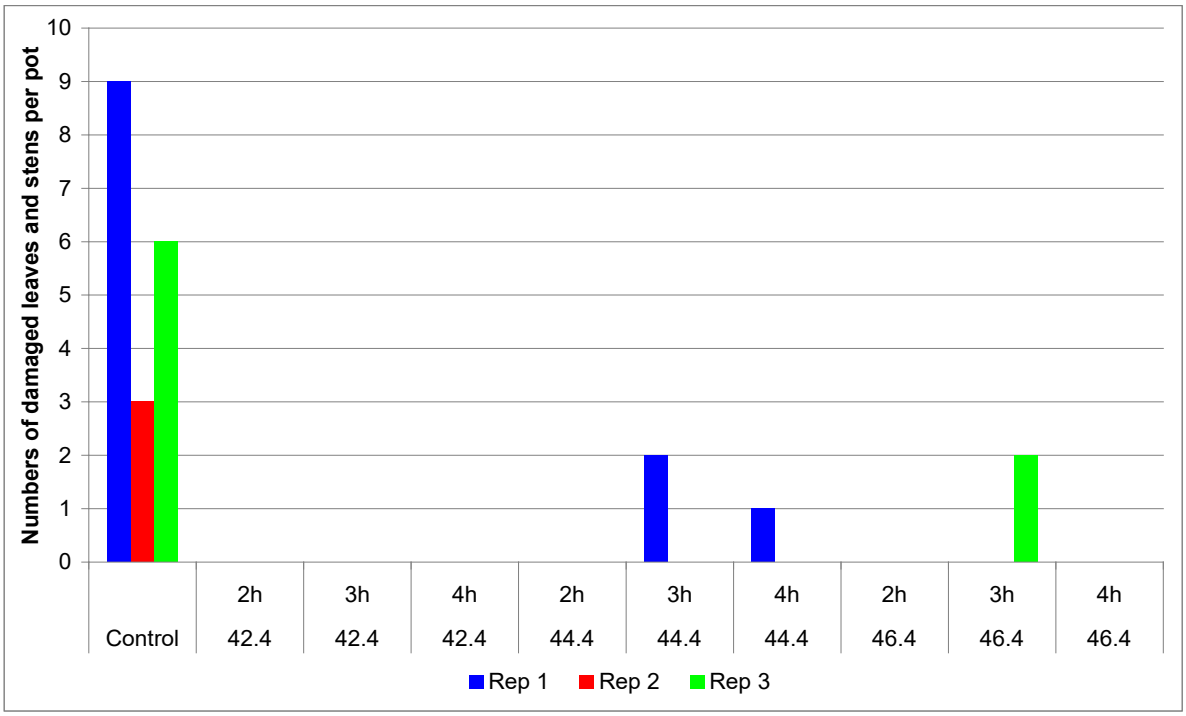


Figure 3.2: Bulb-scale mite damage symptom assessments of forced plants in the glasshouse, following different HWT regimes, 2008-2009 experiment assessed 3 February 2009

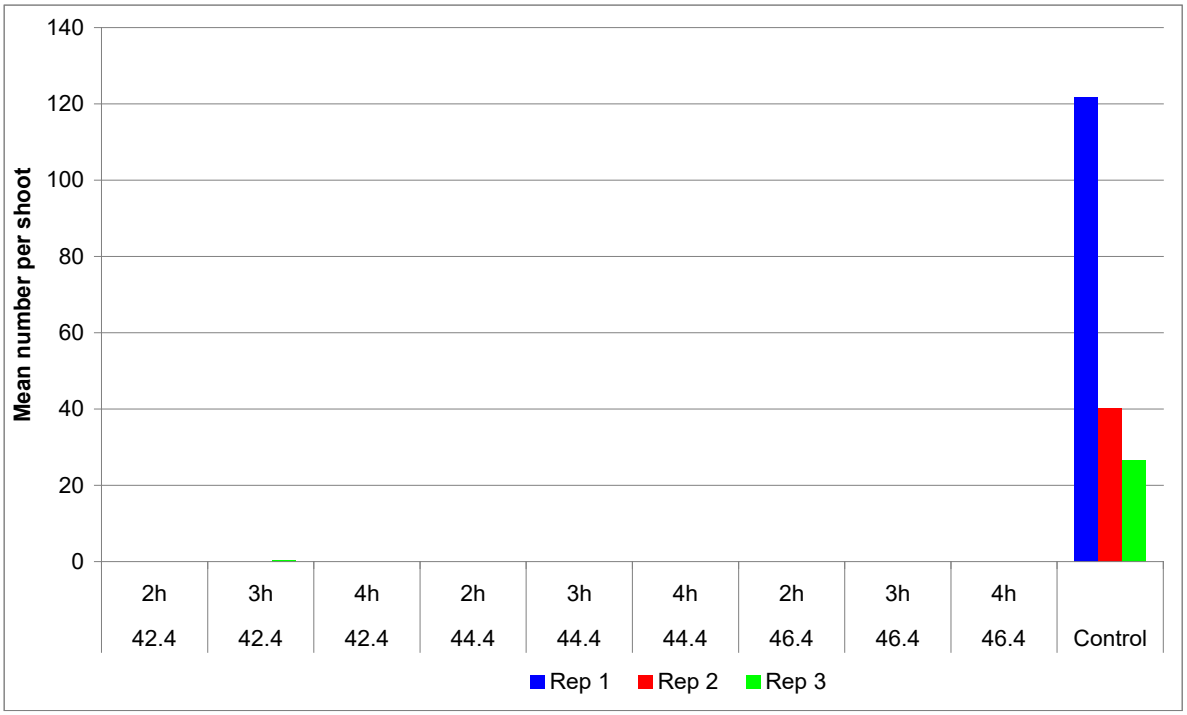


Figure 3.3: Number of live mites per shoot from bulbs previously given HWT in 2008 at the temperatures and durations stated

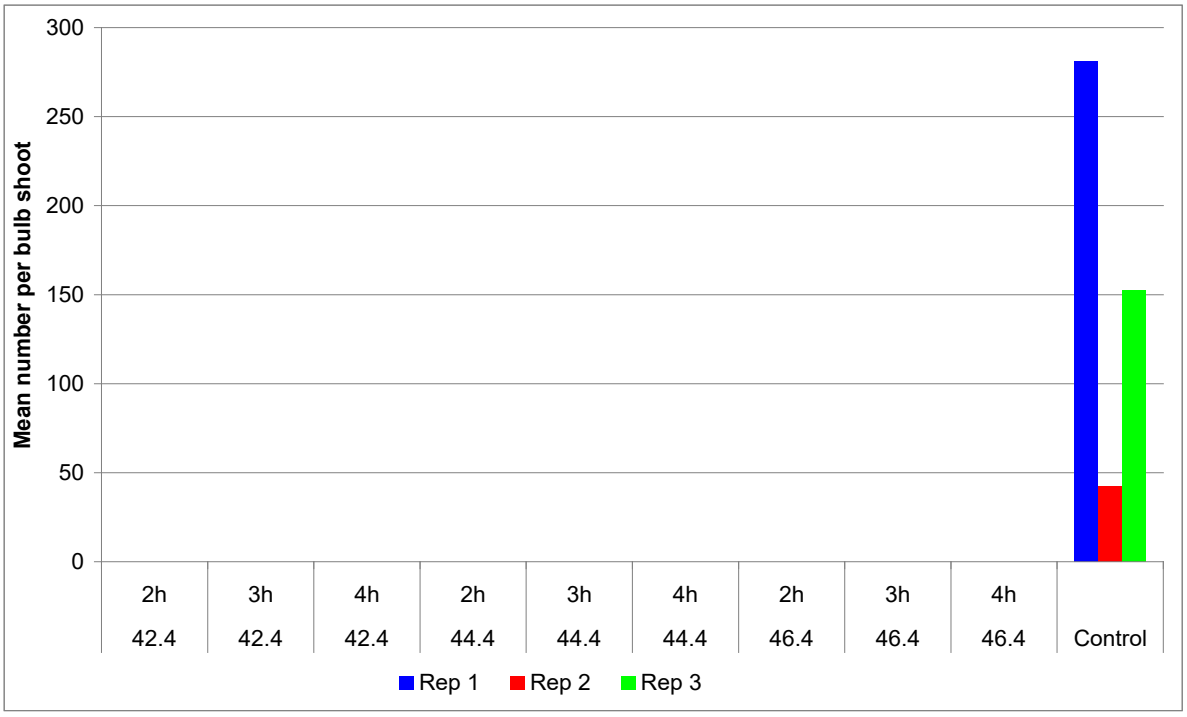


Figure 3.4: Number of eggs per shoot from bulbs previously given HWT in 2008 at the temperatures and durations stated

2009

A further HWT trial was undertaken in 2009. Mite-infested Dutch Master bulbs from the plot at Wellesbourne were treated by a consortium member for 3 hours at 44.4°C with the following dip additives:

- Treatment 1 – no dip additives
- Treatment 2 – FAM30 (4L/1000L water)
- Treatment 3 - FAM 30 (8L/1000L water)
- Treatment 6 - FAM 30 (4L/1000L water) + Bravo 500 (0.5L/1000L water)
- Untreated (n HWT)

The bulbs were returned to Wellesbourne in the week beginning 21 September and then 10 pots x 8 bulbs from each treatment were potted up and stood outside in the Dutch Lights area. They were later moved to a polytunnel to protect them from extreme cold and assessed in April 2010. Live mites and eggs were found in samples from the control treatment. Live mites, but no eggs, were found in samples from three of the other treatments (Figures 3.5-3.6).

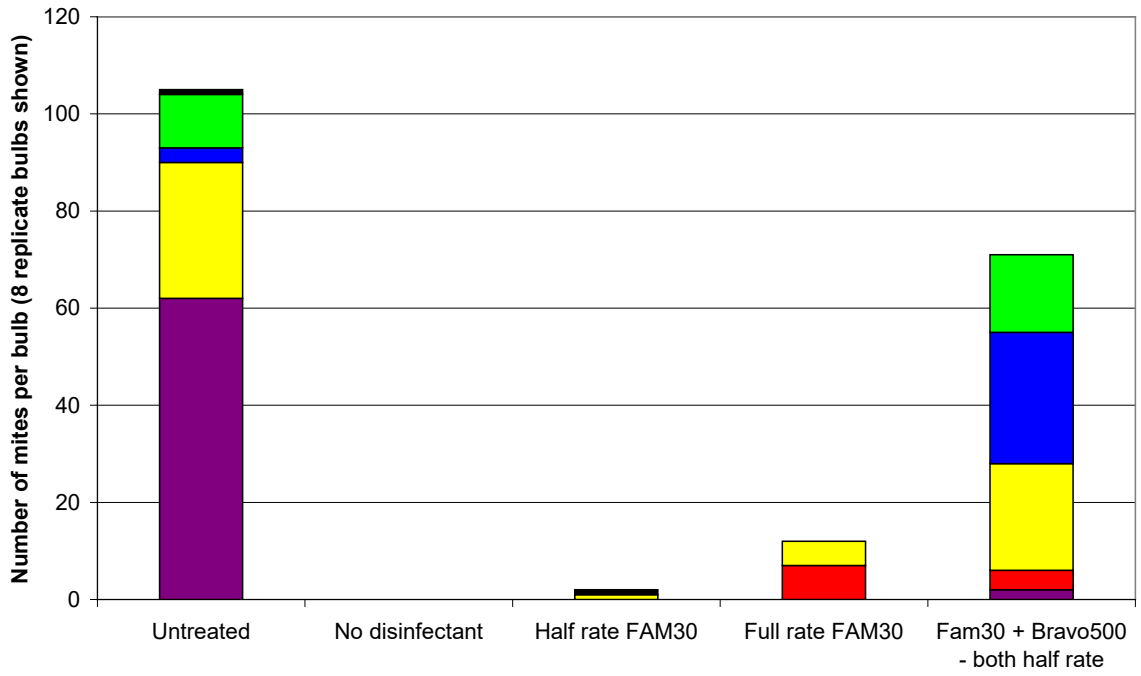


Figure 3.5: Number of live mites per shoot from bulbs previously given HWT in 2009 with the treatments indicated

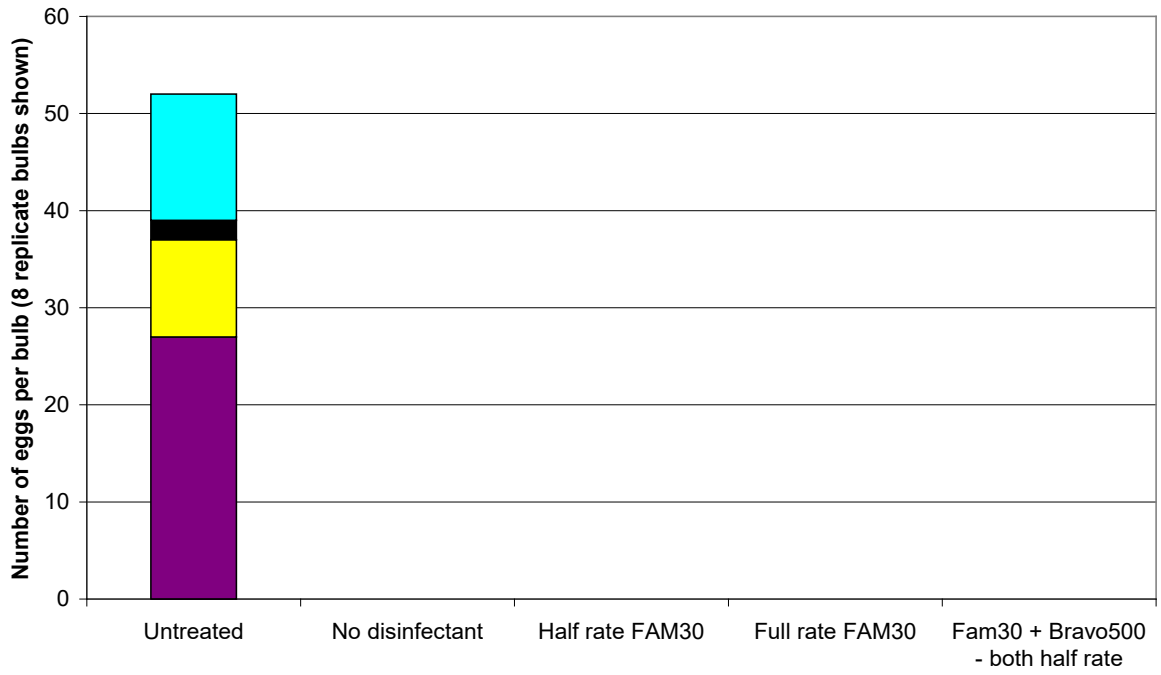


Figure 3.6: Number of eggs per shoot from bulbs previously given HWT in 2009 with the treatments indicated

Determine whether warm-storage treatments are effective in controlling all stages of the bulb-scale mite life-cycle without causing damage to glasshouse-forced bulbs

2007

Bulbs of a mite-infested stock (cv. Carlton, 8-10 cm grade) were used in this trial at Wellesbourne in 2007. Bulbs were treated in netted groups of nine bulbs each, with three replicates (occasions) of each treatment. The treatments were storage for 1, 2 or 3 hours at 42, 44 or 46°C, with further untreated bulbs serving as controls. The three replications were carried out on 2, 3 and 4 October 2007, respectively. Treatments were applied in a laboratory fan oven (Gallenkamp Size 2 'Hotbox', 0.125m^3 capacity) calibrated using a separate thermometer. The bulbs, in nets, were put in the oven on a wooden board, and treatments were timed from when the oven re-gained the target temperature after closing the door. After treatment, the bulbs from each treatment and replicate were put into separate paper bags (to prevent possible transfer of mites between treatments) and they were then transported to the Kirton Research Centre where they were planted in 20 cm-diameter pots with the nine treated bulbs planted around the edge of the pot and five previously hot-water treated Golden Harvest test bulbs were planted in the centre. The pots were cold-stored and moved to a heated glasshouse on 29 February 2008.

After 4 weeks in the glasshouse the incidence and severity of bulb-scale mite symptoms on leaves and stems were recorded (Table 3.5). There was a very low incidence of symptoms, and hence no evidence for a beneficial effect of warm storage.

After a further 1 week in the glasshouse, two random shoots from each pot were excised and all leaf and stem surfaces were examined under a LP microscope for bulb-scale mite and eggs (Table 3.5). There were numerous mites and eggs on control (untreated) plants, and fewer on the warm-stored bulbs, probably indicating a measure of control by these warm storage treatments.

Table 3.5: Bulb-scale mite assessments of forced plants in the glasshouse, following different warm storage treatments, 2007-2008 experiment assessed spring 2008. The figures given are the marginal means for temperature treatments (i.e. means across all treatment durations)

Heat treatment	BSM symptoms on leaves (mean no. of leaves per pot with symptoms)	BSM/eggs on leaves (mean incidence score)
Control (none)	0	2.7
42.0°C	0.2	0.3
44.0°C	0.1	1.0
46.0°C	0.4	0.7

After foliar senescence the pots were moved to a ventilated, non-heated mesh tunnel and grown-on. In November 2008 the bulbs were recovered from their pots, cut transversely and scored for the presence and severity of feeding marks on the scales, also recording whether feeding marks were on the current, previous or new bulb units. However, only two bulbs were found with feeding marks in the current bulb units, and so the data are not presented.

2008

The warm-storage experiment was repeated in 2008, using infested bulbs of cv. Dutch Master. As in 2007, netted groups of bulbs (10 each) were treated for 1, 2 or 3 hours at 42, 44 or 46°C or remained untreated as controls. There was a single treatment of 48 hours at -2°C. The three replicates were treated in the week beginning 6 October 2008. The bulbs were planted in the week beginning 13 October, kept in a glasshouse and shoots from each pot were sampled in April 2009. The numbers of mites per shoot are shown in Figure 3.7. None of the treatments appeared to have controlled the mites.

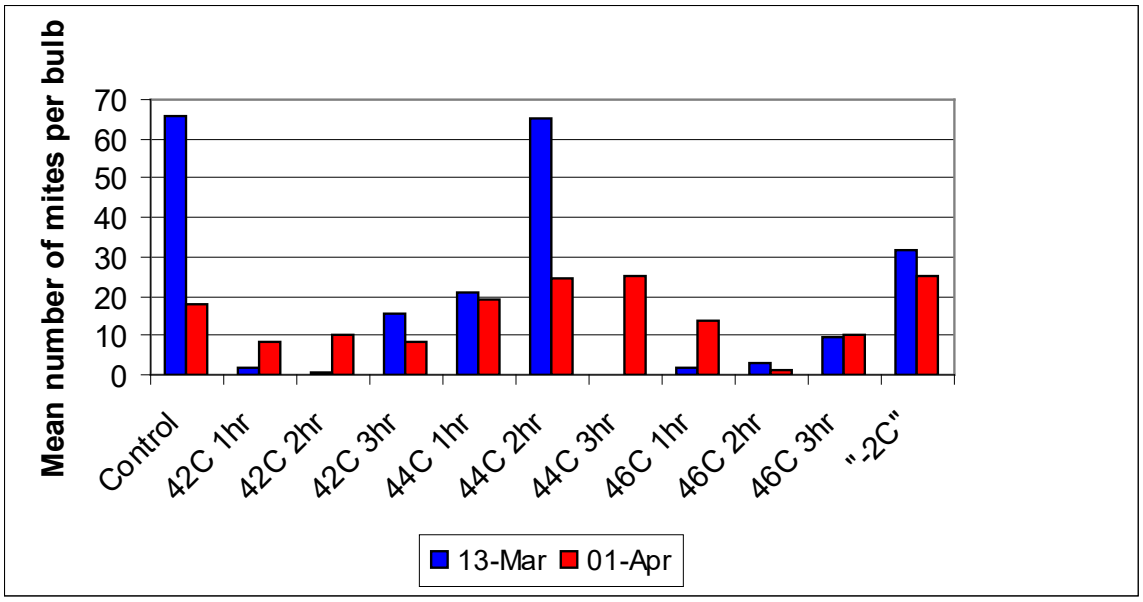


Figure 3.7: Number of live mites per shoot from bulbs previously given warm storage treatment in 2008 at the temperatures and durations stated

2009

Further 'extreme' warm storage treatments were applied to bulbs of a mite-infested stock (cv. Dutch Master) in a trial at Wellesbourne in 2009.

Bulbs were treated in netted groups of nine bulbs each, with three replicates (occasions) of each treatment. The treatments were storage for 2, 4 or 8 hours at 46, 50, 54°C, with further untreated bulbs (6 x 9 bulbs) serving as controls. The three replications were carried out in October 2009. Treatments were applied in a laboratory fan oven (Gallenkamp Size 2 'Hotbox', 0.125m^3 capacity) calibrated using a separate thermometer.

The bulbs, in nets, were put in the oven on a wooden board, and treatments were timed from when the oven re-gained the target temperature after closing the door.

After treatment, the bulbs from each treatment and replicate were put into separate paper bags (to prevent possible transfer of mites between treatments) and they were then planted in 20cm-diameter pots. These were kept outside and then moved to a polytunnel to protect them from extreme cold. Shoots taken from the bulbs were assessed in spring 2010. Mite eggs were found only on samples from the control treatment (Figure 3.8). However, the more extreme warm storage treatments (temperature and duration) had an adverse effect on the numbers of shoots produced by the bulbs (Figure 3.9)

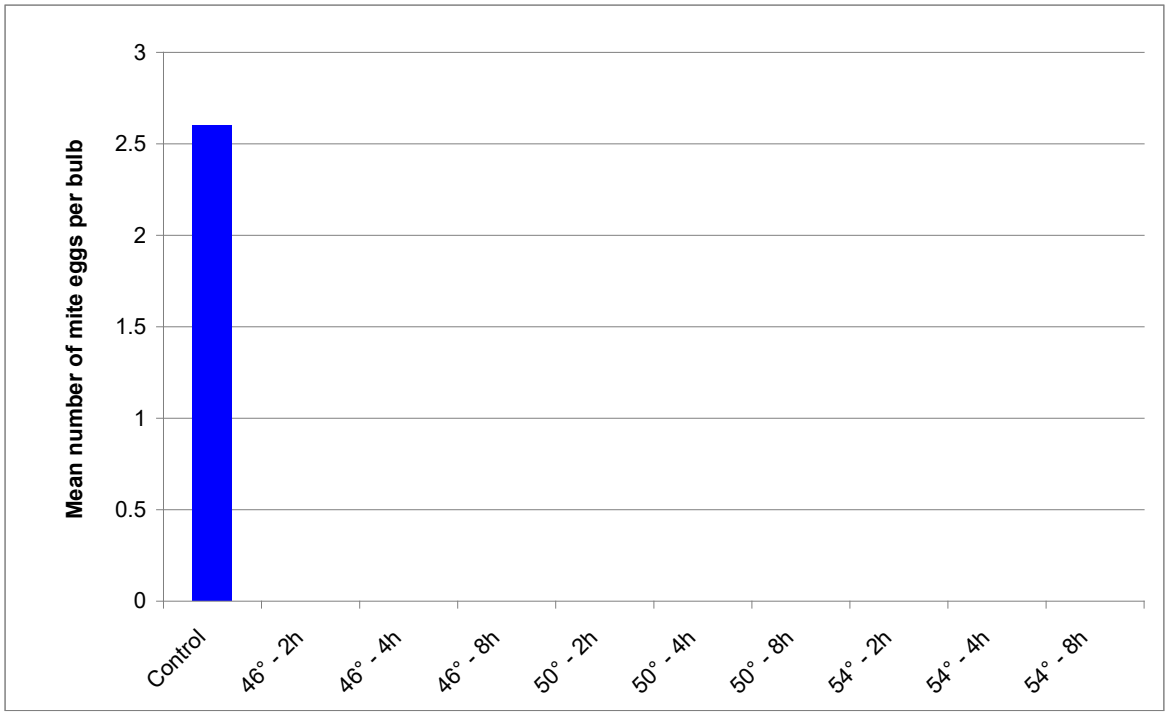


Figure 3.8: Number of mite eggs on shoots in March 2010 from bulbs previously given warm storage treatment in 2009 at the temperatures and durations stated

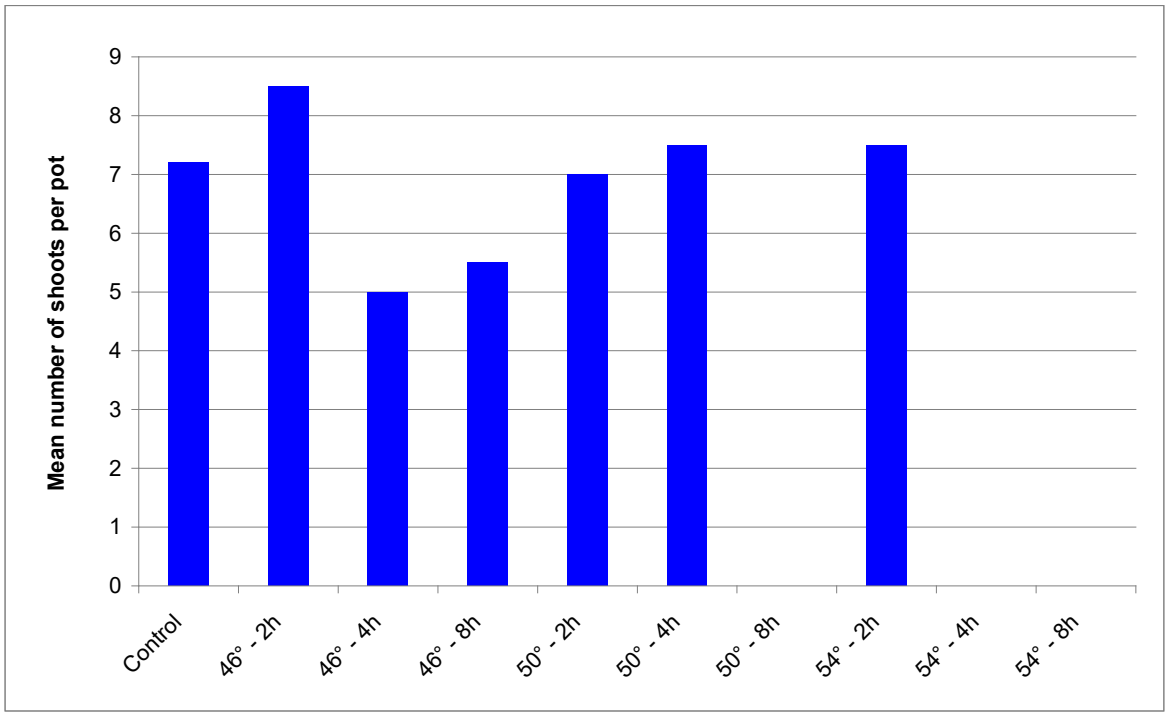


Figure 3.9: Number of shoots on 22 March 2010 from bulbs previously given warm storage treatment in 2009 at the temperatures and durations stated

Determine whether moving forced bulbs from the glasshouse to freezing temperatures overnight ('frosting') is effective in controlling bulb-scale mite without adversely affecting crop quality

2007

On 17 September 2007, groups of nine mite-infested bulbs (cv Carlton, grade 8-10 cm) were placed with five hot-water treated Golden Harvest test bulbs in paper bags (317 x 305 mm). Twenty-four bags were set up, the top of each bag was folded over, and the bags were placed in a controlled-temperature store at 9°C for 5 weeks. On 22 October 2007, the bulbs from each bag were planted in pots, the infected bulbs being planted around the edge of the pot and the five treated Golden Harvest test bulbs planted in the centre. The pots were returned to the 9°C cold store. On 8 January 2008, after a total of 15 weeks at 9°C, the pots were moved to a glasshouse. After 3 days in the glasshouse the pots were subjected to different 'frosting' treatments by placing them in a -1°C cold store for 12, 24 and 36h before being returned to the glasshouse. One set was non-frosted as a control. There were six replicate pots for each of the treatments and control.

After 4 weeks (5 February 2008) the incidence and type of foliar symptoms of bulb-scale mite were recorded (Table 3.6). A relatively low incidence of symptoms was seen at this examination, in all treatments.

After a further week in the glasshouse, random shoots were excised from each pot and the leaf/stem surfaces were examined under a LP microscope (Table 3.6). All six replicates of the control (untreated) bulbs carried several to many live bulb-scale mites. Of the 12 lots of bulbs frosted for 12 or 24 hours, half carried one to many mites. Half of the plants frosted for 36 hours carried one to a few mites. Mite eggs were found on only five pots from among the control plants and plants frosted for 12 hours. This suggests that the 'frosting' treatment has potential and could be further investigated.

Table 3.6: Bulb-scale mite (BSM) assessments of forced plants in the glasshouse, following different 'frosting' treatments, 2007-2008 experiment assessed spring 2008

Frosting treatment	BSM symptoms on leaves (mean no. of leaves per pot with symptoms)	BSM/eggs on leaves (mean incidence score)
Control (none)	0.7	4.3
12h	0.7	1.2
24h	0.2	1.8
36h	0.5	0.8

After foliar senescence the pots were moved to a ventilated, non-heated mesh tunnel and grown-on. In November 2008 bulbs were recovered from their pots, cut transversely and scored for the presence and severity of feeding marks on the scales, also recording whether feeding marks were on the current, previous or new bulb units. Feeding marks occurred frequently on scales of the current bulb units (Table 3.7). The outstanding result is that much lower feeding mark scores occurred in the 12h-frosting treatment, with higher scores in the longer frosting treatments and in the controls, a difficult result to interpret.

Table 3.7: Bulb-scale mite assessments of plants grown-on after glasshouse forcing, following different frosting treatments, 2007-2008 experiment assessed autumn 2008

Frosting treatment	Feeding mark score in current bulb units
12h	0.27
24h	0.50
36h	0.49
None (control)	0.47

2008

A further experiment on 'frosting' was carried out in 2008, to develop these findings. Dutch Master bulbs were potted-up on 24 September 2008, the bulbs for one treatment having been stored at -2°C for the previous 48 hours, after which all pots were placed in a 9°C cold store. On 16 January 2009, all pots were moved to a heated glasshouse. After three days, bulbs allocated to receive 24-, 48- and 72-hour 'frosting' treatments were moved to a cold store at -2°C and were returned to the glasshouse after the appropriate period. One batch of pots received no 'frosting' treatment, as controls. These pots were transported to Wellesbourne in February 2009.

Shoots were examined on 10-11 March 2009 and the average counts of mites and eggs are presented in Figures 3.10-3.11. Only the dry, cold treatment appeared to reduce the numbers of eggs, compared with the control, but this effect may have been due to the initial distribution of mites between the bulbs. No treatment appeared to reduce the number of adults.

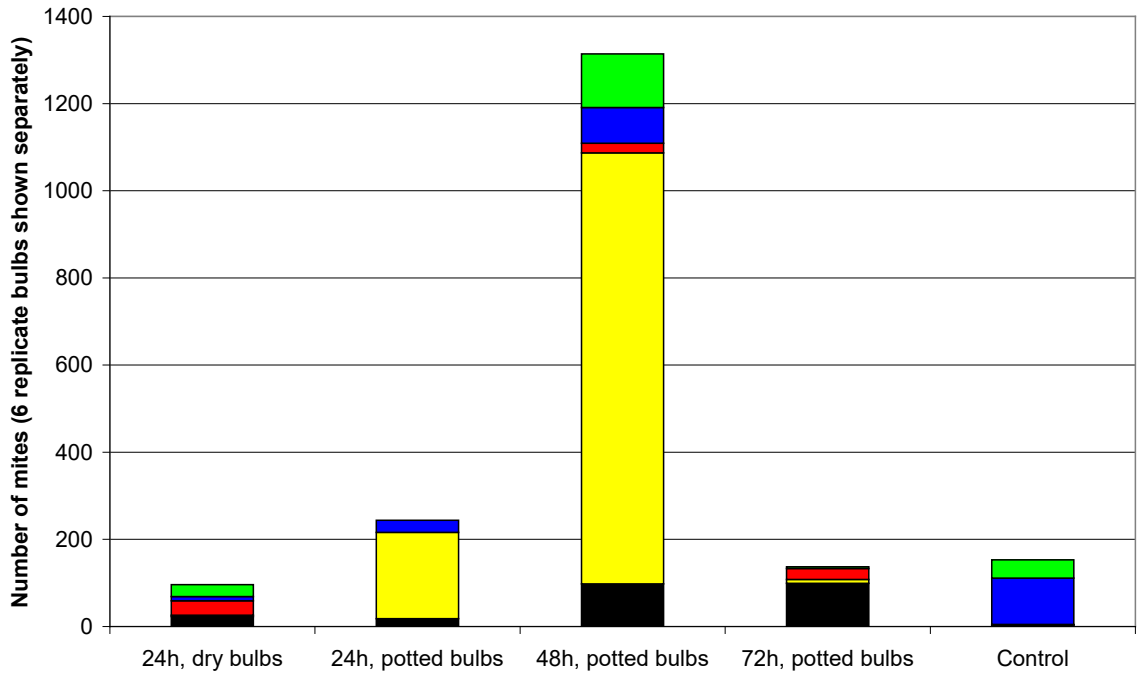


Figure 3.10: Numbers of bulb-scale mite adults in bulb shoots following 'frosting' treatments, 2008-2009 experiment

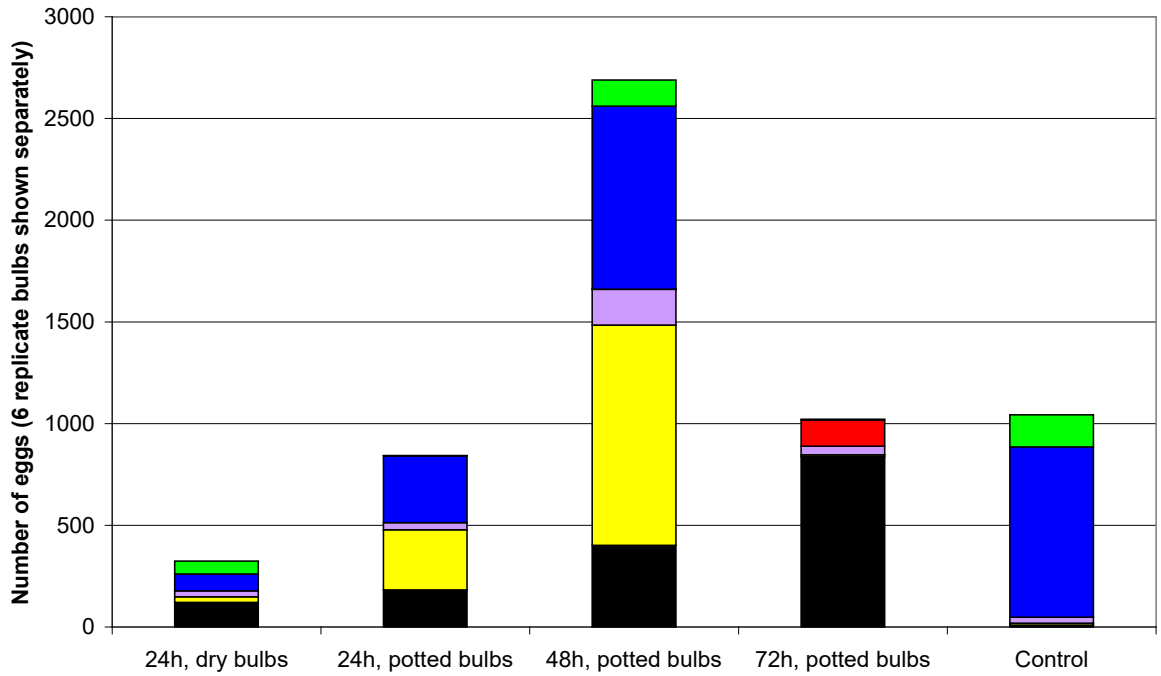


Figure 3.11: Numbers of bulb-scale mite eggs in bulb shoots following 'frosting' treatments, 2008-2009 experiment

Evaluate fogging as a method of mite control in bulb stores

The possibility of using fogging to control bulb-scale mites on bulbs in-store was investigated during 2008. Formalin, Jet 5 and pybuthrin-based insecticides were considered, though formalin was withdrawn from horticultural uses at the end of 2008. Jet 5 is also likely to be withdrawn for such uses. Pybuthrin insecticides have apparently been used extensively for the fogging of produce stores, but discussions with the registration holder clarified that the material cannot be used to treat produce in stores. Consequently this approach was aborted.

Effect of acaricides and disinfectants applied as sprays

The trials focused on acaricides applied as sprays and initial tests were done using pot-grown bulbs. Mite-infested bulbs (grade 8-10 cm) (9 per pot) were potted with un-infested bulbs (5 per pot) in 20cm diameter pots of compost in September 2007. The pots were watered well and placed outside on 'standing ground' at Wellesbourne. In December 2007, the pots were placed inside a polytunnel at Wellesbourne to protect them from extreme weather. A range of acaricides were sourced from pesticide companies and these were applied to the bulbs as foliar sprays (5 replicate pots per treatment). The acaricides tested in this trial were: Actellic (pirimiphos-methyl), Sequel (fenpyroximate), Floramite (bifenazate) a coded treatment (Exp C) and Movento (spirotetramat). There was also an acaricide-free control treatment. There were two trials. Each trial was sprayed twice. The first trial was sprayed on 15 and 29 March the second on 16 and 30 May 2008. The bulbs were then grown on and samples of the foliage were assessed for the presence of live mites in April – June 2008 (one replicate at a time) by sampling 10cm of the lower leaf taken from the bulb neck upwards. The bulbs were later assessed (mid November – early December 2008) for the presence of feeding marks (data not presented). Three similar trials were done in spring 2009 using infested bulbs cv Dutch Master (6 replicates per treatment). The treatments were the same as those used in the previous year in Trials 1 & 2. Additional treatments were tested in Trial 3, namely Dynamec (abamectin) and Masai (tebufenpyrad). A 10cm shoot sample (as described previously) was taken from each pot and examined for mites. The data were subjected to Analysis of Variance (data square-root transformed prior to analysis). The results of Trials 1 and 2 in both years were analysed together. There was a statistically significant effect of 'Year' but not of 'Treatment' (Table 3.8, Figure 3.12). The results of Trial 3 in the second year are summarised in Table 3.9 and Figure 3.13. There was no statistically significant effect of treatment.

Table 3.8: The mean number of bulb-scale mites (all stages) per pot (sampling procedure described in text) recovered from the foliage of potted narcissus treated with foliar sprays (Experiments 1 & 2, 2007-9)

Treatment	Transformed means	Back-transformed means
Actellic	4.36	19.02
Sequel	6.98	48.69
Exp C	5.17	26.75
Floramite	5.77	33.27
Movento	3.7	13.65
Control	8.49	72.06
P-value		
Year	<0.001	
Treatment	0.168	
Year.Treatment	0.159	
Least Significant Difference (5%)		
Year	2.258	
Treatment	3.910	
Year.Treatment	5.530	

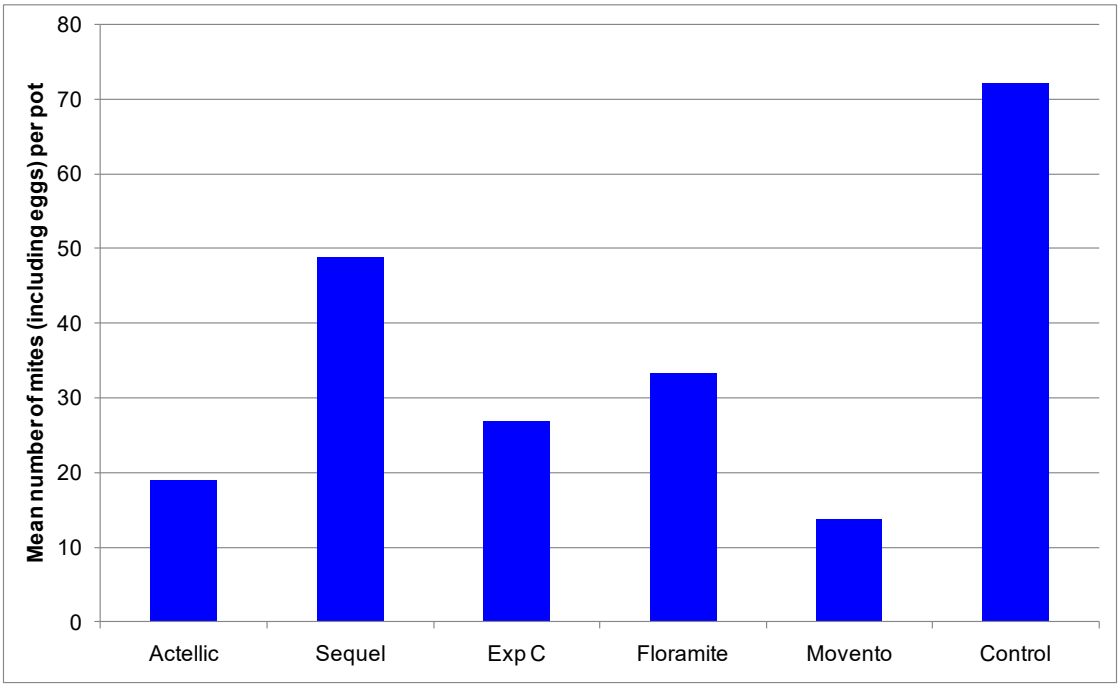


Figure 3.12: The mean number of bulb-scale mites (all stages) per pot (sampling procedure described in text) recovered from the foliage of potted narcissus treated with foliar sprays (Experiments 1 & 2, 2007-9)

Table 3.9: The mean number of bulb-scale mites (all stages) per pot (sampling procedure described in text) recovered from the foliage of potted narcissus treated with foliar sprays (Experiment 3, 2007-9)

Treatment	Transformed means	Back-transformed means
Control	4.11	16.85
Dynamec	4.23	17.92
Masai	4.08	16.64
Floramite	2.15	4.6
P-value	0.444	
Least Significant Difference (5%)	3.099	

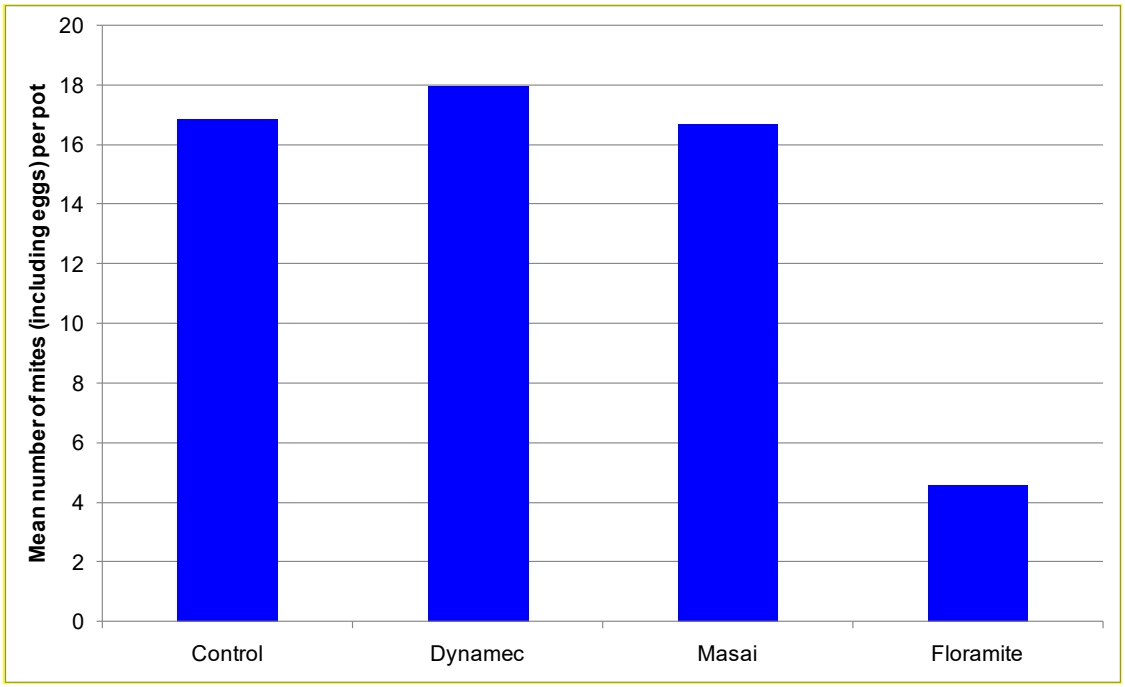


Figure 3.13: The mean number of bulb-scale mites (all stages) per pot (sampling procedure described in text) recovered from the foliage of potted narcissus treated with foliar sprays (Experiment 3, 2007-9)

A final field trial was undertaken in 2010. Part of the block of infested cv Dutch Master at Wellesbourne was divided into 40 plots (each plot 1 row x 3 m). There were 10 treatments including an untreated control and the acaricide treatments were applied to the foliage on 6 occasions at fortnightly intervals from 24 March until 18 June 2010. Samples of bulbs were taken and destructively sampled to assess them for the presence of mites. Initially 4 bulbs

were sampled from each plot and then a further 8 bulbs were assessed from the three treatments which appeared most effective and the untreated control. The treatments are listed in Table 3.10 and were applied at rates recommended by the manufacturers. The results following Analysis of Variance (data square-root transformed prior to analysis) are summarised in Tables 3.11 to 3.13 and in Figures 3.14 to 3.15. None of the treatments provided a statistically-significant level of mite control. Considering all the trials undertaken, Floramite and Movento, in particular, might be worthy of further evaluation.

Table 3.10: Treatments applied as foliar sprays in a field trial at Wellesbourne in 2010

Treatment (active ingredient)	**Product name**
Untreated control	Untreated control
Fenpyroximate	Sequel
Experimental treatment	Exp 1
Bifenazate	Floramite
Spirotetramat	Movento
Experimental treatment	Exp 2
Experimental treatment	Exp 3
Abamectin	Dynamec
Tebufenpyrad	Masai
Bifenthrin	Talstar

Table 3.11: Summary of statistical analysis of treatments applied as foliar sprays in a field trial at Wellesbourne in 2010 (all data)

	All data - eggs		**All data – adult mites**	
Treatment	**Back-transformed**	**Transformed**	**Back-transformed**	**Transformed**
Untreated control	22.48	4.741	7.46	2.731
Sequel	15.11	3.887	14.58	3.818
Exp 1	48.96	6.997	19.9	4.461
Floramite	35.51	5.959	8.67	2.944
Movento	20.35	4.511	13.91	3.73
Exp 2	19.25	4.399	20.69	4.549
Exp 3	17.64	4.2	9.59	3.097
Dynamec	17.04	4.128	8.26	2.874
Masai	12.25	3.5	10.39	3.224
Talstar	31.8	5.639	19.33	4.396
F probability		0.816		0.772
Least significant difference		4.176		2.597

Table 3.12: Summary of statistical analysis of treatments applied as foliar sprays in a field trial at Wellesbourne in 2010; first four samples from each plot

	Samples 1-4 eggs		Samples 1-4 adults	
Treatment	**Back-transformed**	**Transformed**	**Back-transformed**	**Transformed**
Untreated control	14.63	3.825	11.42	3.379
Sequel	15.11	3.887	14.58	3.818
Exp 1	48.96	6.997	19.9	4.461
Floramite	2.65	1.626	3.36	1.832
Movento	20.35	4.511	13.91	3.73
Exp 2	19.25	4.388	20.69	4.549
Exp 3	14.58	3.818	17.98	4.24
Dynamec	14.24	3.774	13.28	3.644
Masai	12.25	3.5	10.39	3.224
Talstar	31.8	5.639	19.33	4.396
F probability		0.419		0.669
Least significant difference		3.946		2.732

Table 3.13: Summary of statistical analysis of treatments applied as foliar sprays in a field trial at Wellesbourne in 2010; samples 5-12 from selected treatments

	Samples 5-12 eggs		Samples 5-12 adults	
Treatment	**Back-transformed**	**Transformed**	**Back-transformed**	**Transformed**
Untreated control	20.88	4.569	5.284	2.299
Floramite	47.7	6.907	10.263	3.204
Exp 3	18.4	4.294	4.28	2.069
Dynamec	16.64	4.079	5.356	2.314
F probability		0.299		0.612
Least significant difference		3.52		2.014

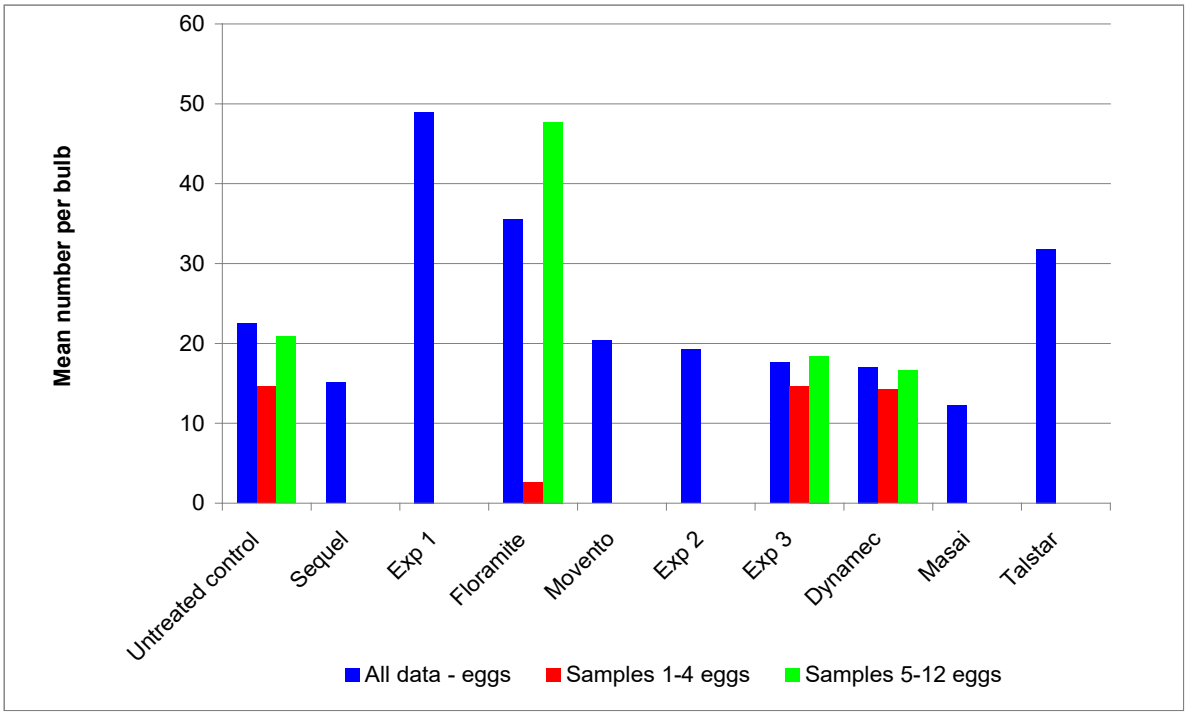


Figure 3.14: Field trial with acaricides – mean number of mite eggs per bulb

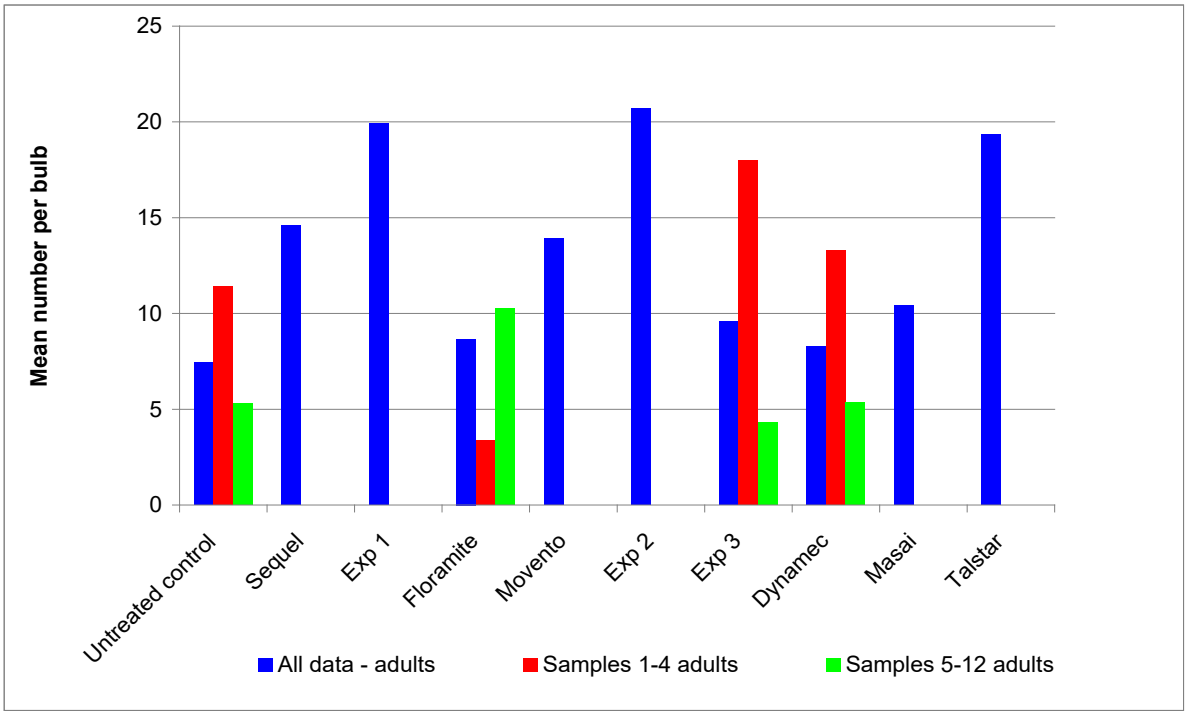


Figure 3.15: Field trial with acaricides – mean number of mite adults per bulb

Objective 4: Design novel biological control strategies and test these as part of an integrated management strategy in commercial crops

Biological control could provide an alternative control strategy. An initial literature review was done to identify potential biological control agents. The predatory mite *Hypoaspis aculeifer* (Canestrini) has demonstrated an ability to suppress the bulb mite, *Rhizoglyphus robini* Claparède on lilies in laboratory trials (Lesna, Sabelis & Conijn, 1996). A limiting factor in the study was the ability of the predatory mite to penetrate between the bulb-scales, where *R. robini* sought refuge. Bulb-scale mite, *Steneotarsonemus laticeps,* is a substantially smaller mite, penetrating deep within bulbs, therefore the potential success of any predator will depend on its ability to reach them. A recent study to identify predators of bulb-scale mite on *Hippeastrum* identified a number of suitable candidates (Messelink & van Holstein-Saj, 2006). They found that predation efficacy was correlated with predator body size and concluded that the best candidate was the small mite *Neoseiulus barkeri* Hughes. This species would be a good candidate to test against bulb-scale mite in this project. Microbes, on the other hand, might provide a more feasible control strategy, since, for example, fungal-infected, but not yet dead mites, could transmit infections to the bulb-scale mite population in the scales, which might be unreachable by predators.

After discussion with Richard GreatRex of Syngenta Bioline, four potential control agents (predators) were provided by Syngenta and tested in small box tests in the laboratory. They were: *Chrysoperla carnea*, *Hypoaspis* mites, *Amblyseius cucumeris* and *Amblyseius (Neoseiulus) barkeri*. Each pack was split between two plastic boxes, a layer of compost was put in bottom of each box, infested bulbs were added and then the surface of the compost was sprinkled with the predators. The lids contained small air holes and the boxes were stood in trays of water to prevent predators moving between treatments. The *A. barkeri* mites arrived later than the other predators as Syngenta Bioline had to source them from another company so these were tested separately. There were control treatments (no predators) in each test. The results are summarized in Figures 4.1 and 4.2. The numbers of bulb-scale mite and bulb mite were very variable, both between treatments and between the two boxes within a treatment, and this is not surprising as the initial numbers of mites per bulb would have varied considerably. All of the bulbs were infested with some live mites at the end of the test. What is most interesting is the larger percentage of dead mites in the boxes containing *A. barkeri* which is the species that the Dutch study on *Hippeastrum* indicated might be effective (Messelink & van Holstein-Saj, 2006).

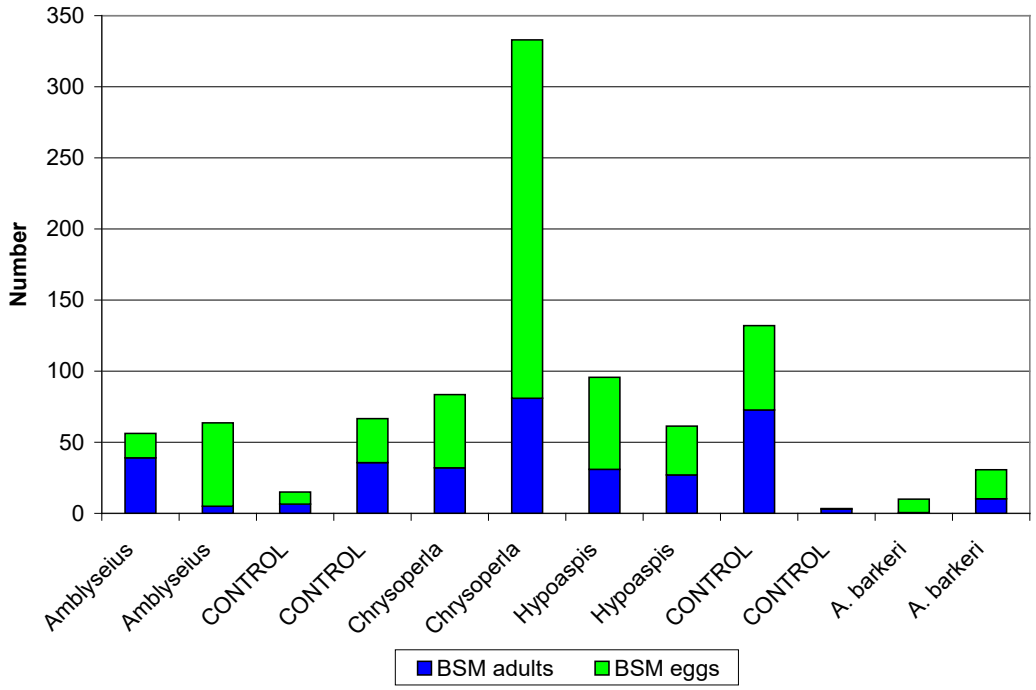


Figure 4.1: Numbers of bulb-scale mite (BSM) in infested bulbs following exposure to biocontrol agents (2 boxes per treatment)

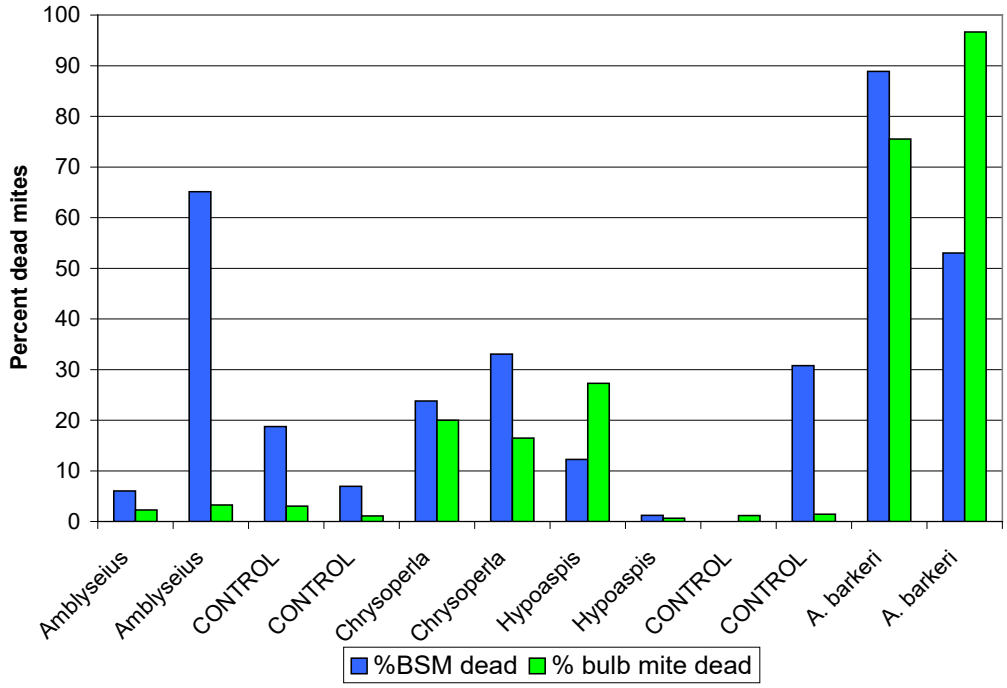


Figure 4.2: Percentage dead bulb-scale mites (BSM) and bulb mites in infested bulbs following exposure to biocontrol agents (2 boxes per treatment)

Objective 5: Examine the link between bulb-scale mite and smoulder disease

2006

It has been reported that bulb-scale mite infestations increase the incidence of smoulder (Gray et al., 1975), and this was investigated in a series of tests, starting with a field trial planted in 2006 at the Kirton Research Centre. In spring 2007 three replicate plots were (1) inoculated with smoulder debris, (2) inoculated with smoulder debris and bulb-scale mite debris/bulbs and (3) left untreated as controls. The trial was left down for two years and was assessed for the incidence and severity of smoulder and bulb-scale mite symptoms each year. Only very occasional symptoms were observed, and, because the inoculation methods appeared to be ineffective, it was decided to carry out further work on pot-grown bulbs.

2007

On 21 September 2007 groups of nine test bulbs of Golden Harvest (previously treated with standard HWT) were placed alone or with five mite-infested bulbs (<8 cm grade) in food-quality brown paper bags (317 x 305mm). Twenty bags were set up with mite-infested bulbs and 20 with test bulbs only. The top of each bag was folded over and the bags were placed in a controlled-temperature store at 17°C for 4 weeks. On 19 October 2007 the bulbs from each bag were planted in pots, with nine test bulbs around the edge of the pot and (where appropriate) five mite-infested bulbs in the centre. As described above, the pots were placed in a cold store and on 22 February 2008, five smoulder-infected leaves were placed among the shoots in each pot of the appropriate treatments and all pots were lightly enclosed in a clear polythene bag. The pots were then moved to the glasshouse, after which the growing medium and shoot surfaces in each pot were sprayed with water and the polythene bags closed for 3 days before the bags were removed. The plants were assessed for foliar symptoms through the growing season, after which they were moved to an unheated mesh tunnel and bulbs bisected and examined in November 2008. In both cases no clear smoulder symptoms, and only a very low incidence of bulb-scale mite symptoms, were observed.

2008

Because of the ineffectiveness of the two earlier experiments, a different approach was used in 2008. No inoculations were attempted, and instead a substantial number of bulb-scale mite-infested bulbs and healthy bulbs were grown and subsequently observed, relying on natural infection to provide smoulder infection. Fifty pots each of the infested ‘Dutch Master’ stock and 50 of the HT-HWT Golden Harvest stock, five bulbs per pot, were planted

on 23 September 2008 and placed in a 9°C cold store. The plants were transferred to Wellesbourne in early February 2009, where they were grown-on in an unheated glasshouse. Examined later in 2009, however, few symptoms of bulb-scale mite, and no symptoms of smoulder, were seen.

Although attempts to infect narcissus with smoulder via the shoots and leaves failed in these cases, the many observations of the extent of bulb-scale mite damage to the young leaf-bases makes it very likely that the extending shoots may easily be infected with fungal inocula within the bulb or in the neck of the bulb.

Objective 6: Deliver a prototype, improved programme for bulb-scale mite control.
The project has provided some insights into the biology of the bulb-scale mite and has questioned the efficacy of the accepted method of control, leading to further research. However, it has also confirmed that, in future, an effective HWT is likely to be the best way of controlling the spread of mite infestations in the field.

Objective 7: Communicate with the industry.
An article about the project was published in HDC News in March 2009 and another one is due to be published in summer 2011. Presentations on the project were made at meetings of the ADAS Bulb Centre on two occasions, at NFU meetings in Cornwall on two occasions and at HDC Narcissus seminars in Lincolnshire and Cornwall in 2011.

DISCUSSION

The project has confirmed that the bulb-scale mite is a challenging pest to work with for growers and researchers alike. In particular, the uneven distribution of mites, together with the considerable amount of time required to dissect a single bulb to count them, were two major challenges when undertaking the experimental work. Every attempt was made to gain as much information as possible for the sampling effort invested, but replication was inevitably poorer than the ideal in some of the studies. Because of the nature of mite distribution it was inevitably simpler to demonstrate when a treatment was ineffective than when it ‘worked’. The presence of a few mites after HWT, for example, readily showed that it was not completely effective, whereas a very large sample of bulbs would need to be taken to demonstrate with a reasonably high level of confidence that a treatment had ‘worked’. It was clear too that assessing mite infestations via the resultant symptoms was ineffective: since the bulb-scales of narcissus bulbs live for about four years, infestations leave an historic record in the feeding marks that does not necessarily represent the current

levels of infestation. At best, even the full bulb examinations used in this study may give an indication only of presence or likely absence.

Whilst we have increased our understanding of mite biology and particularly mite distribution in the field and within the bulb, there are still a number of questions that have not been answered completely and which may never be answered because of the time and resource required to sample at a sufficient level of intensity. This includes the role of dust and debris (in bulb handling areas) and of other agents, such as insects or the wind, in aiding mite dispersal. It is also unclear why mite populations can change so dramatically in size from year to year; to answer this would probably require intensive monitoring over a long period of time in different locations.

The recurring finding that some mites survive HWT suggests that either the recommended treatment time and temperature are insufficient to kill all mites, that the temperatures actually achieved within bulbs varies around the critical levels between dips or even within treatment tanks, or that there have been poor HWT practices (e.g., allowing foam to form on the surface of the dip, or allowing the top surfaces of the bins to become uncovered during dipping). This has lead to further HDC-funded work, to be undertaken in summer 2011 (BOF 63b) to determine the temperatures reached in bulbs exposed in different parts of commercial HWT tanks. HWT will certainly remain the prime means of controlling the pests infesting narcissus: heat treatments, freezing treatments and fumigation were shown to be ineffective, unreliable or unsuitable. Because of the still extant doubts about the importance of wind-blown debris and of mite movement (in the field and in stores) on the spread of bulb-scale mite, the importance of hygiene in bulb-handling operations should still be considered as important as HWT. On the other hand, it was useful to report some positive results from a trial of predatory mites and trials of some new acaricides. Both these possibilities need to be appropriately followed up.

An unexpected finding was the widespread occurrence of bulb mites, as opposed to bulb-scale mites, throughout this project, in bulbs from eastern England and the south-west, and both with and without the simultaneous presence of bulb-scale mites, despite the usual assertion that bulb mites attack only bulbs that have already been damaged. This finding, along with the identity of other mites found in, and in association with, bulbs ought to be investigated further.

ACKNOWLEDGEMENTS

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