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| **Project title:** | Asparagus purple spot: optimising urea application rates and timings to reduce disease on the emerging new crop and ferns |
| **Project number:** | FV 341c |
| **Project leader:** | Angela Huckle, ADAS UK Ltd. |
| **Report:** | Final Report, December 2014 |
| **Previous report:** | Final report, FV 341<br>Annual reports, FV 341a, FV 341b |
| **Key staff:** | Chris Dyer (Statistician)<br>Rob Drummond, Dennis Churchill, Jonny Kerley (Senior Research Technicians)<br><br>Wilson Dyer (Independent asparagus consultant) |
| **Location of project:** | ADAS Boxworth and grower sites in Norfolk and Hereford |
| **Industry Representative:** | Peter Knight (previously Ron Marshall) |
| **Date project commenced:** | 1 January 2014 |
| **Date project completed (or expected completion date):** | 29 January 2015 |

 

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The results and conclusions in this report are based on investigations conducted over a one- or two-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.

Angela Huckle
Research Consultant, Field Vegetables
ADAS UK Ltd.

Signature A.Huckle Date ..29/01/15

**Report authorised by:**

Dr Tim O'Neill
Horticulture Research Manager
ADAS

Tim O'Neill
Signature Date ..29/01/15

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## *GROWER SUMMARY*

### Headline

- In field trials Tech Grade Urea (46% N) applied as a pre-harvest spray application to soil and asparagus debris at 100 kg N/ha in 400 L water/ha gave a significant reduction in spore release of *Stemphylium vesicarium* for up to 14 days post application at one out of the two field sites; *however* there was no subsequent reduction in Stemphylium purple spot on spears or ferns at either site.
- In pot trials Silwett-L77 was the best adjuvant for the urea, the combination resulted in the most spore suppression.

### Background

Stemphylium purple spot of asparagus caused by the fungus *Stemphylium vesicarium* occurs on spears during the harvest season, leading to loss in productivity and spear quality. The disease mainly develops on the asparagus ferns after harvest, affecting main stems, secondary branches and needles where survival structures (pseudothecia) of the fungus are produced. These overwinter on fern debris and this is often the initial source of the disease. Once purple spot is present in a crop, asexual spores (conidia) are produced on lesions in wet weather and are readily spread by wind and water splash leading to rapid disease increase, and the disease can be very difficult to control once established.

Fern debris from the previous season is commonly found on the soil surface as spears emerge. Burial of the debris by ridging up after the crop has senesced reduces the risk of *Stemphylium* spore release in the following spring, but in older crops where crowns and roots develop closer to the soil surface this can be difficult due to the risk of damage to the extensive root system and debris may remain on the surface. Heavy rainfall and footfall during harvest can also cause the debris to become exposed again, and this trash will still have viable fruiting bodies which will release spores. Work done in FV 341b showed that the disease can be reduced in the ferns by application of urea pre-harvest to reduce *S. vesicarium* spores being released from asparagus fern debris. An additional application of urea could be useful post-harvest after the debris has been disturbed by machinery and footfall to give a clean up before the ferns develop. The benefit of this extra application was evaluated in the 2014 trial. In addition to granular urea there are liquid urea products available (Nufol20 and Nuram37) which may be more convenient to use than dissolving granular urea on a large scale. A range of commercially available adjuvants were also tested in this trial to determine the best tank mix partner for urea for efficacy against spore release.

The aim of this project was to improve the control of *Stemphylium* purple spot on asparagus by decreasing overwintering inoculum on crop debris. The objectives were to:

- determine the most effective granular urea rate, form and timing for reduction of the level and persistence of spore production from debris;
- determine the most effective granular urea rate, form and timing for reduction of purple spot on the emerging spears and ferns;
- assess whether additional efficacy is gained from addition of adjuvants;
- assess the influence of water volume on efficacy of the urea.

## Summary

<u>Objective 1 and 2: Effect of urea rate, form and timing for reduction of spore production from debris and reduction of purple spot on spears and ferns - field trials.</u>

A field experiment was carried out in 2014 using commercial crops of asparagus cv. Gijnlim in Norfolk and Herefordshire. Each field site had a known history of *Stemphylium* purple spot and resting bodies (pseudothecia) of the pathogen were present on the crop debris at each site and at each treatment timing. Nine treatments were applied to the crop over three timings as shown in Table 1 below.

**Table 1.** Urea and Nufol20 treatments applied – Norfolk & Herefordshire sites, 2014

| | Treatment description | Product | Water volume L/ha | Application timing (kg N/ha) | |
|---|---|---|---|---|---|
| | | | | T1 Mar/Apr Pre-harvest (post harrowing and ridging) | T2 Post-harvest |
| 1 | Untreated control | - | - | 0 | 0 |
| 2 | Standard pre-harvest (high vol, 100 kg) | Urea* | 1000 | 100 | 0 |
| 3 | Pre-harvest only (moderate vol, 100 kg) | Urea* | 400 | 100 | 0 |
| 4 | Pre-harvest only (moderate vol, 50 kg) | Urea* | 400 | 50 | 0 |
| 5 | Pre-harvest only (low vol, 50 kg) | Urea* | 250 | 50 | 0 |
| 6 | Post-harvest only (low vol, 50 kg) | Urea* | 250 | 0 | 50 |
| 7 | Liquid urea | Nufol20* | 250 | 50 | 0 |
| 8 | Post-harvest app only (moderate vol, 100 kg once) | Urea* | 400 | 0 | 100 |
| 9 | Pre and post-harvest (moderate vol, 100 kg as two applications) | Urea* | 400 | 50 | 50 |

* applied in mixture with Silwett L77 at 0.15%

There were two application timings for each treatment:

- **Timing 1** – applied after ridging but before the residual herbicide application (April 1st 2014 at the Herefordshire site and April 15th 2014 at the Norfolk site)
- **Timing 2** – applied after harvesting of the spears had finished (July 3rd 2014 at the Herefordshire site and June 25th 2014 at the Norfolk site)

Assessments were done to determine the effect of treatments on i) the incidence and intensity of *Stemphylium* spore release from debris collected from experimental plots up to 56 days after treatment application and, ii) incidence and severity of Stemphylium purple spot on spears and fern.

Urea applied as a pre-harvest spray significantly reduced spore production at the Norfolk site in 2014 (Figure 1). At 1 day after treatment, urea (100 kg N/ha in 400 L water) significantly reduced spore release from debris compared with the untreated control (Table 2). At 14 days, although spore production was not completely suppressed, all urea treatments significantly reduced the intensity of spore release to the low category (0-50 spores per piece of debris), compared with the untreated which showed approximately 40% spore release in the medium category (50-150 spores per piece of debris). At 28 days after treatment at the Norfolk site, although there were no significant differences in the suppression of spore production, there was a trend for urea applied at 100 kg N/ha in 400 L/ha to give the greatest reduction in spore release of 72% compared to the untreated. At the experimental site in Hereford, there was no significant reduction in spores released from the debris. This differs from the results seen in 2013 where the application of urea at 100 kg N/ha post-ridging gave a significant reduction in spore release for up to 28 days post application at both trial sites in the east and west (Annual Report, FV 341b). In 2014 the treatments were not as effective or persistent, or alternatively the weather may not have been so conducive to ascospore release when the debris was collected for assessment.

**Table 2.** Summary of the effect of urea treatments on production of *Stemphylium vesicarium* ascospores from asparagus crop debris after pre-harvest applications – 2014

| Treatment and application timing | | | | % spore production relative to the untreated (= 100%) at intervals after treatment | | | | |
|---|---|---|---|---|---|---|---|---|
| | | *Water volume (L/ha)* | *KgN/ha* | Hereford | | | Norfolk | |
| | | | | 1 Days after T1 | 14 Days after T1 | 56 Days after T1 | 1 Days after T1 | 28 Days after T1 |
| 1 | Untreated | 0 | 0 | 100 | 100 | 100 | 100 | 100 |
| 2 | Urea | 1000 | 100 | 73 | 100 | 233 | 100 | 57 |
| 3 | Urea | 400 | 100 | 45 | 100 | 166 | **63** | 28 |
| 4 | Urea | 400 | 50 | 100 | 135 | 266 | 105 | 100 |
| 5 | Urea | 250 | 50 | 82 | 227 | 183 | 89 | 114 |
| 7 | Nufol 20 | 250 | 50 | 64 | 208 | 166 | 74 | 72 |
| 9 | Urea | 400 | 50 | 64 | 112 | 216 | 95 | 100 |

Values in bold are significantly different from the untreated.
Results for treatments 6 and 8 are not shown as they had not been applied at this point.

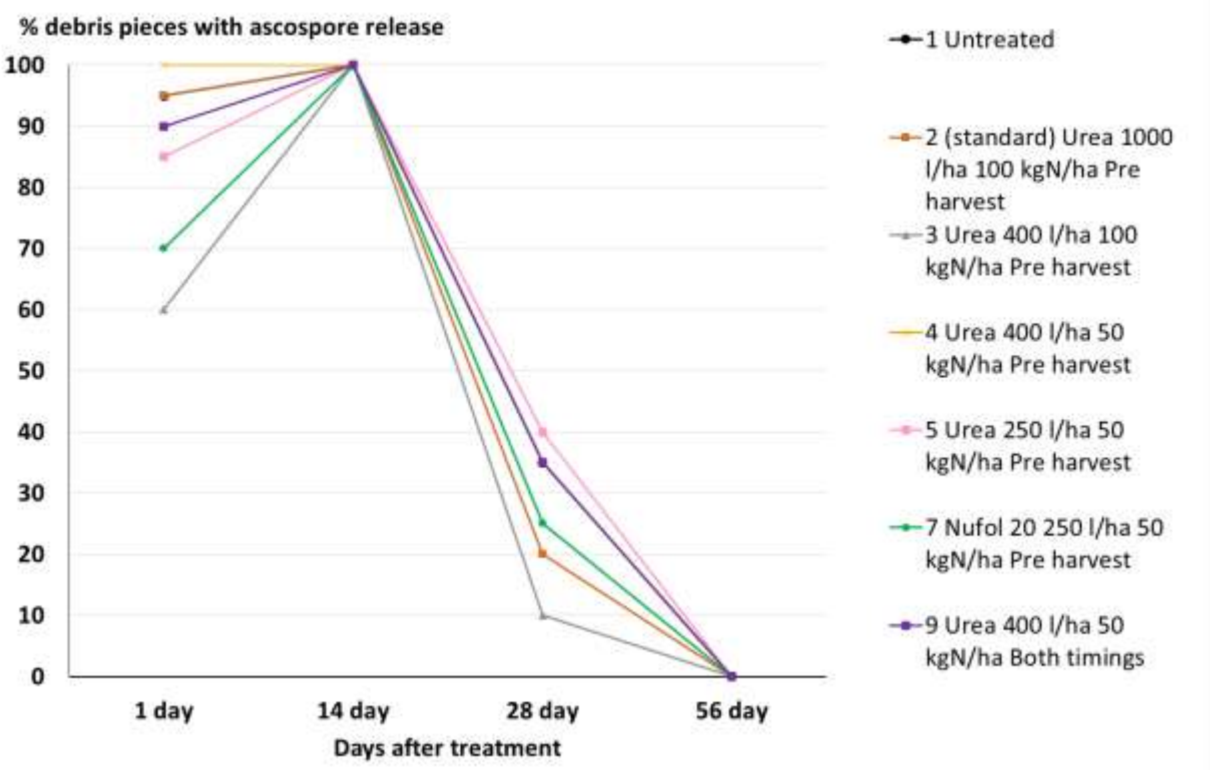


**Figure 1.** Trend in percentage of ascospores released from resting bodies on debris for a period of 8 weeks after urea treatments applied before harvest at the Norfolk site, April to June 2014 (i.e. after the first treatment application and before the second).
***N.B.*** *The data line for the untreated (treatment 1) tracks that of treatment 4 and after day 14 both these treatments track treatment 9. Results for treatments 6 and 8 are not shown as they had not been applied at this point.*

At both sites, there was low or no spore release from the debris after the 2$^{nd}$ application of treatments was made (post-harvest) and there was no effect of urea treatments on spore release. Results from HDC project FV 341 indicated that *Stemphylium* spores on asparagus debris mature between January and March. Therefore the majority of spores are released from infected crop debris just prior to or during the crop harvest which takes place from April to June.

In the dry summer of 2014, conditions were not conducive to infection and development of Stemphylium purple spot. The disease was at low levels during spear harvest and was well controlled during fern development by the commercial fungicide programmes used. Urea treatments applied did not provide additional control of the disease.

The exact mode by which urea suppresses spore release is unknown. However, it has some effect and is used in other crops for control of pathogens such as in apple orchards where urea has been used for a number of years for control of apple scab (*Venturia inequalis*). Here the main mode of action is to hasten the breakdown of leaves, and so destroy the host on which the pathogen needs to survive, it is also acknowledged that the urea changes the microbial and chemical populations on the leaf which could be affecting ascospore release. Where the exact mode of action is unknown, and a number of unpredictable biological factors could be contributing to spore reduction, it is more difficult to reliably predict the efficacy of urea for management of Stemphylium purple spot.

Objective 3: Effect of adjuvants on efficacy of urea in suppressing S.vesicarium spore release.

A pot trial was completed to test a range of adjuvants for their influence on the efficacy of urea against *Stemphylium* spore release from asparagus debris, as well as to consider other forms of nitrogen that could be substituted for urea (which may be more convenient for the grower to use). Eleven treatments were applied to the debris on 28 April as below (Table 3).

**Table 3.** Fertiliser treatments applied, tank mixed with a selection of adjuvants – ADAS Boxworth, 2014

| Treatment number | Treatment | Active Ingredient | Rate N (rate adjuvant) | Water volume |
|---|---|---|---|---|
| 1 | Untreated | | | |
| 2 (standard) | Urea + Silwett – L77 | 46% N + silicone based adjuvant | 50 kg N/ha 0.15% | 400 L/ha |
| 3 | Urea + Tween 20 | 46% N + surfactant (polyoxyethylene (20) sorbitan monolaurate) | 50 kg N/ha 0.1 - 0.5% | 400 L/ha |
| 4 | Urea + Activator 90 | 46% N + non-ionic wetter | 50 kg N/ha 0.1% | 400 L/ha |
| 5 | Urea + X-change | 46% N + water conditioner | 50 kg N/ha 0.25% | 400 L/ha |
| 6 | Urea + Grounded | 46% N + petroleum oil | 50 kg N/ha 1.0% | 400 L/ha |
| 7 | Urea + Bond | 46% N + sticker/extender | 50 kg N/ha 0.1% | 400 L/ha |
| 8 | Urea + Toil | 46% N + methylated rape seed oil | 50 kg N/ha 1.5 L/ha | 400 L/ha |
| 9 | Ammonium nitrate+ Silwett – L77 | 34% N + silicone based adjuvant | 50 kg N/ha 0.15% | 400 L/ha |
| 10 | Nuram37+ Silwett – L77 | 37% N (w/w) + silicone based adjuvant | 50 kg N/ha 0.15% | 135 L/ha |
| 11 | Nufol20+ Silwett – L77 | 20% N (w/w) + silicone based adjuvant | 50 kg N/ha 0.15% | 250 L/ha |
| 12 | Ammonium sulphate+ Silwett – L77 | 21% N + silicone based adjuvant | 50 kg N/ha 0.15% | 400 L/ha |

In the pot experiments carried out at ADAS Boxworth significant differences could be seen at 14 days after application between the adjuvants used. Of the adjuvants trialled with the urea, Silwett-L77 facilitated the greatest spore suppression, and Activator-90, X-change, Grounded and Bond also significantly reduced the spore release at 14 days after application. This result provides a range of choices to the grower of products available in the chemical store which could be substituted for Silwett-L77 if more convenient. Alternative forms of nitrogen were also evaluated for their effects on spore release in the trial and ammonium nitrate, ammonium sulphate and Nufol20 also significantly reduced spore release but did not perform as well as the urea and Silwett-L77 combination. In the field experiments, Nufol20 did not give a significant reduction in spore release as it did in the pot experiments (Table 2, p4).

Objective 4: Effect of water volume on efficacy of urea in suppressing S.vesicarium spore release.

A pot trial was completed to test the influence of a range of water volumes on the efficacy of the urea against *Stemphylium* spore release from asparagus debris. Eleven treatments were applied to the debris on 28 April as below (Table 4).

**Table 4.** Urea treatments applied at different water volumes per hectare – ADAS Boxworth, 2014

| Treatment | Product | Water volume L/ha | Rate (kgN/ha) |
|---|---|---|---|
| 1 | Untreated control | - | 0 |
| 2 (standard) | Urea + Silwett – L77* | 1000 | 50 |
| 3 | Urea + Silwett – L77* | 800 | 50 |
| 4 | Urea + Silwett – L77* | 600 | 50 |
| 5 | Urea + Silwett – L77* | 400 | 50 |
| 6 | Urea + Silwett – L77* | 200 | 50 |

* wetter applied at 0.15%

The influence of water volumes on spore release was also tested in an adjacent experiment to the adjuvants trial with significant differences seen between treatments at 1 day and 14 days after application. The greatest reduction in spore release compared to the untreated was at 14 days after application when urea was applied in 1000 L/ha. There was no significant difference between treatments from 200 to 800 L/ha in the experiment, suggesting that once volume is reduced below 1000 L/ha then the choice of volume used does not have a large influence on increasing efficacy.

## Financial Benefits

The project shows that urea reduced spore release from debris for 14 days post application at one field site only. When applied instead of or as an additional fertiliser application after ridging and prior to spear emergence, urea has potential to reduce the inoculum pressure during harvest. The urea spray would most likely be best used as part of an integrated approach to disease control alongside existing fungicide and fertiliser programmes. It could be argued that with reduced inoculum pressure post-harvest, the first spray application could be delayed and numbers of sprays reduced. However since most available fungicides are primarily protectant in activity, then this would probably be inadvisable. The use of urea for *Stemphylium* control therefore represents an additional input on top of any fungicide programs.

Applying an extra urea spray as well as ammonium nitrate and alongside a fungicide programme may add to input costs for disease control by c. £60/ha using current urea costs of £280/tonne. However urea application seems to contribute to management of *Stemphylium* by reducing spore release near the stem bases, in turn possibly reducing the risk of infection, in an area which is difficult to reach with fungicide sprays when the canopy is closed. Assuming an average yield of 2.5 tonne/ha and a farm gate price of £5,500/tonne, a yield loss of only 0.45 % represents a reduction in sales equivalent to the cost of the additional urea input. Therefore if urea provides a following year yield benefit greater than 0.45 % by additional control of *Stemphylium*, it is worth considering as part of an integrated programme of *Stemphylium* control in asparagus at current urea prices.

## Action Points

- Asparagus fields with high infection pressure from Stemphylium purple spot may benefit from a pre-harvest treatment of urea to suppress spore release from crop debris. Of the treatments tested in this project, urea (combined with Silwett-77) at the rate of 100 kg N/ha in 400 L/ha water gave the most consistent suppression of spore release, up to 14 days after treatment. This finding was supported by results from similar field trials in 2013 (FV 341 b).

- To reduce spore release and possibly purple spot in the emerging crop without compromising crop nutrition or environmental risk, consider applying urea twice as 50 kg N/ha prior to and during harvest instead of nitrogen as a fertiliser. Applying the majority of the asparagus crop's nitrogen requirement, (bearing in mind the NMax in an NVZ is 180 kg N/ha) as urea at 100 kg N/ha just prior to spear growth is not environmentally advisable given that N offtake by the spears at this point in growth is very low, and would increase the risk of loss of N to leaching in a wet spring.

- Urea sprays if used are best targeted to a pre-harvest timing. There appears to be no gain from an application of urea between spear harvest and fern growth, as the majority of ascospore release occurs prior to or during spear harvest.

- The timing of spore release from debris indicates that the highest risk of infection by *S. vesicarium* is to the spears during harvest. But ultimately if protectant fungicides are not applied post-harvest early enough before canopy closure to protect stem bases, infection from debris uncovered by footfall during harvest is still a risk during later fern development as it is the initial source of infection as shown in work in FV 341.

- Commence protectant fungicide programmes soon after the end of harvest and prior to canopy closure, in order to protect asparagus stem bases from infection.

- The work indicates that Silwett-L77 is the best tank mix partner for urea for greatest spore suppression, but if this is not conveniently available to the grower, Activator-90, X-change, Grounded and Bond offer viable alternatives.

- Although Nufol20 gave a significant reduction in spore release in the pot experiments, this significant effect was not borne out in the field trials. However, if used this formulation would be a more practical option for larger scale growers.

- Applying urea in 1000 L/ha gives the greatest efficacy for suppressing spore release but this is not practical, especially for larger growers. Once volumes were reduced below 1000 L/ha then the choice of volume used did not have a large influence on increasing efficacy.

# *SCIENCE SECTION*

## Introduction

Stemphylium purple spot of asparagus, caused by the fungus *Stemphylium vesicarium*, occurs on spears during the harvest season, and reduces productivity and spear quality. The disease develops predominantly on the asparagus ferns after harvest, affecting main stems, secondary branches and needles where survival structures (pseudothecia) of the fungus are produced. These overwinter on fern debris and act as an inoculum source to infect the following crop, causing further crop damage if left untreated.

Once purple spot lesions are present, asexual spores (conidia) can be produced and are readily spread by wind and water splash. This can lead to a rapid increase in disease which can be very difficult to control in the fern canopy once established. Fern debris from the previous season is often present on the soil surface as spears emerge. Burial of the debris by ridging up after the crop has senesced can reduce the risk of *Stemphylium* spore release the following spring. In older crops, where crowns and roots develop closer to the soil surface, this can be difficult due to the risk of damage to the extensive root system and debris may remain. Heavy rainfall and footfall during harvest can also cause the debris to become exposed again, and this trash will still have viable fruiting bodies which will release spores.

Previous research to control blackleg (*Leptosphaeria maculans*) on oilseed rape showed that treatment of stubble with products such as urea and certain adjuvants could reduce inoculum survival between seasons (Humpherson-Jones and Burchill, 1982; Wherrett *et al.,* 2003). A similar approach was evaluated for potential to reduce survival of *Stemphylium* on asparagus crop debris between seasons (FV 341). From August 2008 to March 2009, microscopic examination of asparagus debris was done to determine the timing of development of different stages of the pathogen life-cycle on asparagus fern, to give insight into the timing at which treatment of debris might be effective. Fruiting bodies (pseudothecia) of the perfect (sexual spore) overwintering stage of the *Stemphylium* fungus (*Pleospora herbarum*) developed from August onwards in crops with *Stemphylium* lesions. Maturation of ascospores contained in the fruiting bodies occurred 5 months later (from January).

In previous studies for FV 341, a range of fertiliser and fungicide treatments was applied once to asparagus debris placed into trays once release of ascospores was detected. The same treatments were applied to a separate set of plots 2 weeks later and evaluated for their efficacy in reducing the release of spores from the fruiting bodies on the debris. Urea (200 kg/ha product, equivalent to 98 kgN/ha) applied at 1000 L/ha was one of the two most effective products at reducing the percentage of pieces of debris releasing spores and was more persistent than Switch (cyprodinil + fludioxonil) in reducing spore levels of *S. vesicarium* until 14 days post treatment. Urea was further trialled in 2013 in FV 341b at a range of timings and rates, as well as Perlka (calcium cyanamide) to evaluate the treatments in a field situation for their efficacy at reducing the ascospores released from debris. The treatments of urea were applied at three timings in asparagus field trials in FV 341b (pre-ridging in January/February, post ridging in March/April and post ridging + 4 weeks later) to determine the best time to reduce inoculum levels of *S. vesicarium* pre-harvest. Preliminary results showed that inoculum levels (obtained from spore release counts) can be significantly reduced by a urea spray applied to soil and fern debris before harvest. This also led to a reduction in disease levels on the ferns post-harvest compared to the untreated plots. All plots had a full fungicide programme applied throughout the year, and the urea gave a reduction in disease severity on stem bases that was additional to the control achieved from the fungicide applications. With repeated use in consecutive years, there could be potential for the urea to be used in combination with cultural practices to reduce inoculum levels on infected debris further each year. This could in turn reduce the number of fungicide applications needed.

An additional application of urea could be useful post-harvest after the debris has been disturbed by machinery and footfall to give a clean-up before the ferns develop. The benefit of this extra application was evaluated in the 2014 trial. There are also liquid urea products available (Nufol20 and Nuram37) which may be more convenient to use than dissolving granular urea on a large scale. The aim of the field trial was to determine the most suitable rate, timing and form of urea for commercial application in field crops by examining their effect on (i) level and persistence of spore production (*S. vesicarium*) from debris and (ii) occurrence of purple spot on spears and ferns.

Water volume, adjuvants and other similar products can improve the spread and dispersal of products such as urea, and in turn improve efficacy. The aim of the pot/tray trials was to determine the effect of a wider range of water volumes and products than could be included in the field trial on reducing the level and persistence of spore production from debris. As shown in FV 341b, decreases in spore counts can be related to decreases in purple spot in the crop, and this project considered if it is possible to further refine the treatments in the field.

The aim of this project was to improve control of *Stemphylium* purple spot on asparagus by decreasing overwintering inoculum on crop debris. This was determined against the objectives:

(i) To determine the most effective granular urea rate, form and timing for reduction of the level and persistence of spore production from debris,
(ii) And determine the most effective granular urea rate, form and timing for reduction of purple spot on the emerging spears and ferns,
(iii) To assess whether additional efficacy is gained from addition of adjuvants,
(iv) To assess the influence of water volume on efficacy of the urea.

## Materials and methods

### Field trial – two locations

Site selection

A trial was carried out on a commercial crop of asparagus cv. Gijnlim in each of Norfolk and Herefordshire. Both crops had a history of Stemphylium purple spot infection, with abundant pseudothecia present on the overwintering debris. The trial areas were located within commercial crops and arranged so that treatments could be applied by spraying equipment mounted on a Kawasaki mule or by an Oxford Precision knapsack sprayer. The trial areas were at a different site to the areas used in FV 341b, and therefore there would not be a cumulative or confounding effect from the plots treated in 2013. The trial area at each site including surrounding discard was approximately 11, 000 $m^2$ and individual plots were marked out within these areas using a randomised block design, with each plot measuring 20 m long x six crop rows. One crop row of discard was placed either side and 5-metre discard areas were used between the ends of each plot to mitigate against debris movement during farm operations. This gave a total plot width of eight rows, of which only the central six were assessed.

*Stemphylium* infection was naturally occurring and present as resting bodies (pseudothecia of *P. herbarum*) on the debris in the chosen crop at each site at the first application timing. The trial crops were managed as per commercial practice, e.g. routine treatments were applied as well as crop husbandry and the crop was marketed as normal. A USB logger was placed in each crop to measure temperature and relative humidity hourly and a soil sample was taken.

Trial design and treatments

At both sites the trial consisted of eight treatments plus an untreated control, set out in fully randomised blocks with four replicates (Table 5). All treatments fell within NVZ regulations current at the time of the trial.

**Table 5.** Urea and Nufol20 treatments applied – Norfolk & Herefordshire sites, 2014

| | Treatment description | Product | Water volume L/ha | Application timing (kgN/ha) | |
|---|---|---|---|---|---|
| | | | | T1 Mar/Apr Pre-harvest (post harrowing and ridging) | T2 Post-harvest |
| 1 | Untreated control | - | - | 0 | 0 |
| 2 | Standard pre-harvest (high vol, 100 kg) | Urea* | 1000 | 100 | 0 |
| 3 | Pre-harvest only (moderate vol, 100 kg) | Urea* | 400 | 100 | 0 |
| 4 | Pre-harvest only (moderate vol, 50 kg) | Urea* | 400 | 50 | 0 |
| 5 | Pre-harvest only (low vol, 50 kg) | Urea* | 250 | 50 | 0 |
| 6 | Post-harvest only (low vol, 50 kg) | Urea* | 250 | 0 | 50 |
| 7 | Liquid urea | Nufol20* | 250 | 50 | 0 |
| 8 | Post-harvest app only (moderate vol, 100 kg once) | Urea* | 400 | 0 | 100 |
| 9 | Pre and post-harvest (moderate vol, 100 kg as two applications) | Urea* | 400 | 50 | 50 |

* applied using Silwett L77 at 0.15%

There were two application timings for each treatment:

- **Timing 1** – applied after ridging but before the residual herbicide application (March/April)
- **Timing 2** –applied after harvesting of the spears had finished

Timing 1 applications occurred on April 1st 2014 at the Herefordshire site, and on April 15th 2014 at the Norfolk site. Timing 2 applications occurred on July 3rd 2014 at the

Herefordshire site, and on June 25th 2014 at the Norfolk site. Timing 2 required close grower liaison, as there is often only a small window of opportunity after harvest before residual herbicides are applied.

At the Norfolk site, the urea treatments were applied to the plots as a spray over the debris and soil using a 6 m spray boom mounted on the back of a Kawasaki mule (Figure 2). At the Hereford site, treatments were applied using an OPS backpack sprayer and appropriate spray boom. The urea used was industrial specification technical grade urea supplied by Yara, and all treatments were dissolved in hot water before use to prevent re-crystallisation in the sprayer and nozzle blockages. Urea treatments were applied at the water volumes detailed, using the appropriate nozzles and pressures to achieve the rates required. All the treatments were applied with a wetter (Silwett – L77) to maintain continuity with previous work in FV 341, and encourage coverage of the debris. All experimental treatments were applied by ADAS staff as detailed in the crop diary (Appendix 2).


**Figure 2.** Mounted spray equipment applying urea to a plot, Norfolk, 2014.

Assessments

Spore release from debris was assessed prior to the first urea spray application and 1, 14, 28 and 56 days after each spray timing. This was done by collecting debris at each assessment timing (1, 14, 28 and 56 days) from five random points from the central six rows of each plot, and excluding 1 m around the plot edges to avoid edge effects. The pieces (approx. 0.5 x 0.5 cm) were attached inside Petri dish lids using vaseline and suspended above tap water agar, with five pieces of debris on each plate, and four plates per plot.

Plates were incubated in ambient conditions with a 16 h day/8 h night cycle. Intensity of release of ascospores was estimated on the agar below, once at 5 days after debris plating, using a microscope at x 400 magnification. Ascospore release was categorised as nil, low, medium or high, (nil = nil spores per piece, low = 0-50 spores per piece, moderate = 50-150 spores per piece, high > 150 spores per piece). The absence of mycelium was also noted for samples where ascospore release was recorded as nil. This differentiated between pieces of debris where nil spore release was seen but mycelium grew, and was recorded as 'nil' and those where in addition to no spore release, there was also no mycelial growth, recorded as 'nil mycelium'. The results were categorised rather than counted, as counting the number of spores released was very time consuming, and there were clear differences between each category chosen.

In the field, disease incidence was assessed as presence or absence of purple spots on the spears from 10 spears in the central two rows of each plot. Disease severity was assessed as percentage area affected. Number of spears per metre was also recorded. These in-crop assessments occurred at appropriate timings throughout harvest when infection was expected, e.g. after high rainfall events. In Herefordshire, spear assessments were carried out on May 3$^{rd}$, May 13$^{th}$ and June 7$^{th}$. In Norfolk, spear assessments were carried out on May 14$^{th}$ and June 5$^{th}$. Incidence and severity assessments of purple spot during the fern growth stage was assessed at five points in each plot, on the stem bases and the fern canopy separately. In Herefordshire, fern assessments were carried out on August 26$^{th}$, September 10$^{th}$, September 26$^{th}$ and October 16$^{th}$. In Norfolk, fern assessments were carried out on September 1$^{st}$ and October 20$^{th}$. Crop vigour was also recorded on a 0-9 scale (0 = dead, 9 = excellent) at each assessment in Herefordshire. In Norfolk, greenness and defoliation were recorded at each assessment with; for greenness, 0 = brown, 9 = green, and for defoliation, 0 = all needles dropped and 9 = all needles present.

## Pot trials

Stemphylium-infected asparagus debris was collected from the grower site in Norfolk in March, with abundant pseudothecia present. Debris was laid out in plots on a sheltered hard standing area of soil covered with Mypex® at ADAS Boxworth. A plot was one 1 x 0.5 m free draining chitting tray of debris, and plots were laid out in a fully randomised block design. Trays were covered with mesh to keep debris in the appropriate plots and prevent it blowing into others. The urea used was industrial specification technical grade urea supplied by Yara, and all treatments were dissolved in hot water before use to prevent re-crystallisation in the sprayer and nozzle blockages. Urea treatments were applied using the appropriate nozzles and pressures to achieve the rates required. All experimental treatments were applied by ADAS staff as detailed in the crop diary (Appendix 1).

Trial design and treatments - Effect of water volumes for urea treatments on ascospore release from asparagus debris

This trial consisted of five treatments (of five different water volumes per hectare) plus an untreated control replicated four times and set out in a fully randomised design (Table 6). Spray treatments were applied once, on April 28$^{th}$, using an OPS backpack sprayer.

**Table 6.** Urea treatments applied at different water volumes per hectare – ADAS Boxworth, 2014

| Treatment | Product | Water volume L/ha | Rate (kg N/ha) |
|---|---|---|---|
| 1 | Untreated control | - | 0 |
| 2 (standard) | Urea + Silwett – L77* | 1000 | 50 |
| 3 | Urea + Silwett – L77* | 800 | 50 |
| 4 | Urea + Silwett – L77* | 600 | 50 |
| 5 | Urea + Silwett – L77* | 400 | 50 |
| 6 | Urea + Silwett – L77* | 200 | 50 |

* wetter applied at 0.15%.

Trial design and treatments - Effect of adjuvants with urea treatments on ascospore release from asparagus debris

This trial consisted of 11 treatments plus an untreated control and was replicated four times (Table 7). Spray treatments were applied once, on April 28$^{th}$, using an OPS backpack sprayer in 400 L water/ha with single nozzle at 2 -3 Bar pressure with an 02F110 flat fan nozzle. T10 (Nuram37) was applied in a rate of 135 L water/ha, and T11 (Nufol20) was applied in a rate of 250 L water/ha to achieve an application of 50 kg N/ha consistent with the other treatments.

 

**Table 7.** Fertiliser treatments applied, tank mixed with a selection of adjuvants – ADAS Boxworth, 2014

| Treatment number | Treatment | Active Ingredient | Rate N (rate adjuvant) | Water volume |
|---|---|---|---|---|
| 1 | Untreated | | | |
| 2 (standard) | Urea + Silwett – L77 | 46% N + silicone based adjuvant | 50 kg N/ha<br>0.15% | 400 L/ha |
| 3 | Urea + Tween 20 | 46% N + surfactant (Polyoxyethylene (20) sorbitan monolaurate) | 50 kg N/ha<br>0.1 - 0.5% | 400 L/ha |
| 4 | Urea + Activator 90 | 46% N + non-ionic wetter | 50 kg N/ha<br>0.1% | 400 L/ha |
| 5 | Urea + X-change | 46% N + water conditioner | 50 kg N/ha<br>0.25% | 400 L/ha |
| 6 | Urea + Grounded | 46% N + petroleum oil | 50 kg N/ha<br>1.0% | 400 L/ha |
| 7 | Urea + Bond | 46% N + sticker/extender | 50 kg N/ha<br>0.1% | 400 L/ha |
| 8 | Urea + Toil | 46% N + methylated rape seed oil | 50 kg N/ha<br>1.5 L/ha | 400 L/ha |
| 9 | Ammonium nitrate+ Silwett – L77 | 34% N + silicone based adjuvant | 50 kg N/ha<br>0.15% | 400 L/ha |
| 10 | Nuram37+ Silwett – L77 | 37% N (w/w) + silicone based adjuvant | 50 kg N/ha<br>0.15% | 135 L/ha |
| 11 | Nufol20+ Silwett – L77 | 20% N (w/w) + silicone based adjuvant | 50 kg N/ha<br>0.15% | 250 L/ha |
| 12 | Ammonium sulphate+ Silwett – L77 | 21% N + silicone based adjuvant | 50 kg N/ha<br>0.15% | 400 L/ha |

<u>Assessments</u>

Plots were sampled prior to spray application on April 28$^{th}$ in both trials to check for release of ascospores. Plots were then sampled 1, 14, 28 and 56 days after spray application to assess the effect of treatment on spore release. Assessments took place on April 29$^{th}$, May 12$^{th}$, May 27$^{th}$ and June 25$^{th}$.

The pieces (approx. 0.5 x 0.5 cm) were attached inside Petri dish lids using vaseline and suspended above tap water agar, with five pieces of debris on each plate, and four plates per plot. Plates were incubated in ambient conditions with a 16 h day/8 h night cycle. Intensity of release of ascospores was estimated on the agar below, once at 5 days after debris plating, using a microscope at x 400 magnification. Ascospore release was categorised as nil, low, medium or high, (nil = nil spores per piece, low = 0-50 spores per piece, moderate = 50-150 spores per piece, high > 150 spores per piece). The absence of mycelium was also noted for samples where ascospore release was recorded as nil. This differentiated between pieces of debris where nil spore release was seen but mycelium grew, and was recorded as 'nil' and those where in addition to no spore release, there was also no mycelial growth, recorded as 'nil mycelium'.

Statistical analyses

The proportions in each category (nil, low, medium, high as detailed above) were analysed and tested for statistical differences by use of Generalised Linear Models (GLM), using the logistic regression model. The proportion of debris releasing ascospores was calculated by converting the predicted values reported for the treatment to percentages, by subtracting the predicted value from 1 and multiplying by 100 (see data analysis in Appendix 2 for predicted values and details of significant differences). The standard errors vary for each treatment, so 95% confidence intervals were used to identify differences between treatments.

The incidence and severity of purple spot on spears, stem bases and canopy were analysed using ANOVA.

## Results

The trial was carried out at two sites (Hereford and Norfolk) with differing climate conditions. Results from each site are presented separately with reference to the project objectives.

### Objective 1 - Efficacy of urea treatments: rates, forms and timing for reduction of ascospore release from crop debris by *S. vesicarium* – field trials.

Hereford site

The first urea treatments were applied on 1st April just prior to spear emergence and harvest. Spore release was very variable in the untreated plots after the pre-harvest treatments were applied, and ranged from 55% at 1 day after application to 0% spore release at 14 and 28 days after application. Spore release from debris pieces subsequently increased to 30% at the final assessment 2 months after the pre-harvest application (Figure 3a). The treated plots showed a similar variability and at the 14 day and 2 month post-application assessments suggested greater spore release than the untreated. There were, however, no significant differences between treatments with regards to % debris pieces with ascospore release at any of the assessments carried out. Therefore the data must be treated with caution as any trends could be coincidental variability.

Proportions of debris that gave ascospore release in different categories varied between the treatments, and there were significant differences between percentages in the low, medium and high categories (P = 0.052 to <0.001, Appendix 2) at 1 and 14 days after treatment (Figure 3b and 3c). But this did not further help to distinguish which, if any, was the treatment with the greatest efficacy at reducing spore release because of data variability and inconsistent treatment effects.

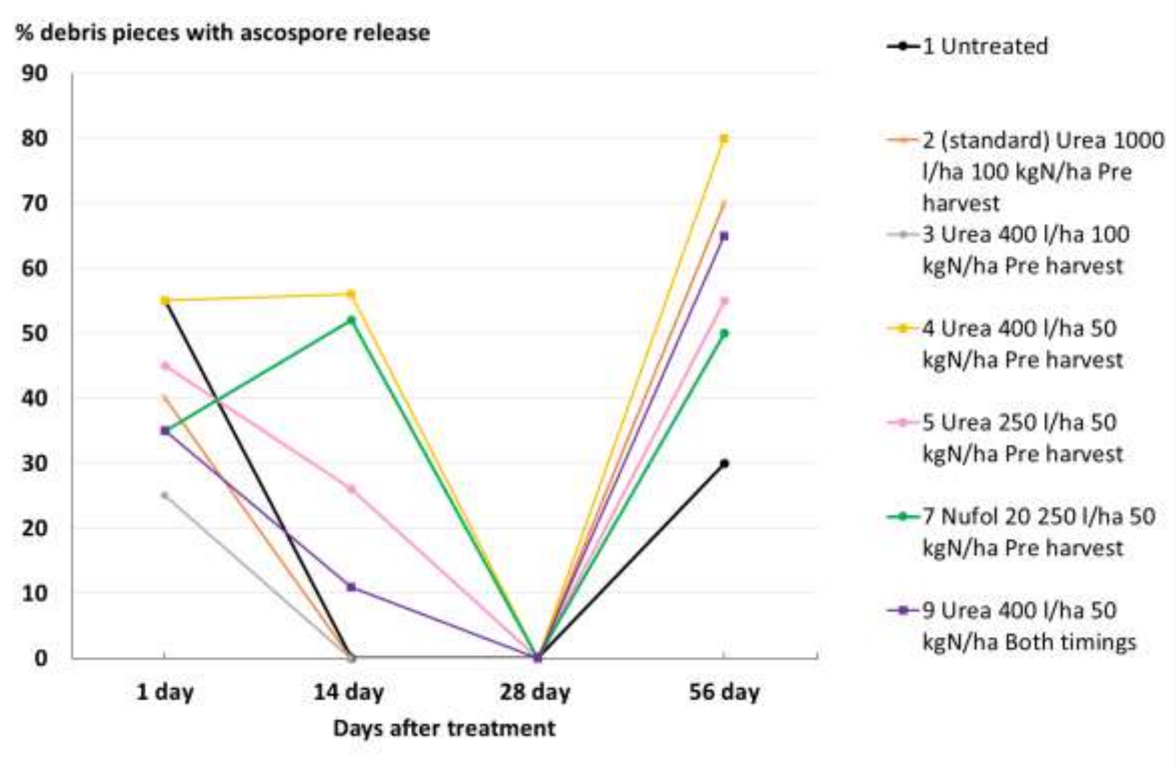


**Figure 3a:** Trend in percentage of ascospores released from resting bodies on debris for a period of 8 weeks after urea treatments applied before harvest at the Hereford site, April to June 2014 (i.e. after the first treatment application and before the second).

In this figure and those following results for treatments 6 and 8 are not shown as they had not been applied at this point.

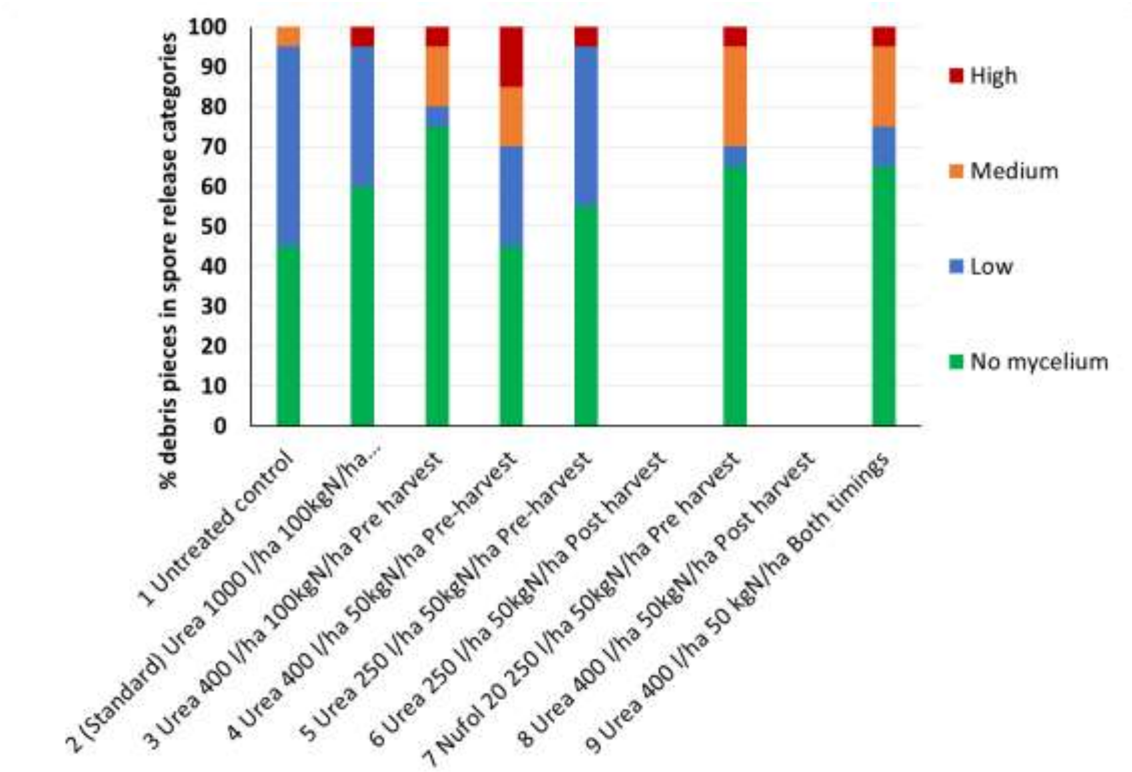


**Figure 3b:** Proportion of debris pieces with ascospore release in categories - no mycelium, low, medium and high, 1 day after treatment application. (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece).

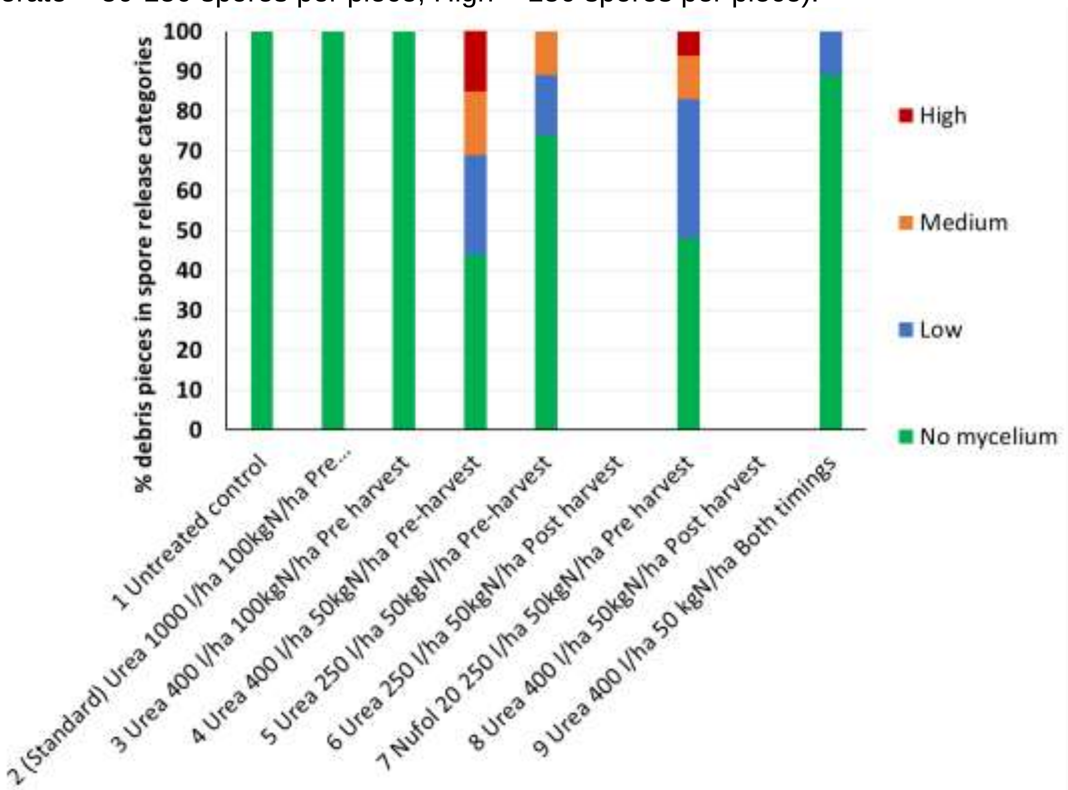


**Figure 3c.** Proportion of debris pieces with ascospores released in categories no mycelium, low, medium and high, 14 days after treatment application. (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece). N.B. There was no ascospore release at 28 days after treatment application

 

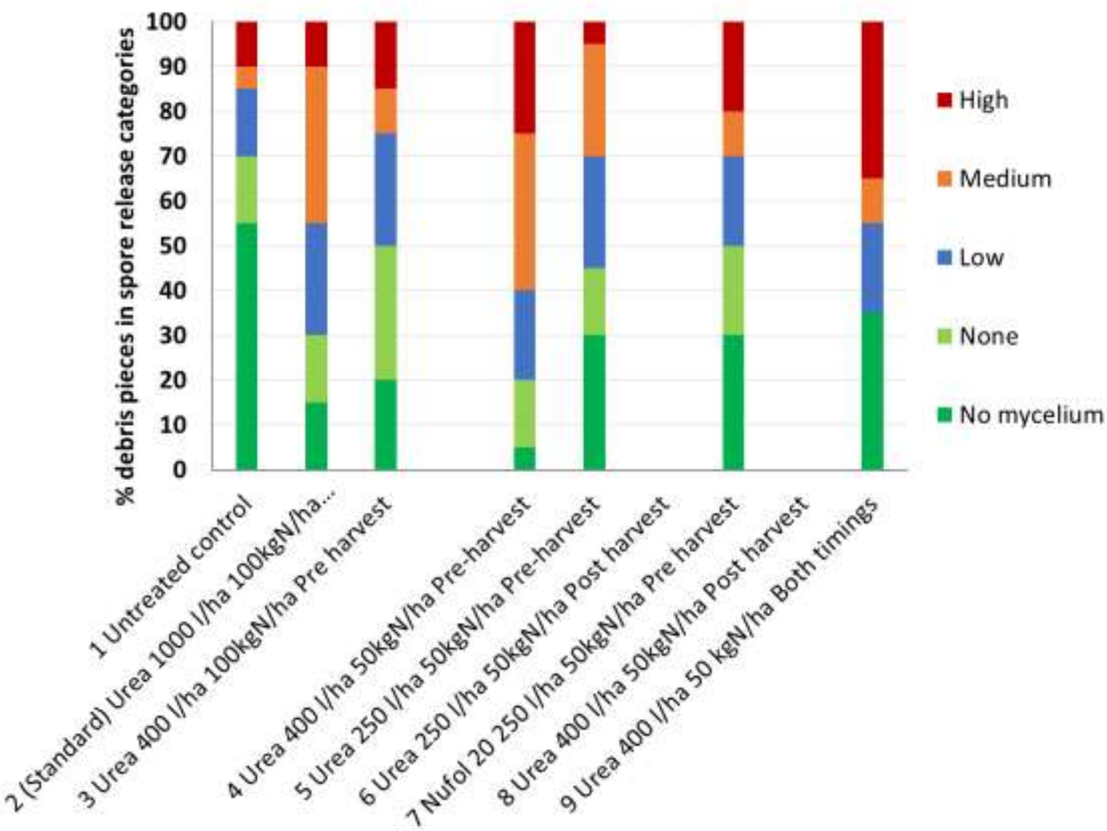


**Figure 3d.** Proportion of debris pieces with ascospores released in categories no mycelium, none, low, medium and high 56 days after treatment. (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece).

At day 1, there is an apparent reduction in spore release due to urea treatments (Figure 3a), with higher values in the 'no mycelium' category for urea treatments compared with the untreated control (Figure 3b), however these differences are not statistically significant. At all other assessments no clear treatment effects are seen.

The second urea treatments were applied on 3 July after harvest ceased and before the spears developed into ferns. At subsequent assessments, there was no spore release from any of the debris collected from any of the plots, treated or untreated. This may have been due to the robust protectant fungicide spray programme that would have been applied from this point, and its influence on any ascospores released at this time.

Norfolk site

At this site the first treatments were applied on 15 April just prior to spear emergence and harvest. Spore release was high in the untreated plots at greater than 95% at 1 day and 14 days after the pre-harvest treatments were applied. Spore release then subsequently fell to 35% at 1 month after application and finally 0% at 2 months after application. Spore release in the treated plots followed a similar trend. At 1 day after the treatments were applied, a significant reduction in spore release was seen between the untreated plots and those where urea was applied at 100 kg N/ha in 400 L/ha (P = 0.008, full data analysis in Appendix 2) (Figure 4a - N.B. The data line for the untreated tracks that of treatment 4 and is difficult to see).

Proportions of debris that gave ascospore release varied between the treatments and there were significant differences between percentages in the nil mycelium category at 1 day after application (P = 0.008; Figure 4b), with urea treatment 100 kg N/ha in 400 L/ha giving significantly lower levels of spore release than the untreated control. At 14 days after treatment there were no plots without spore release but there were significant reductions in the intensity of spores released in the medium category following urea treatment at any water volume (except Nufol 20) ($P < 0.001$), compared with the untreated control(Figure 4c).

At 28 days post treatment there were no significant reductions in levels of spore release in any of the categories, but there was a trend for Urea applied as 100 kg N/ha in 400 L/ha to give the highest reduction in spore release (Figure 4d). At 56 days after treatment there was no spore release from any of the debris collected from any of the plots. (Figure 4a)

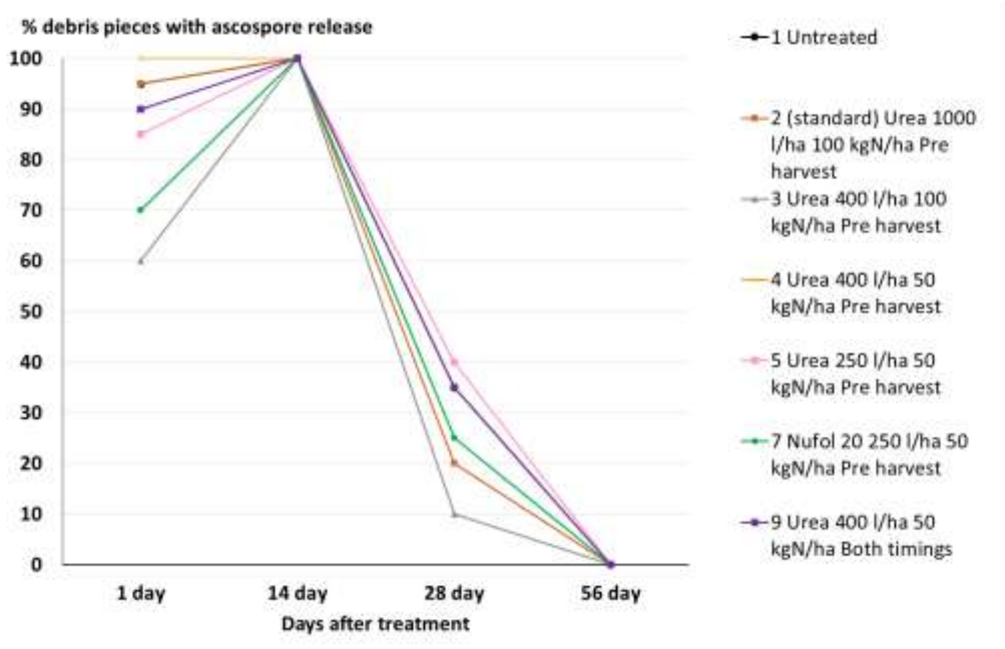


**Figure 4a.**Trend in percentage of ascospores released from resting bodies on debris for a period of 8 weeks after urea treatments applied before harvest at the Norfolk site, April to June 2014 (i.e. after the first treatment application and before the second).
In this figure and those following results for treatments 6 and 8 are not shown as they had not been applied at this point.

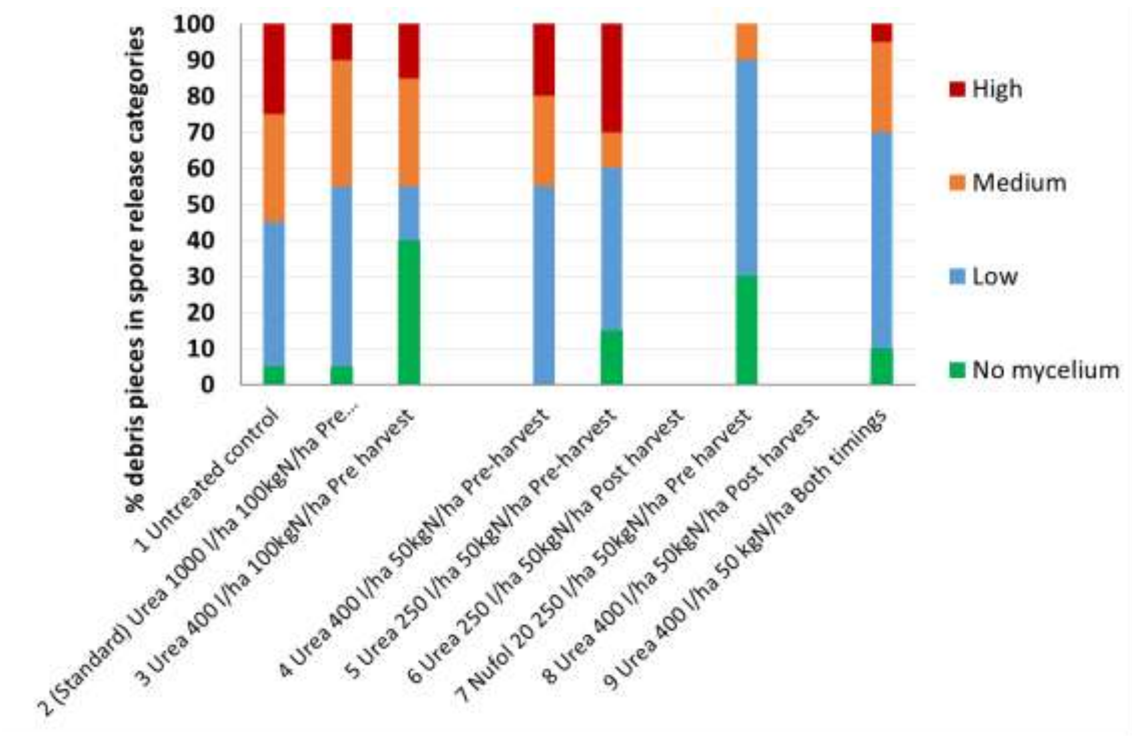


**Figure 4b.** Proportion of debris pieces with ascospores released in categories no mycelium, low, medium and high 1 day after treatment application. (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece).

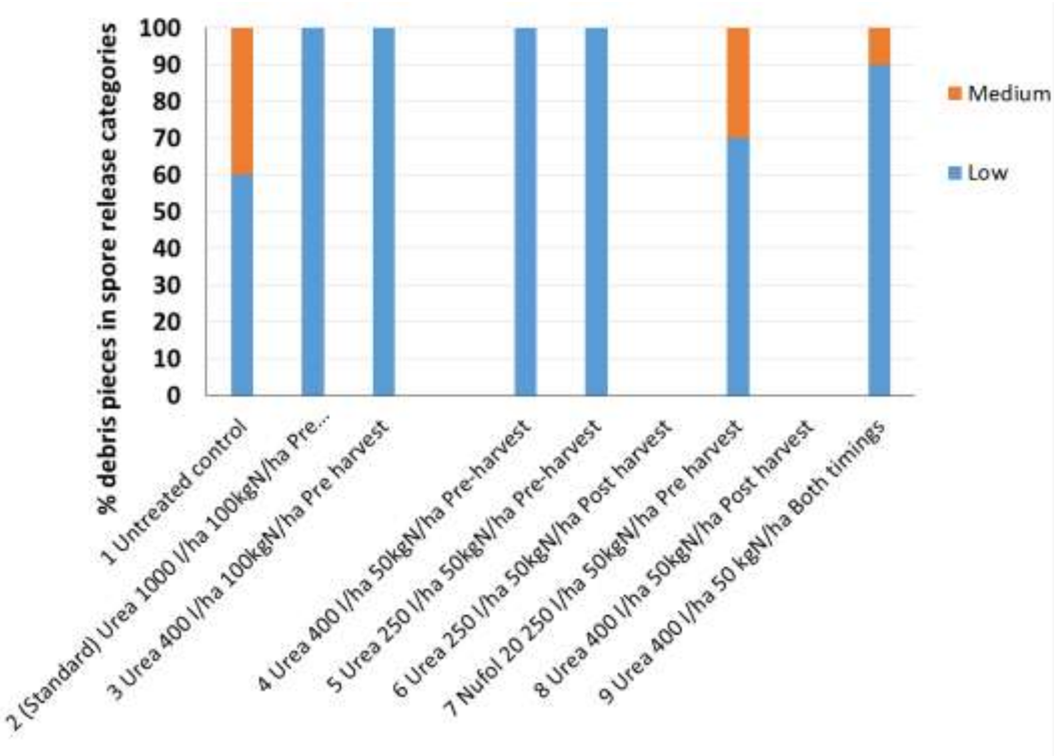


**Figure 4c.** Proportion of debris pieces with ascospores released in categories low and medium 14 days after treatment application. (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece).

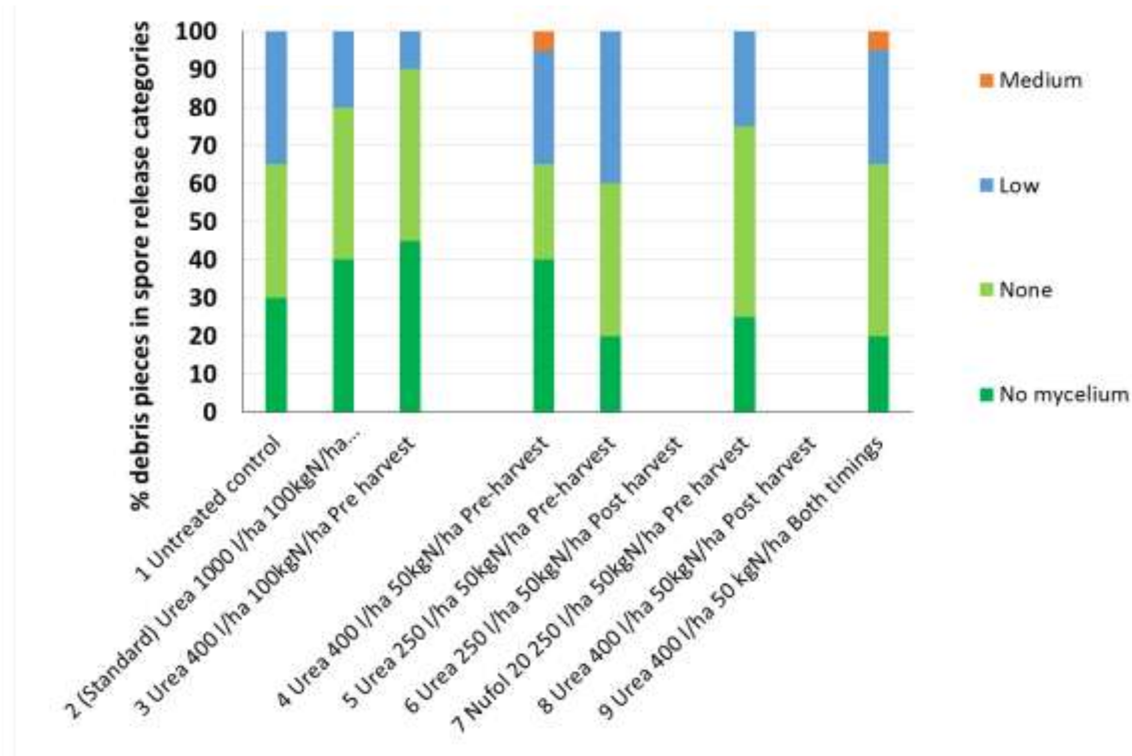


**Figure 4d.** Proportion of debris pieces with ascospores released in categories no mycelium, none, low, and medium 28 days after treatment. (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece). N.B. There was no ascospore release at 56 days after treatment application.

The second urea treatments were applied on 25 June after harvesting had ceased and before the spears started to expand to ferns. Spore release was high in the untreated plots at 90% at 1 day post application, and then fell steadily to 60% and 55% at 14 and 28 days respectively after the pre-harvest treatments and finally nil spore release was seen 2 months after application (Figure 5a). Levels of spore release from individual debris pieces were lower at this point in the crop cycle, with no pieces recorded with spore release in the medium and high categories. There were no significant differences between treatments in either % debris pieces with ascospores released or proportions of spore release in the nil mycelium growth or low spore release categories at any of the assessments (Figures 5b - d).

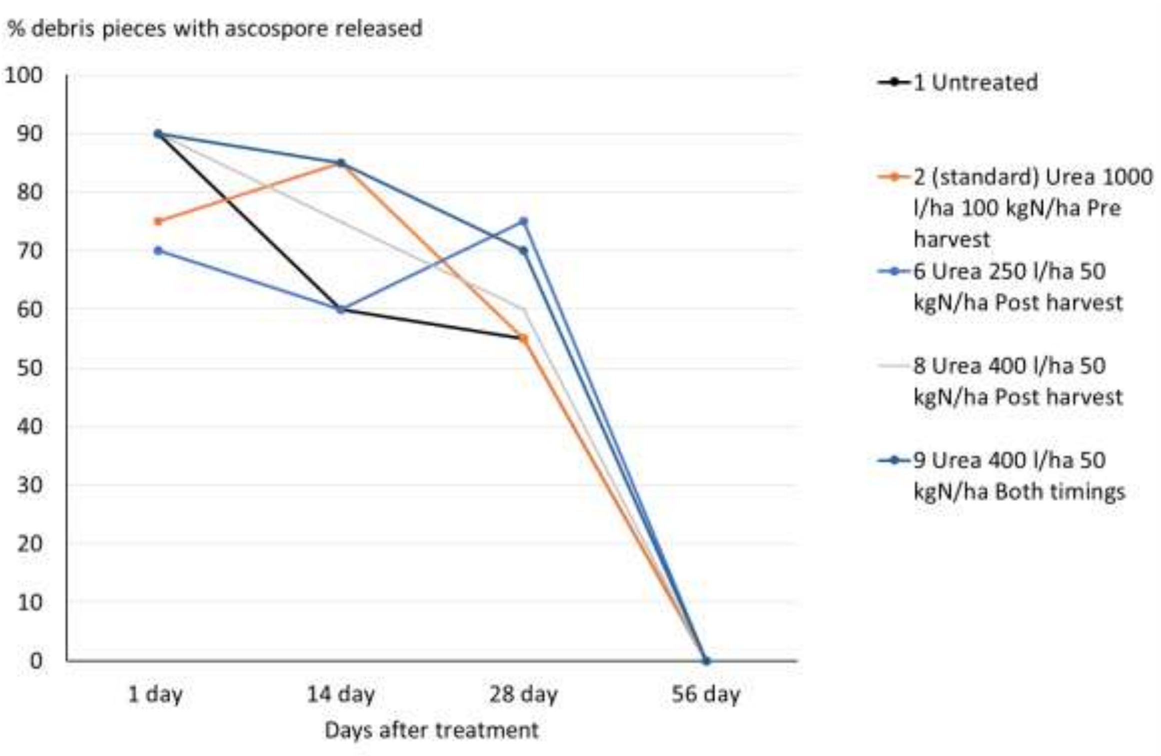


**Figure 5a:** Trend in percentage of ascospores released from resting bodies on debris for a period of 8 weeks after urea treatments applied post-harvest at the Norfolk site, June to September 2014.

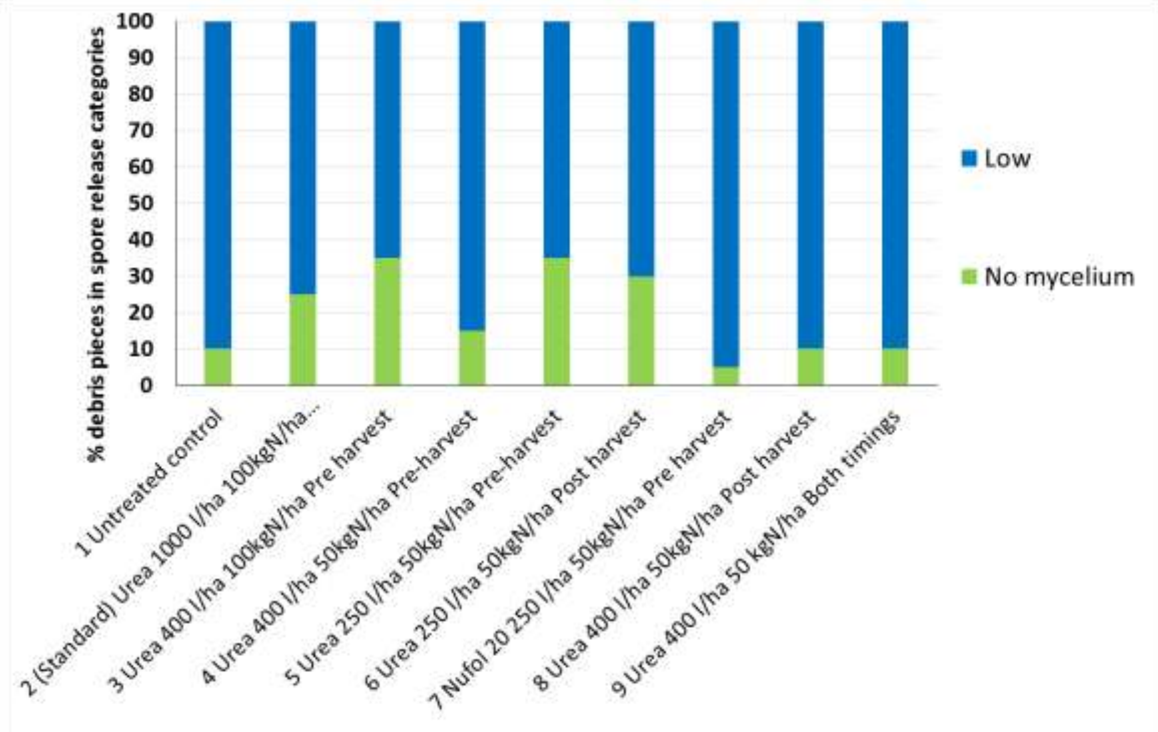


**Figure 5b.** Proportion of debris pieces with ascospores released in categories no mycelium and low 1 day after treatment application. (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece).

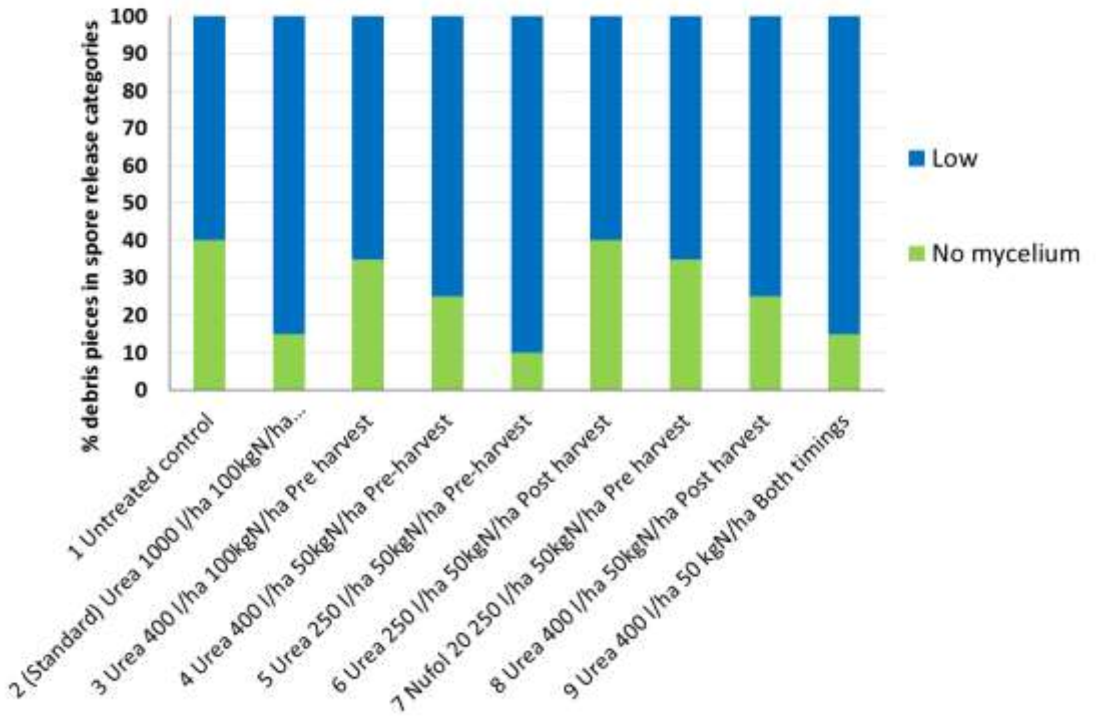


**Figure 5c.** Proportion of debris pieces with ascospores released in categories no mycelium and low 14 days after treatment application. (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece).

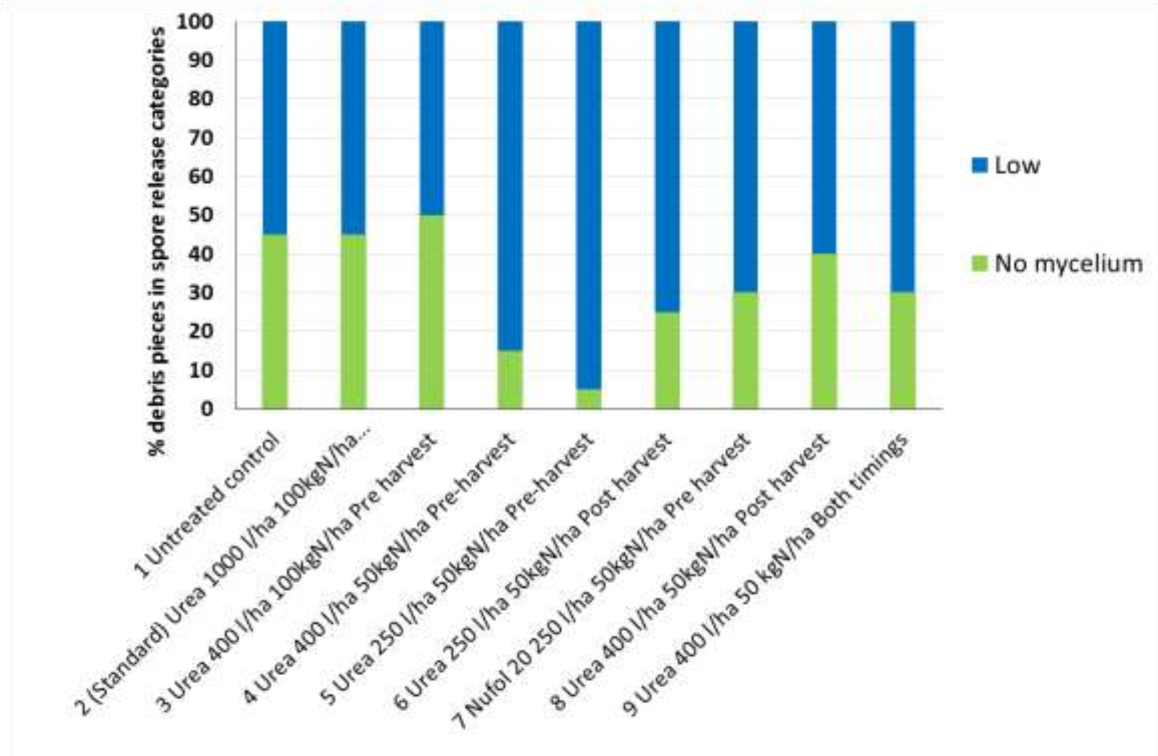


**Figure 5d.** Proportion of debris pieces with ascospores released in categories no mycelium and low 28 days after treatment. (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece). N.B. There was no ascospore release at 56 days after treatment application

**Objective 2: Efficacy of urea: rates, forms and timing for reduction of purple spot in the emerging crop and ferns – field trials.**

Hereford site

The spear assessments took place after spells of heavy rainfall, after which infection by *Stemphylium* purple spot would be expected to take place. The incidence and severity of purple spot on spears was assessed as described in the materials and methods section, three times on 3 May, 13 May and 7 June. Severity was low (0.5 to 1%), with only one or two small lesions per spear at each assessment despite the rainfall in the week beforehand and no significant differences were seen between treatments (data not shown). Therefore only the incidence results are shown in Figure 6. Incidence was also low with a maximum of 18.5% in the untreated at the assessment on 13 May. There were no significant differences in incidence of *Stemphylium* for urea treatments when compared to the untreated. Temperature and rainfall at the time of application and throughout the trial are shown in Figure 7. Tables of the full datasets can be found in Appendix 2.

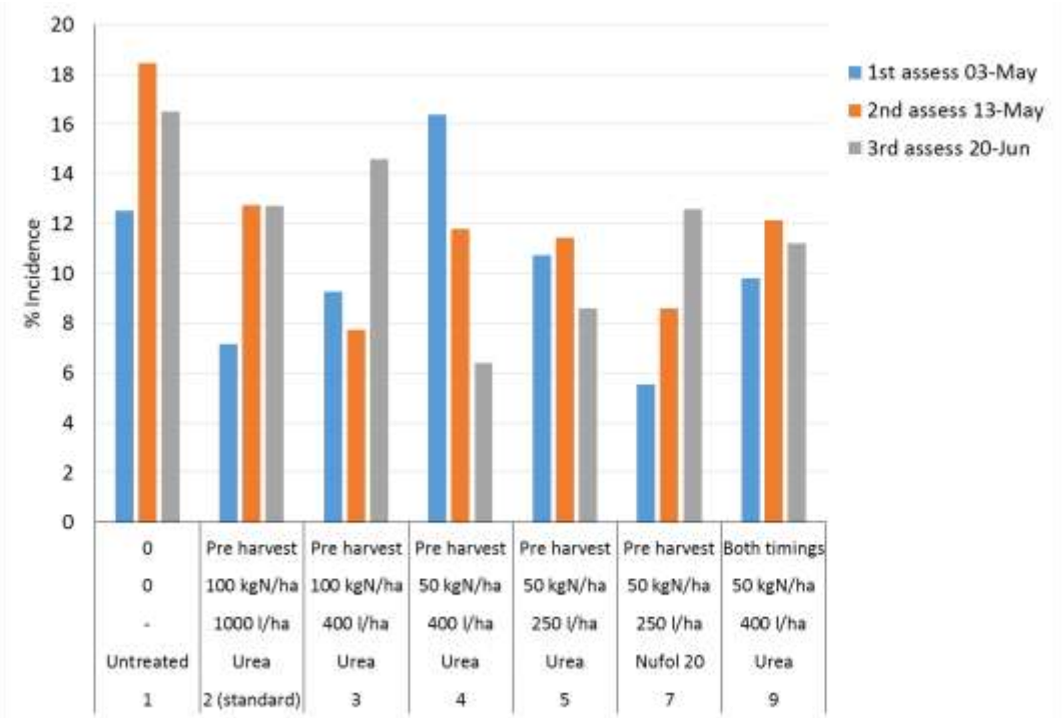


**Figure 6.** Incidence of purple spot on the emerging spears at assessments on 3 May, 13 May and 20 June, Hereford, 2014. In this figure results for treatments 6 and 8 are not shown as they had not been applied at this point.

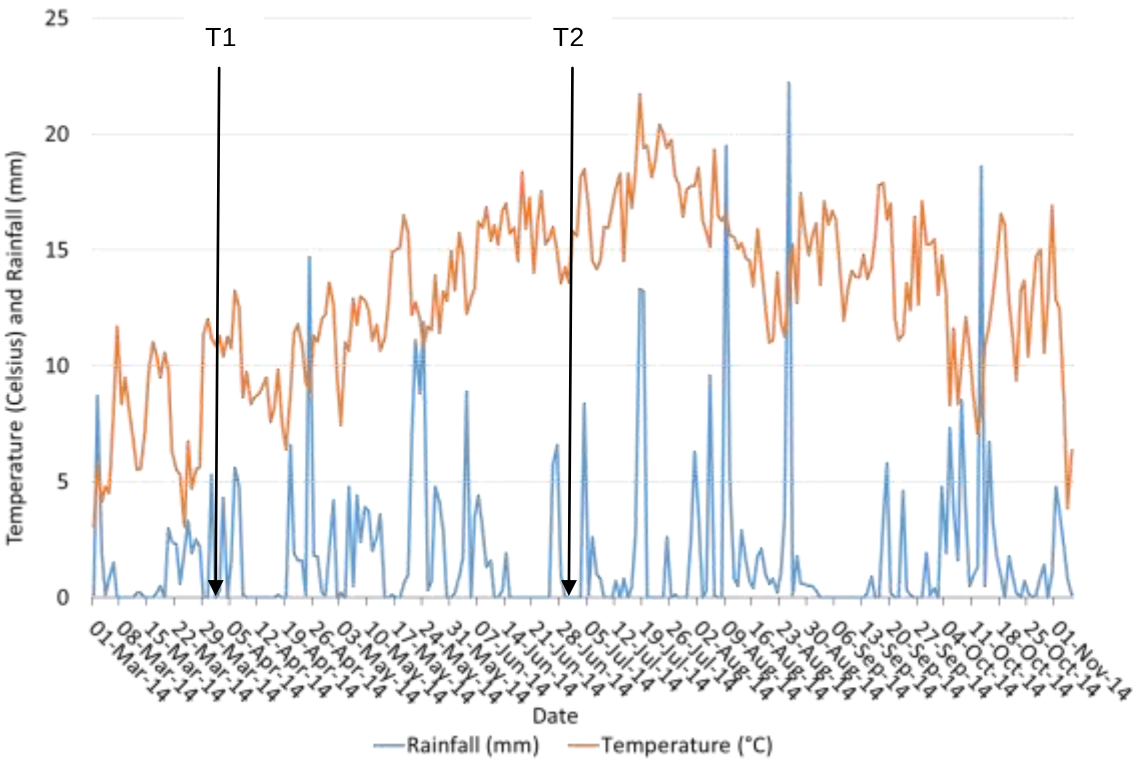


**Figure 7.** Temperature and rainfall throughout the duration of the trial, with arrows showing treatment application dates, Hereford, 2014.

Fern assessments were carried out on 26 August, 10 September, 26 September and 16 October and disease levels were low with 0.01% incidence in the untreated plots in stem bases and canopy and 0.01% severity. In the dry summer of 2014 *Stemphylium* was well controlled in the crop by the commercial fungicide programmes used and no significant differences were seen from the addition of urea treatments (data not shown). Vigour was assessed at the same time as purple spot, also showing no significant differences. However, a trend was seen for higher vigour scores in the treated plots as shown in Figure 8 but this was more likely a nitrogen effect rather than disease reduction effect, as disease was not a limiting factor.

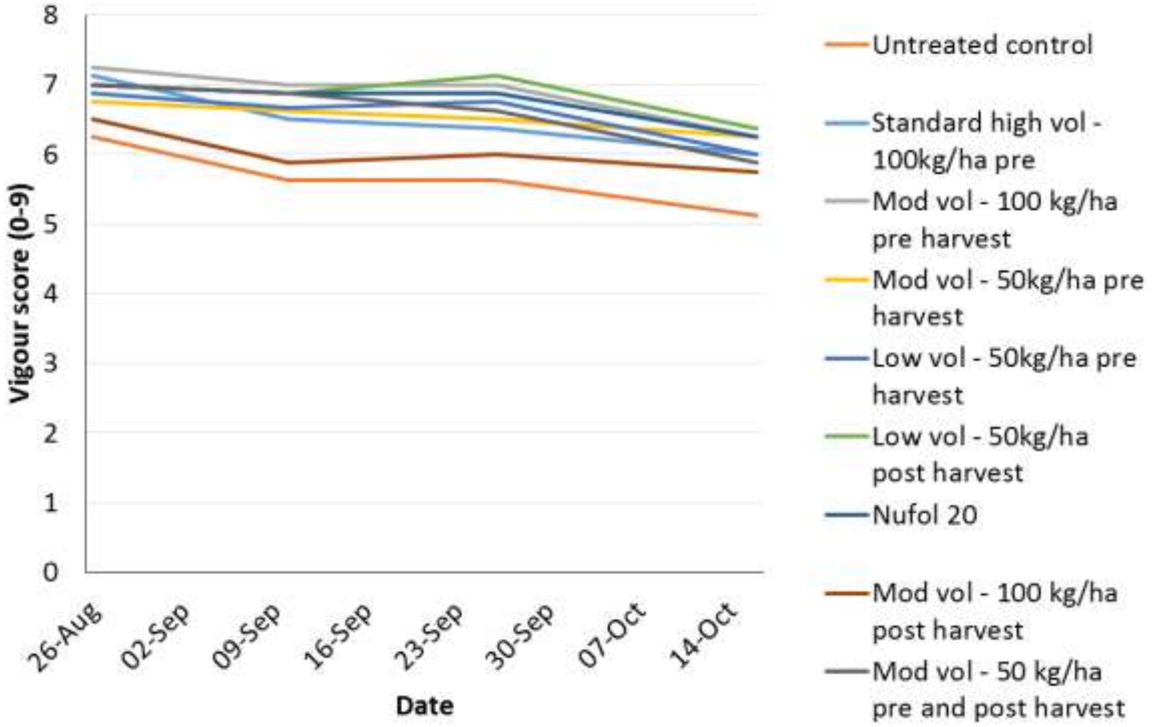


**Figure 8.** Vigour of fern canopy at assessments 26 August, 10 September, 26 September and 16 October, Hereford, 2014.

<u>Norfolk site</u>

Two spear assessments were carried out on 14 May and 5 June, the third spear assessment was not carried out as there was very little purple spot seen on the spears through June when debris was being collected. There was only a trace of purple spot seen on the spears (0.01% incidence) on 5 June so these data are not presented. At the spear assessment carried out on 14 May the disease levels were still low with only 9.6% of spears affected by purple spot in the untreated (Figure 9). As in 2013 in FV 341b the weather was considerably drier at the eastern site during harvest, and these less favourable conditions for *Stemphylium* infection led to less symptoms on the spears with severity of purple spot only 0.2% stem area affected in the untreated plots. Temperature and rainfall at the time of application and throughout the trial are shown in Figure 10. All treatments except Nufol 20 appeared to reduce incidence on the spears, but these reductions did not achieve statistical significance or consistent effects in assessments at both sites.

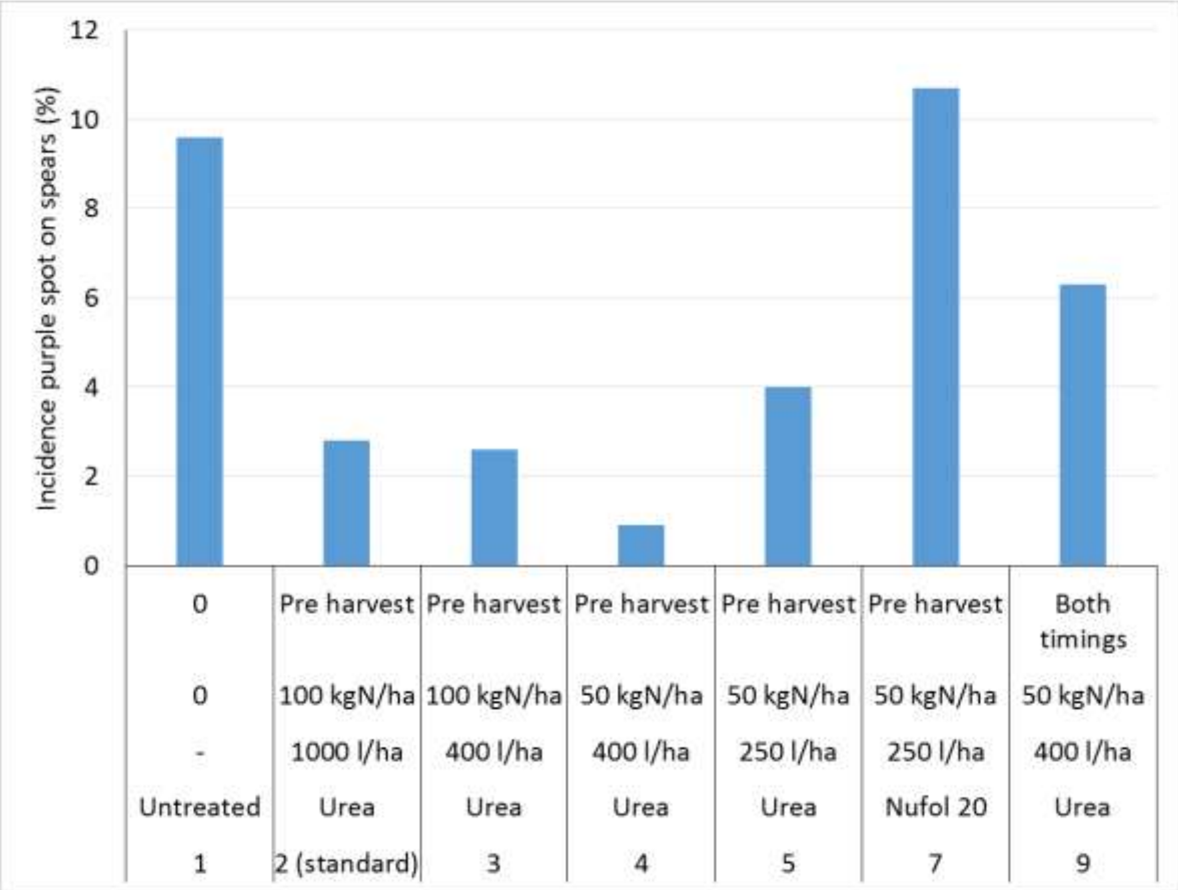


**Figure 9.** Incidence of purple spot on the emerging spears at assessment on 14 May, Norfolk, 2014. In this figure results for treatments 6 and 8 are not shown as they had not been applied at this point.

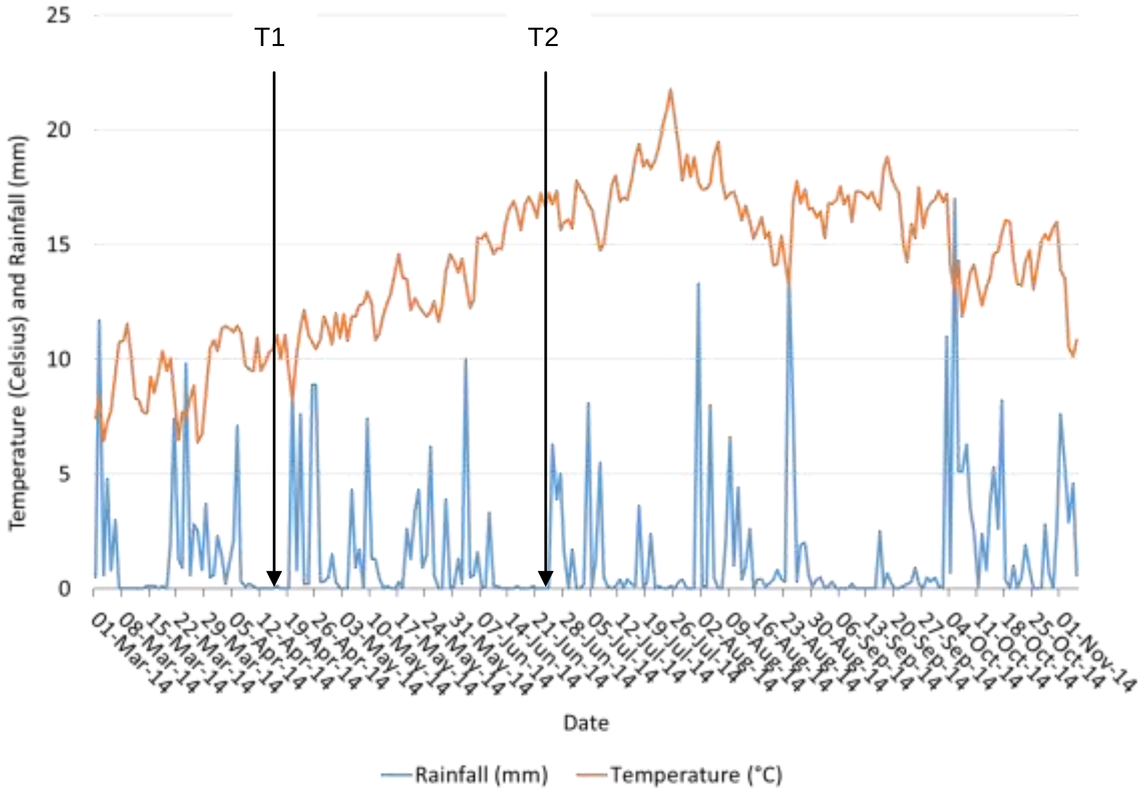


**Figure 10.** Temperature and rainfall throughout the duration of the trial, with arrows showing treatment application dates, Norfolk, 2014.

Once the crop was left to grow away to ferns after harvest, there was a greater incidence of purple spot in the ferns on the stem bases and canopy at the Norfolk site with 62.5% incidence in the untreated plots at the 1st assessment on 1 September, and 100% in the untreated plots at the 2nd assessment on 20 October. However, there were no significant differences between treatments in the incidence of purple spot. Severity of purple spot on 1 September recorded as % area of the stem base or canopy was moderate at 10.3% in the untreated on the stem bases and 5.3% in the canopy in the untreated plots (Figure 11 and 12). There were no significant differences between treatments for incidence or severity at either assessment.

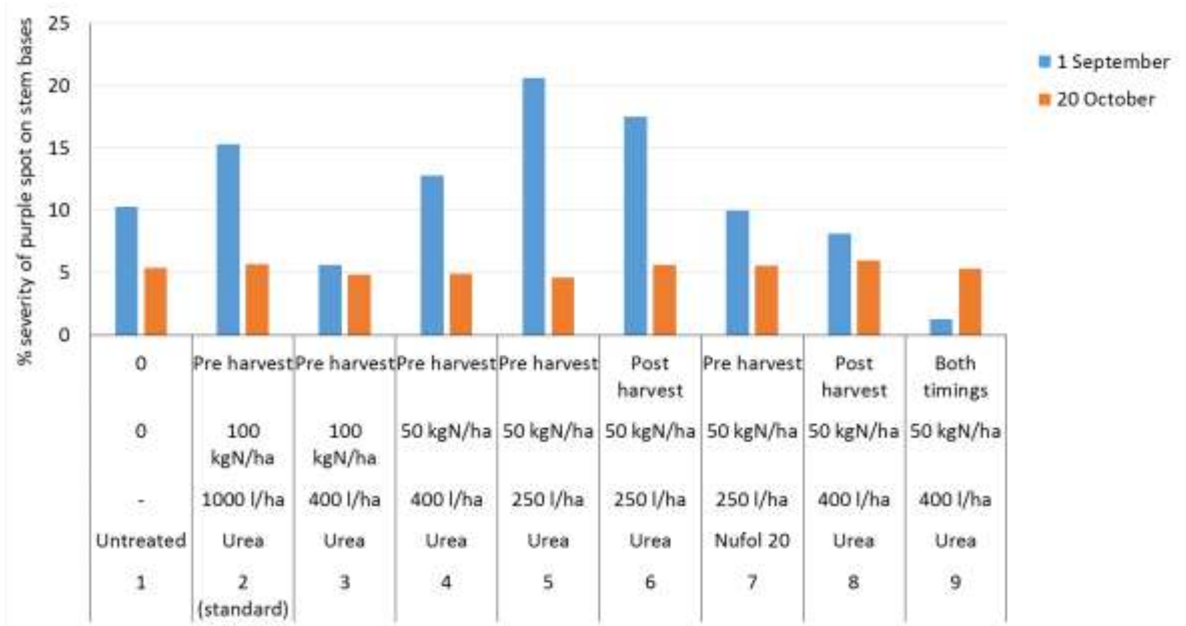


**Figure 11**. Severity of purple spot on the stem bases at assessment on 1 September and 20 October, Norfolk, 2014.

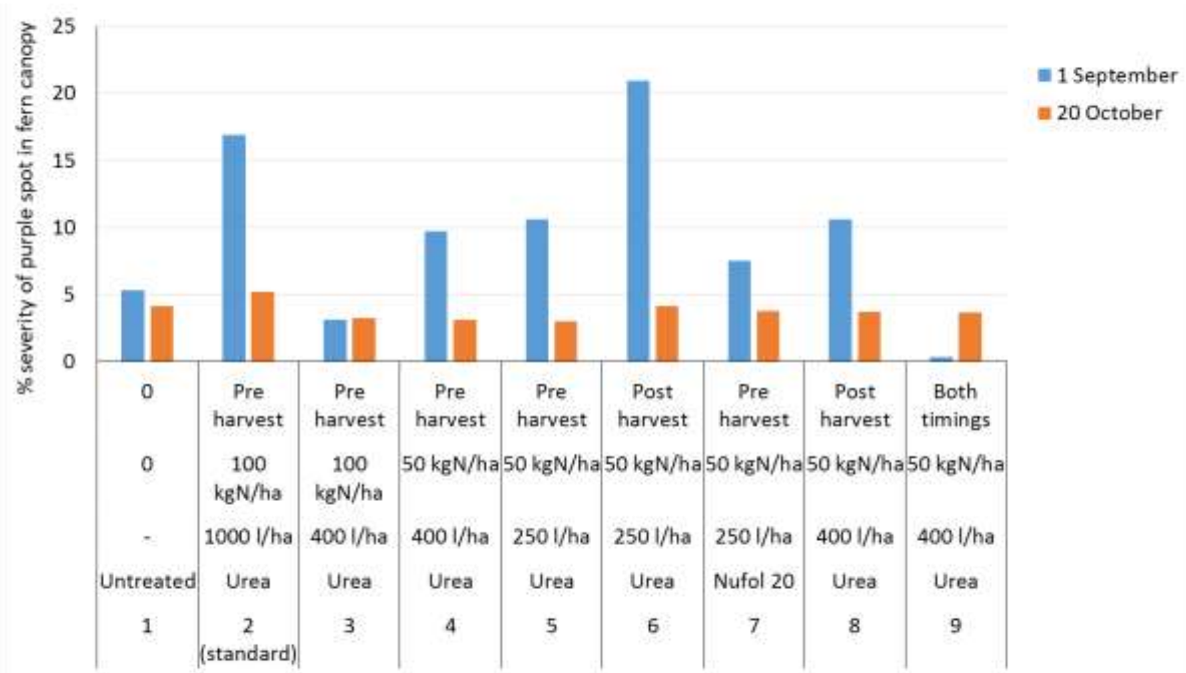


**Figure 12.** Severity of purple spot in the canopy at assessment on 1 September and 20 October, Norfolk, 2014.

Greenness and vigour of the fern were recorded, with no significant treatment effects. (Figure 13 and 14).

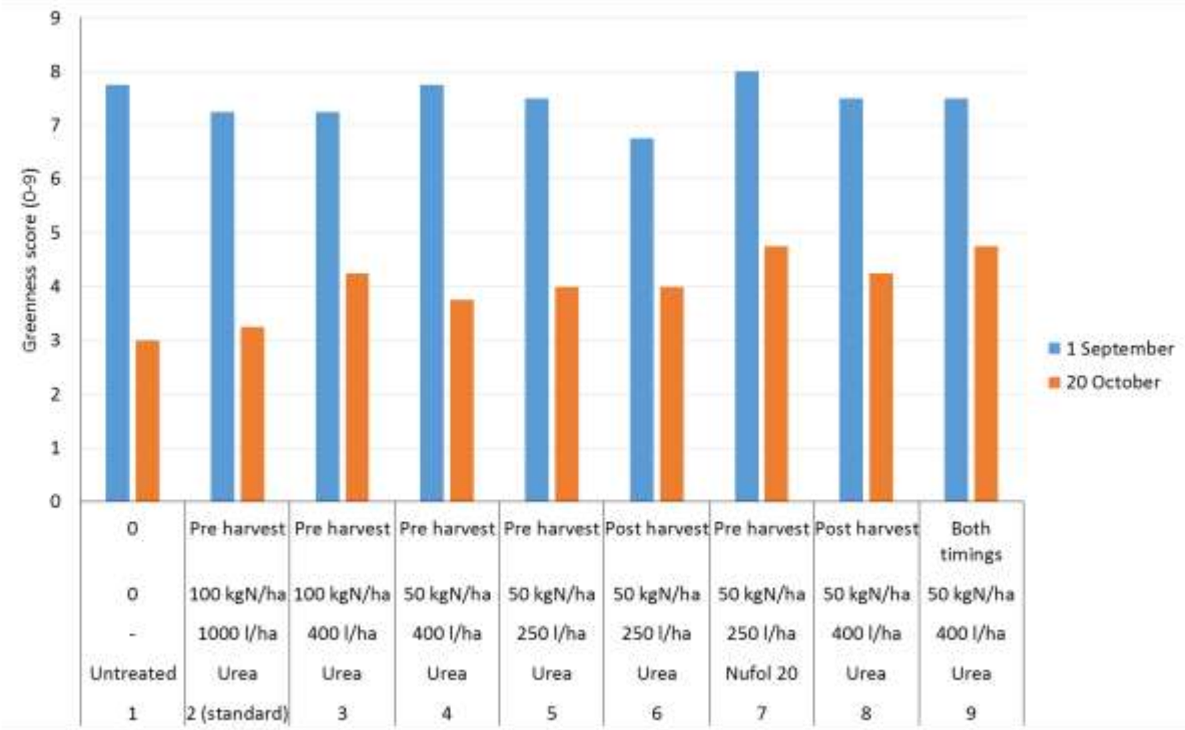


**Figure 13.** Greenness of the canopy at assessment on 1 September and 20 October (0 = brown, 9 = green), Norfolk, 2014

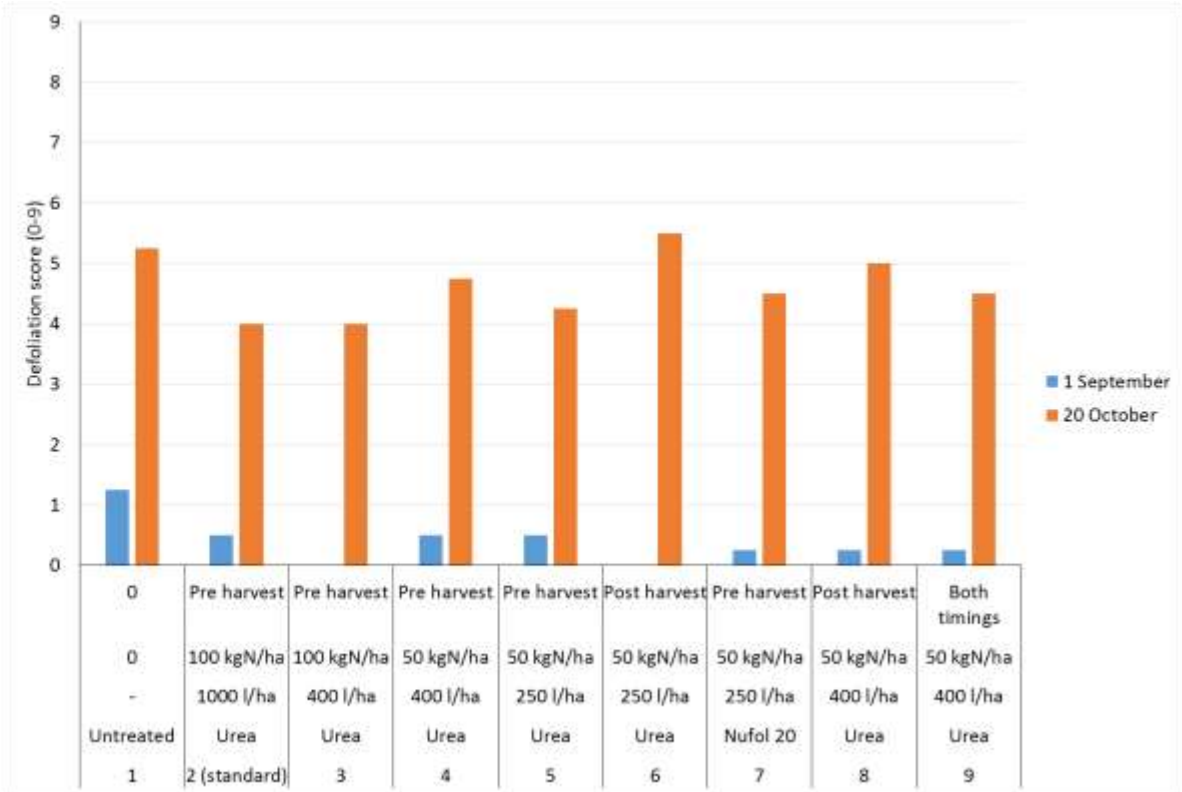


**Figure 14.** Defoliation of the canopy at assessment on 1 September and 20 October (0 = no needles, 9 = full needles), Norfolk, 2014

### Objective 3: – Assessing any additional efficacy from the addition of adjuvants to urea treatments for reduction of ascospore release from crop debris by *S. vesicarium* – pot trial.

*Stemphylium* infected asparagus debris with abundant pseudothecia present was collected from the grower site in Norfolk on 18 March. The treatments were applied on 28 April. Spore release was high in the untreated plots at 100% at 1 day and 14 days after the treatments were applied, then subsequently fell to 0% at 1 and 2 months after application. (Figure 15a N.B. The data line for the untreated tracks that of Urea and Tween 20 and is difficult to see). Proportions of ascospore release varied between the treatments (Figure 15b - c) and there were significant differences between percentages in nil, low and high categories (P = 0.006 to <0.001) at 1 and 14 days after treatment.

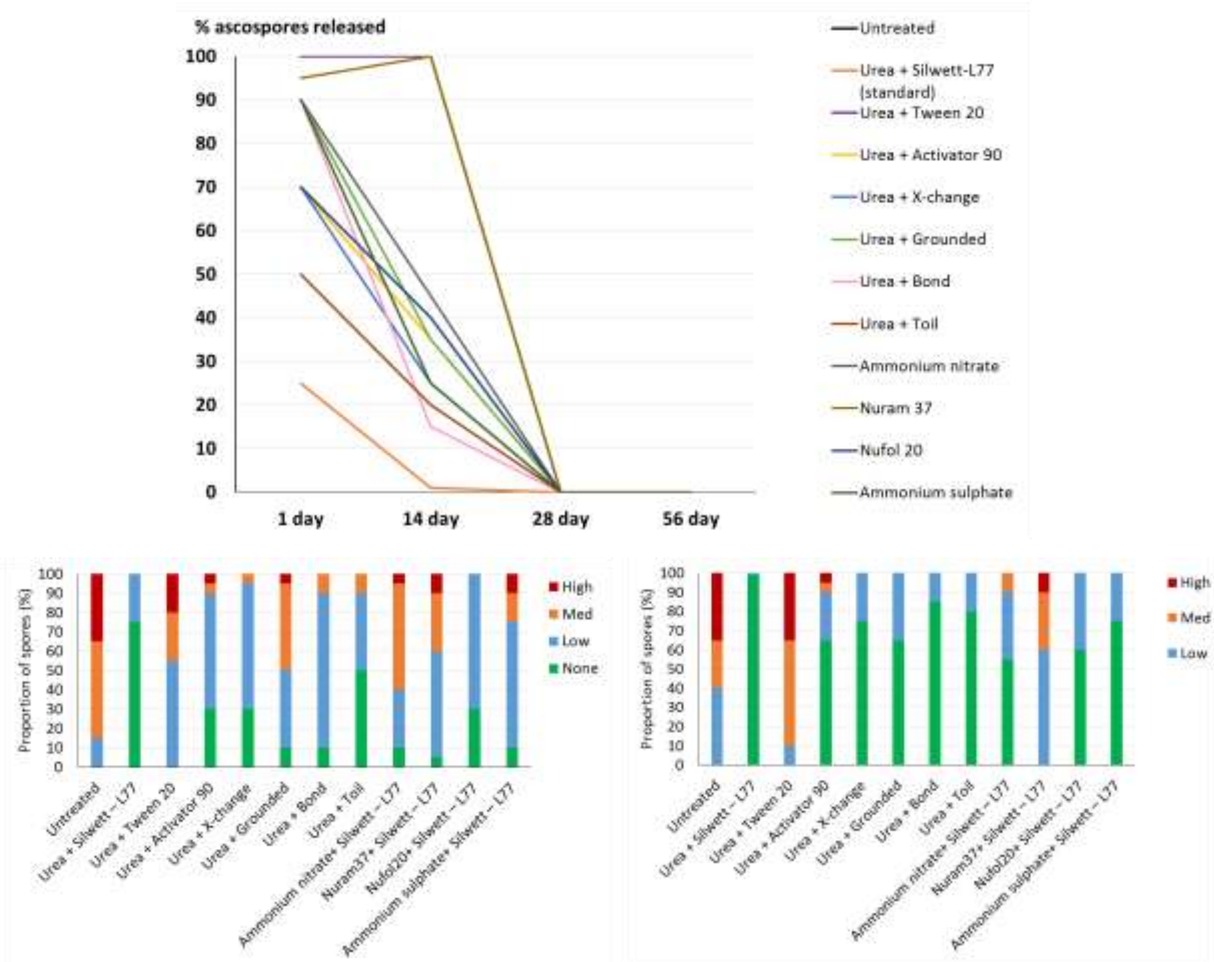


**Figure 15. a (Top):**Trend in percentage of ascospores released from resting bodies on debris for a period of 8 weeks after urea treatments applied at ADAS Boxworth, April to June 2014; **b (Bottom left):** Proportion of ascospores released in categories none, low, medium and high 1 day after treatment application; **c (Bottom right):** Proportion of ascospores released in categories none, low, medium and high 14 days after treatment application; (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece). N.B. There was no ascospore release at 56 days after treatment application

At 1 and 14 days after the treatments were applied, a significant reduction in spore release was seen between the untreated plots and those where urea was applied with Silwett-L77, Toil, Activator-90, and X-Change (P 0.006, full data analysis in Appendix 2). Additionally, Nufol 20 gave a significant reduction in spore release at both 1 and 14 days after application (Figure 15). Also at 14 days after application, treatments of urea with adjuvants Grounded and Bond gave a significant reduction of spore release, as did ammonium nitrate and ammonium sulphate. However, the standard treatment of urea applied with Silwett-L77 still gave the greatest reduction in spore release. There was no spore release from any of the treatments after 4 and 8 weeks post treatment application, and thus efficacy at these timings could not be assessed.

### Objective 4: Assessing influence of water volume on the urea treatments for reduction of ascospore release from crop debris by *S. vesicarium* – pot trials.

The asparagus debris for the water volumes trial was collected from Norfolk on 18 March and set up on the hard standing at ADAS Boxworth in the same trial design as the one on adjuvants. Spore release was high in the untreated plots at 100% at 1 day and 14 days after the treatments were applied, then subsequently fell to 80% at 1 month after application and finally to 0% at 2 months after application (Figure 16a). There were significant differences between treatments at 1 day after the spray application, with 800 L/ha and 200 L/ha being the only treatments with nil spore release. At 14 days after the urea was applied all urea treatments significantly reduced spores, with 1000 L giving the greatest reduction. By 28 days after application there were no significant differences between any of the treatments, and by 56 days after application, spore release had ceased in all plots (Figures 16b – d).

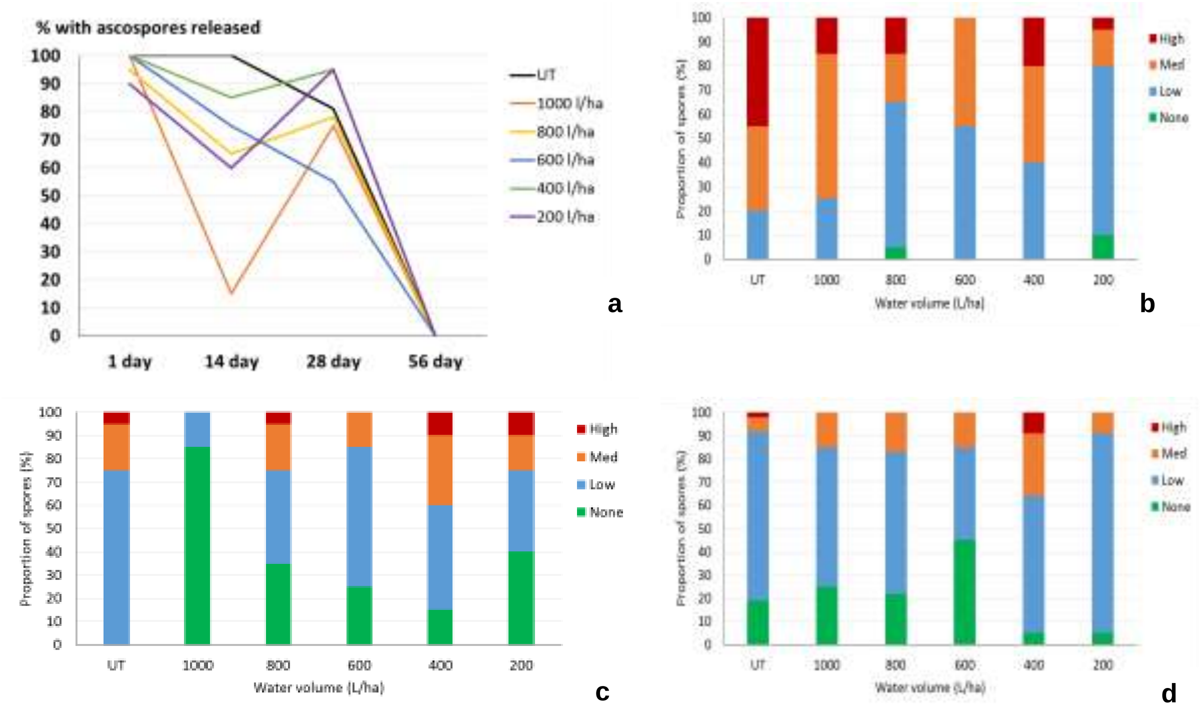


**Figure 16. a:**Trend in percentage of ascospores released from resting bodies on debris for a period of 8 weeks after urea treatments applied at ADAS Boxworth, April to June 2014; **b:** Proportion of ascospores released in categories none, low, medium and high 1 day after treatment application; **c:** Proportion of ascospores released in categories none, low, medium and high 14 days after treatment application; (Low = 0-50 spores per piece, Moderate = 50-150 spores per piece, High > 150 spores per piece). N.B. There was no ascospore release at 56 days after treatment application

## Discussion

The aim of this project was to improve control of *Stemphylium* purple spot on asparagus by decreasing overwintering inoculum on crop debris

This was determined against the objectives:

(i) To determine the most effective granular urea rate, form and timing for reduction of the level and persistence of spore production from debris,
(ii) And determine the most effective granular urea rate, form and timing for reduction of purple spot on the emerging spears and ferns,
(iii) To assess whether additional efficacy is gained from addition of adjuvants,
(iv) To assess the influence of water volume on efficacy of the urea.

## Efficacy of urea treatments: rates, forms and timing for reduction of ascospore release from crop debris by *S. vesicarium*

Urea applied as a spray significantly reduced spore production at the Norfolk site in 2014. At 1 day after treatment, urea (100 kg N/ha in 400 L water) significantly reduced spore release from debris compared with the untreated control. (Table 8). At 14 days, although spore production was not completely suppressed, all urea treatments significantly reduced the intensity of spore release to the low category (0-50 spores per piece of debris), compared with the untreated which showed approximately 40% spore release in the medium category (50-150 spores per piece of debris) (Figure 4c back in the field trial section). At 28 days after treatment at the Norfolk site, although there were no significant differences in the suppression of spore production, there was a trend for urea applied at 100 kg N/ha in 400 L/ha to give the greatest reduction in spore release of 72% compared to the untreated. At the experimental site in Hereford, there was no significant reduction in spores released from the debris. This is differing to the results seen in 2013 where the application of urea at 100 kg N/ha post-ridging gave a significant reduction in spore release for up to 28 days post application at both trial sites in the east and west (Table 9). In 2014 the treatments were not as effective or persistent, or alternatively the weather may not have been so conducive to ascospore release when the debris was collected for assessment.

**Table 8.** Summary of the effect of urea treatments on production of *Stemphylium vesicarium* ascospores from asparagus crop debris after pre-harvest applications – 2014

| Treatment and application timing | | | | % spore production relative to the untreated (= 100%) at assessment timings as indicated after treatment | | | | |
|---|---|---|---|---|---|---|---|---|
| | | *Water volume (L/ha)* | *kgN/ha* | Hereford | | | Norfolk | |
| | | | | 1 Days after T1 | 14 Days after T1 | 56 Days after T1 | 1 Days after T1 | 28 Days after T1 |
| 1 | Untreated | 0 | 0 | 100 | 100 | 100 | 100 | 100 |
| 2 | Urea | 1000 | 100 | 73 | 100 | 233 | 100 | 57 |
| 3 | Urea | 400 | 100 | 45 | 100 | 166 | **63** | 28 |
| 4 | Urea | 400 | 50 | 100 | 135 | 266 | 105 | 100 |
| 5 | Urea | 250 | 50 | 82 | 227 | 183 | 89 | 114 |
| 7 | Nufol 20 | 250 | 50 | 64 | 208 | 166 | 74 | 72 |
| 9 | Urea | 400 | 50 | 64 | 112 | 216 | 95 | 100 |

Values in bold are significantly different from the untreated
Results for treatments 6 and 8 are not shown as they had not been applied at this point.

**Table 9.** Summary of the effect of urea and Perlka treatments on production of *Stemphylium vesicarium* ascospores from asparagus crop debris – 2013 (from FV 341b)

| Treatment and application timing | | | | | % spore production relative to the untreated (= 100%) at 1 and 2 months after treatment | | | |
|---|---|---|---|---|---|---|---|---|
| | | T1 | T2 *kgN/ha* | T3 | Hereford | | Norfolk | |
| | | | | | 28 Days after T1 | 56 Days after T1 | 28 Days after T1 | 56 Days after T1 |
| 1 | Untreated | 0 | 0 | 0 | 100 | 100 | 100 | 100 |
| 2 | Urea | 0 | 100* | 0 | **15** | **24** | **45** | 108 |
| 3 | Urea | 0 | 100 | 0 | **63** | **67** | **34** | 90 |
| 4 | Urea | 0 | 50 | 0 | 94 | 95 | 76 | 82 |
| 5 | Urea | 50 | 0 | 0 | **69** | 87 | **51** | 87 |
| 6 | Urea | 0 | 50 | 50 | **46** | **49** | **50** | **49** |
| 7 | Urea | 50 | 50 | 0 | **52** (38) | 98 (35) | **32** (84) | 47 (74) |
| 8 | Perlka | 60** | 0 | 0 | 108 | 133 | 97 | 113 |
| 9 | Perlka | 0 | 60** | 0 | 73 | 89 | **24** | **13** |

Values in bold are significantly different from the untreated
Figures in brackets are days after T2 treatment applied
*1,000 L/ha **Top dressed

At both sites, there was low or no spore release from the debris after the 2nd application of treatments was made and no significant differences were seen between treatments applied. This is likely to be related to the maturation of ascospores which was monitored in HDC project FV 341. This showed that from January to March, mature pseudothecia contained abundant ascospores, which were starting to erupt through the epidermal stem tissues and that the proportion of pseudothecia erupting through skin tissue increased from January to March. Therefore the majority of spores are released just prior to or during the crop harvest which takes place from April to June.

In the pot experiments carried out at ADAS Boxworth significant differences could be seen between the adjuvants used at 14 days after application. Of the adjuvants trialled with the urea, Silwett-L77 facilitated the greatest spore suppression, and Activator-90, X-change, Grounded and Bond also significantly reduced the spore release at 14 days after application. This result provides a range of choices to the grower of products available in the chemical store which could be substituted for Silwett-L77 if more convenient. Alternative forms of nitrogen were also evaluated for their effects on spore release in the trial and ammonium nitrate, ammonium sulphate and Nufol 20 also significantly reduced spore release but did not perform as well as the urea and Silwett-L77 combination. In the field experiments, Nufol 20 did not give a significant reduction in spore release as it did in the pot experiments (Table 8 above). The influence of water volumes on spore release was also tested in an adjacent experiment with significant differences seen between treatments at 1 day and 14 days after application. The greatest reduction in spore release compared to the untreated was at 14 days after application when urea was applied in 1000 L/ha. There was no significant difference between treatments from 200 to 800 L/ha in the experiment, suggesting that once volume is reduced below 1000 L/ha then the choice of volume used does not have a large influence on increasing efficacy. The trend for a high volume such as 1000 L/ha giving the greatest suppression in spore release was also seen in work completed by Humpherson-Jones and Burchill (1982), and Gladders (1980) to control blackleg (*Leptosphaeria maculans*) on oilseed rape.

The exact mode of urea is unknown and this means it is not as predictable to use as conventional fungicides. However, it has some effect and is used in other crops for control of pathogens such as in apple orchards where urea has been used for a number of years for control of apple scab (*V. inaequalis*). Although the main mode of action is to hasten the breakdown of leaves, and so destroy the host on which the pathogen needs to survive, it is also acknowledged that the urea changes the microbial and chemical populations on the leaf which could be affecting ascospore release (Ross and Burchill, 1968). In work by Sutton and MacHardy (1993) bacterial populations on leaves of apple trees were increased by the urea, and this could play a part in spore suppression of some pathogenic organisms. Other work has shown urea to have a direct effect on the pathogen *V. inaequalis* (apple scab), reducing the maturation of pseudothecia (Mezska and Bielenin, 2006). Where the exact mode of action is unknown, and a number of unpredictable biological factors could be contributing to the spore reduction effects it is more difficult to reliably predict the efficacy of urea as a control measure for *Stemphylium* purple spot.

### Field trials - Efficacy of urea treatments: rates, forms and timing for reduction of purple spot in the emerging crop and ferns.

In the dry summer of 2014 *Stemphylium* was well controlled in the crop by the commercial fungicide programmes used. Few significant treatment differences were seen from the addition of urea treatments at any of the assessments because conditions were dry, and not conducive to infection with *Stemphylium* purple spot.

Although treatments applied post-harvest and before the development of ferns did not significantly reduce spore release compared with the untreated at either site, the associated monitoring of spore release carried out through fern growth indicated that the crop in Norfolk in which spore release was seen, had a higher incidence and severity of *Stemphylium* purple spot in the ferns than in Hereford where no or very little spore release was seen.

## Conclusions

- In a Norfolk asparagus crop naturally infected with *S. vesicarium*, urea applied to soil and crop debris before spear emergence in 2014 provided some suppression of ascospore release from infected debris. Of the treatments tested, urea (combined with Silwett-77) at the rate of 100 kg N/ha in 400 L/ha water gave the most consistent suppression of spore release, up to 14 days after treatment. This finding was supported by results from similar field trials in 2013 (FV 341 b). Results from a second field site in Herefordshire in 2014 were inconclusive since spore release from debris was at lower levels.

- Urea treatments (pre- and post-harvest) did not reduce the incidence or severity of purple spot on spears or fern in 2014. It should be noted, however, that disease pressure was low during the harvest season and was subsequently controlled by fungicides during fern growth, such that levels of Stemphylium were low for all treatments including the untreated control. Similarly in 2013, there was only a limited reduction in Stemphylium levels due to urea treatment.

- Applying the majority of the asparagus crop's nitrogen requirement, (bearing in mind the NMax in an NVZ is 180 kg N/ha) as urea at 100 kg N/ha just prior to spear growth is not environmentally advisable given that N offtake by the spears at this point in growth is very low, and would increase the risk of loss of N to leaching in a wet spring. Therefore, to reduce spore release and possibly purple spot in the emerging crop without compromising crop nutrition or environmental risk, applying urea twice as 50 kg N/ha prior to and during harvest instead of nitrogen as a fertiliser could still be considered.

- There appears to be no gain from an application of urea between spear harvest and fern growth, as the majority of ascospore release occurs prior to or during spear harvest and urea sprays if used would be best targeted to a pre-harvest timing.

- This timing of spore release from debris indicates that the highest risk of infection by *S. vesicarium* is to the spears during harvest. But ultimately if protectant fungicides are not applied post-harvest early enough before canopy closure to protect stem bases, infection from debris is still a risk during later fern development as it is the initial source of infection as shown in work in FV 341.

- The work indicates that Silwett-L77 is the best tank mix partner for urea for greatest spore suppression, but if this is not conveniently available to the grower, Activator-90, X-change, Grounded and Bond offer viable alternatives.

- Although Nufol 20 gave a significant reduction in spore release in the pot experiments, this significant effect was not borne out in the field trials. However, if used this formulation would be a more practical option for larger scale growers.

- Applying urea in 1000 L/ha gives the greatest efficacy but this is not practical, especially for larger growers. Once volumes were reduced below 1000 L/ha then the choice of volume used did not have a large influence on increasing efficacy.

## Further work

Further work may be desirable to determine the influence of external variables such as weather, environment and intensity of spore release on the efficacy of the urea treatment. This would be achieved by applying the most promising treatment of 100 kg N/ha in 400L water at several grower sites as a split application of 50 kg N/ha applied twice in place of the usual fertiliser application, and then monitoring spore release, environmental parameters and efficacy. This would aim to determine in what situations urea would provide benefit to the grower for greatest spore suppression, and the ideal conditions for application.

## Knowledge and Technology Transfer

Short written update for Asparagus Growers Association meeting in October 2014
HDC News article (in preparation)

 

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# Appendices

## Appendix 1 – Trial diaries

Hereford site

| Date | Task/application |
|---|---|
| 24th March 2014 | Trial marked out |
| **1st April** | **Timing 1 treatments applied** |
| 2nd April | T1 Day 1 debris collected |
| 7th April | Spore count |
| 15th April | T1 Day 14 debris collected |
| 21st April | Spore count |
| 30th April | T1 Day 28 debris collected |
| 6th May | Spore count |
| 13th May | Early Stemphylium purple spot assessment on asparagus spears |
| 28th May | T1 Day 56 debris collected |
| 2nd June | Spore count |
| 7th June | Late Stemphylium purple spot assessment on asparagus spears |
| **3rd July** | **Timing 2 treatments applied** |
| 7th July | T2 Day 1 debris collected |
| 14th July | Spore count |
| 14th July | T2 Day 14 debris collected |
| 21st July | Spore count |
| 5th August | T2 Day 28 debris collected |
| 12th August | Stemphylium assessment on ferns- no disease present |
| 15th August | Spore count |
| 1st September | T2 Day 56 debris collected |
| 8th September | Spore count |
| 10th September | Early Stemphylium assessment on ferns |
| 16th October | Late Stemphylium assessment on ferns |
| 20th October 2014 | Trial completed and markers collected |

Norfolk site

| Date | Task/application |
|---|---|
| 18th March 2014 | Trial marked out |
| **15th April 2014** | **Timing 1 treatments applied** |
| 16th April | T1 Day 1 debris collected |
| 19th April | Spore count |
| 29th April | T1 Day 14 debris collected |
| 2nd May | Spore count |
| 14th May | T1 Day 28 debris collected |
| 14th May | Early Stemphylium purple spot assessment on asparagus spears |
| 18th May | Spore count |

| | |
|---|---|
| 5th June | Late Stemphylium purple spot assessment on asparagus spears |
| 16th June | T1 Day 56 debris collected |
| 18th June | Spore count |
| **25th June** | **Timing 2 treatments applied** |
| 26th June | T2 Day 1 debris collected |
| 29th June | Spore count- |
| 9th July | T2 Day 14 debris collected |
| 12th July | Spore count |
| 23rd July | T2 Day 28 debris collected |
| 26th July | Spore count |
| 1st September | T2 Day 56 debris collected |
| 1st September | Early Stemphylium assessment on ferns |
| 3rd September | Spore count |
| 20th October | Late Stemphylium assessment on ferns |
| 20th October 2014 | Trial completed and markers collected |

## Appendix 2: Statistical tables – spore counts

The standard errors vary for each treatment, so the 95% confidence intervals for each treatment were calculated to make it easier to identify differences between treatments. If when comparing two treatments, the confidence intervals do not overlap we can say that they are significantly different. This is denoted where the letter is different from the untreated.

Confidence interval (CI) was estimated using standard errors (SE) for each predicted value (CI = SE x $t_{95\%}$ , where for the field trials, df = 18 after timing 1, and 24 after timing 2; and for the pot trials, df = 33 for adjuvants, and 15 for water volumes experiments).

The predictions were converted to % ascospores released by converting the fractions to percentages, by subtracting the predicted value from 1 and multiplying by 100 to plot the graphs in the main body of the report.

<u>Hereford site</u>

Assessments after Timing 1 application

| | | **Day 1** | **Proportion with score No mycelium** | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.45 | 0.129 | 0.271029 | 0.18 | 0.72 | a |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 1000L/100 | 2 | 0.6 | 0.135 | 0.283635 | 0.32 | 0.88 | a |
| 400L/100 | 3 | 0.75 | 0.1417 | 0.2977117 | 0.45 | 1.05 | a |
| 400L/50 | 4 | 0.45 | 0.129 | 0.271029 | 0.18 | 0.72 | a |
| 250L/50 | 5 | 0.55 | 0.13 | 0.27313 | 0.28 | 0.82 | a |
| Nufol 20 | 7 | 0.65 | 0.1398 | 0.2937198 | 0.36 | 0.94 | a |
| 400L/50 x2 | 9 | 0.65 | 0.1398 | 0.2937198 | 0.36 | 0.94 | a |

| | | **Day 1** | **Proportion with score L** | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.5 | 0.12554 | 0.26375954 | 0.24 | 0.76 | a |
| 1000L/100 | 2 | 0.35 | 0.12032 | 0.25279232 | 0.10 | 0.60 | a |
| 400L/100 | 3 | 0.05 | 0.06325 | 0.13288825 | -0.08 | 0.18 | a* |
| 400L/50 | 4 | 0.25 | 0.11255 | 0.23646755 | 0.01 | 0.49 | a |
| 250L/50 | 5 | 0.4 | 0.12274 | 0.25787674 | 0.14 | 0.66 | a |
| Nufol 20 | 7 | 0.05 | 0.06356 | 0.13353956 | -0.08 | 0.18 | a* |
| 400L/50 x2 | 9 | 0.1 | 0.08467 | 0.17789167 | -0.08 | 0.28 | a |
| | | **Day 1** | **Proportion with score M** | | | | |
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.05 | 0.03045 | 0.06397545 | -0.01 | 0.11 | a |
| 1000L/100 | 2 | 0 | 0.00013 | 0.00027313 | 0.00 | 0.00 | b |
| 400L/100 | 3 | 0.15 | 0.04541 | 0.09540641 | 0.05 | 0.25 | a |
| 400L/50 | 4 | 0.15 | 0.04541 | 0.09540641 | 0.05 | 0.25 | a |
| 250L/50 | 5 | 0 | 0.00013 | 0.00027313 | 0.00 | 0.00 | b |
| Nufol 20 | 7 | 0.25 | 0.04908 | 0.10311708 | 0.15 | 0.35 | c |
| 400L/50 x2 | 9 | 0.2 | 0.0482 | 0.1012682 | 0.10 | 0.30 | a |

| | | **Day 1** | **Proportion with score H** | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0 | 0.11665 | 0.00014 | 0.00 | 0.00 | a |
| 1000L/100 | 2 | 0.05 | 0.08139 | 0.03556 | 0.01 | 0.09 | a |
| 400L/100 | 3 | 0.05 | 0.09659 | 0.03556 | 0.01 | 0.09 | a |
| 400L/50 | 4 | 0.15 | 0.10795 | 0.05289 | 0.10 | 0.20 | a |
| 250L/50 | 5 | 0.05 | 0.12329 | 0.03556 | 0.01 | 0.09 | a |
| Nufol 20 | 7 | 0.05 | 0.00087 | 0.03556 | 0.01 | 0.09 | a |
| 400L/50 x2 | 9 | 0.05 | 0.05934 | 0.03556 | 0.01 | 0.09 | a |

| | | **Day 14** | **Proportion with score No mycelium** | | | | |
|---|---|---|---|---|---|---|---|
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 1 | 0.00118 | 0.00247918 | 1.00 | 1.00 | a |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 1000L/100 | 2 | 1 | 0.00118 | 0.00247918 | 1.00 | 1.00 | a |
| 400L/100 | 3 | 1 | 0.00126 | 0.00264726 | 1.00 | 1.00 | a |
| 400L/50 | 4 | 0.4347 | 0.18943 | 0.39799243 | 0.04 | 0.83 | b |
| 250L/50 | 5 | 0.7393 | 0.17211 | 0.36160311 | 0.38 | 1.10 | ab |
| Nufol 20 | 7 | 0.4847 | 0.19115 | 0.40160615 | 0.08 | 0.89 | b |
| 400L/50 x2 | 9 | 0.8951 | 0.12401 | 0.26054501 | 0.63 | 1.16 | ab |

| | | **Day 14** | **Proportion with score L** | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0 | 0.00071 | 0.00149171 | 0.00 | 0.00 | a |
| 1000L/100 | 2 | 0 | 0.00071 | 0.00149171 | 0.00 | 0.00 | a |
| 400L/100 | 3 | 0 | 0.00064 | 0.00134464 | 0.00 | 0.00 | a |
| 400L/50 | 4 | 0.2511 | 0.13606 | 0.28586206 | -0.03 | 0.54 | a |
| 250L/50 | 5 | 0.151 | 0.11332 | 0.23808532 | -0.09 | 0.39 | a |
| Nufol 20 | 7 | 0.3509 | 0.14912 | 0.31330112 | 0.04 | 0.66 | b |
| 400L/50 x2 | 9 | 0.1008 | 0.09587 | 0.20142287 | -0.10 | 0.30 | a |
| | | **Day 14** | **Proportion with score M** | | | | |
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0 | 0.00013 | 0.00027313 | 0.00 | 0.00 | a |
| 1000L/100 | 2 | 0 | 0.00013 | 0.00027313 | 0.00 | 0.00 | a |
| 400L/100 | 3 | 0 | 0.00016 | 0.00033616 | 0.00 | 0.00 | a |
| 400L/50 | 4 | 0.16154 | 0.07498 | 0.15753298 | 0.00 | 0.32 | b |
| 250L/50 | 5 | 0.10769 | 0.0638 | 0.1340438 | -0.03 | 0.24 | a |
| Nufol 20 | 7 | 0.10769 | 0.0638 | 0.1340438 | -0.03 | 0.24 | a |
| 400L/50 x2 | 9 | 0 | 0.00016 | 0.00033616 | 0.00 | 0.00 | a |

| | | **Day 14** | **Proportion with score H** | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0 | 0.00013 | 0.00027313 | 0.00 | 0.00 | a |
| 1000L/100 | 2 | 0 | 0.00013 | 0.00027313 | 0.00 | 0.00 | a |
| 400L/100 | 3 | 0 | 0.00016 | 0.00033616 | 0.00 | 0.00 | a |
| 400L/50 | 4 | 0.16154 | 0.07498 | 0.15753298 | 0.00 | 0.32 | b |
| 250L/50 | 5 | 0 | 0.0638 | 0.1340438 | -0.13 | 0.13 | a |
| Nufol 20 | 7 | 0.05385 | 0.0638 | 0.1340438 | -0.08 | 0.19 | a |
| 400L/50 x2 | 9 | 0 | 0.00016 | 0.00033616 | 0.00 | 0.00 | a |

| | | **Day 56** | **Proportion with score O** | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| UT | 1 | 0.15 | 0.10047 | 0.21108747 | -0.06 | 0.36 | a |
| 1000L/100 | 2 | 0.15 | 0.10047 | 0.21108747 | -0.06 | 0.36 | a |
| 400L/100 | 3 | 0.3 | 0.12802 | 0.26897002 | 0.03 | 0.57 | a |
| 400L/50 | 4 | 0.15 | 0.10047 | 0.21108747 | -0.06 | 0.36 | a |
| 250L/50 | 5 | 0.15 | 0.10047 | 0.21108747 | -0.06 | 0.36 | a |
| Nufol 20 | 7 | 0.2 | 0.11225 | 0.23583725 | -0.04 | 0.44 | a |
| 400L/50 x2 | 9 | 0 | 0.00091 | 0.00191191 | 0.00 | 0.00 | a |

| | | **Day 56** | **Proportion with score No mycelium** | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.55 | 0.1973 | 0.4145273 | 0.14 | 0.96 | a |
| 1000L/100 | 2 | 0.15 | 0.142 | 0.298342 | -0.15 | 0.45 | a |
| 400L/100 | 3 | 0.2 | 0.1587 | 0.3334287 | -0.13 | 0.53 | a |
| 400L/50 | 4 | 0.0501 | 0.0854 | 0.1794254 | -0.13 | 0.23 | a |
| 250L/50 | 5 | 0.3 | 0.1815 | 0.3813315 | -0.08 | 0.68 | a |
| Nufol 20 | 7 | 0.3 | 0.1819 | 0.3821719 | -0.08 | 0.68 | a |
| 400L/50 x2 | 9 | 0.35 | 0.1892 | 0.3975092 | -0.05 | 0.75 | a |
| | | **Day 56** | **Proportion with score L** | | | | |
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.15 | 0.1294 | 0.2718694 | -0.12 | 0.42 | a |
| 1000L/100 | 2 | 0.25 | 0.1567 | 0.3292267 | -0.08 | 0.58 | a |
| 400L/100 | 3 | 0.25 | 0.1567 | 0.3292267 | -0.08 | 0.58 | a |
| 400L/50 | 4 | 0.2 | 0.1448 | 0.3042248 | -0.10 | 0.50 | a |
| 250L/50 | 5 | 0.25 | 0.1556 | 0.3269156 | -0.08 | 0.58 | a |
| Nufol 20 | 7 | 0.2 | 0.1448 | 0.3042248 | -0.10 | 0.50 | a |
| 400L/50 x2 | 9 | 0.2 | 0.1448 | 0.3042248 | -0.10 | 0.50 | a |

| | | **Day 56** | **Proportion with score M** | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.05 | 0.05873 | 0.12339173 | -0.07 | 0.17 | a |
| 1000L/100 | 2 | 0.35 | 0.12686 | 0.26653286 | 0.08 | 0.62 | a |
| 400L/100 | 3 | 0.1 | 0.08062 | 0.16938262 | -0.07 | 0.27 | a |
| 400L/50 | 4 | 0.35 | 0.12868 | 0.27035668 | 0.08 | 0.62 | a |
| 250L/50 | 5 | 0.25 | 0.11554 | 0.24274954 | 0.01 | 0.49 | a |
| Nufol 20 | 7 | 0.1 | 0.08062 | 0.16938262 | -0.07 | 0.27 | a |
| 400L/50 x2 | 9 | 0.1 | 0.08062 | 0.16938262 | -0.07 | 0.27 | a |

| | | **Day 56** | **Proportion with score H** | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |

 

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| UT | 1 | 0.1 | 0.0987 | 0.2073687 | -0.11 | 0.31 | a |
| 1000L/100 | 2 | 0.1 | 0.0988 | 0.2075788 | -0.11 | 0.31 | a |
| 400L/100 | 3 | 0.15 | 0.1167 | 0.2451867 | -0.10 | 0.40 | a |
| 400L/50 | 4 | 0.25 | 0.1397 | 0.2935097 | -0.04 | 0.54 | a |
| 250L/50 | 5 | 0.05 | 0.0722 | 0.1516922 | -0.10 | 0.20 | a |
| Nufol 20 | 7 | 0.2 | 0.1299 | 0.2729199 | -0.07 | 0.47 | a |
| 400L/50 x2 | 9 | 0.35 | 0.152 | 0.319352 | 0.03 | 0.67 | a |

Assessments after Timing 2 application

| | | **Day 56** | **Proportion with score O** | | | | |
|---|---|---|---|---|---|---|---|
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.55 | 0.168 | 0.346752 | 0.20 | 0.90 | a |
| 1000L/100 | 2 | 0.95 | 0.0763 | 0.1574832 | 0.79 | 1.11 | a |
| 400L/100 pre | 3 | 0.85 | 0.1241 | 0.2561424 | 0.59 | 1.11 | a |
| 400L/50 | 4 | 0.75 | 0.1484 | 0.3062976 | 0.44 | 1.06 | a |
| 250L/50 pre | 5 | 0.8 | 0.138 | 0.284832 | 0.52 | 1.08 | a |
| 250L/50 post | 6 | 0.6 | 0.1657 | 0.3420048 | 0.26 | 0.94 | a |
| Nufol 20 | 7 | 0.5 | 0.1687 | 0.3481968 | 0.15 | 0.85 | a |
| 400L/50 post | 8 | 0.9 | 0.105 | 0.21672 | 0.68 | 1.12 | a |
| 400L/50 x2 | 9 | 0.9 | 0.105 | 0.21672 | 0.68 | 1.12 | a |

| | | **Day 56** | **Proportion with score No mycelium** | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.45 | 0.168 | 0.346752 | 0.10 | 0.80 | a |
| 1000L/100 | 2 | 0.05 | 0.077 | 0.158928 | -0.11 | 0.21 | a |
| 400L/100 pre | 3 | 0.15 | 0.1241 | 0.2561424 | -0.11 | 0.41 | a |
| 400L/50 | 4 | 0.25 | 0.1484 | 0.3062976 | -0.06 | 0.56 | a |
| 250L/50 pre | 5 | 0.2 | 0.138 | 0.284832 | -0.08 | 0.48 | a |
| 250L/50 post | 6 | 0.4 | 0.1657 | 0.3420048 | 0.06 | 0.74 | a |
| Nufol 20 | 7 | 0.5 | 0.1687 | 0.3481968 | 0.15 | 0.85 | a |
| 400L/50 post | 8 | 0.1 | 0.1051 | 0.2169264 | -0.12 | 0.32 | a |
| 400L/50 x2 | 9 | 0.1 | 0.1051 | 0.2169264 | -0.12 | 0.32 | a |

Norfolk site

Assessments after Timing 1 application

| | | Day 1 | Proportion with score No mycelium | | | | |
|---|---|---|---|---|---|---|---|
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.05 | 0.04487 | 0.09427187 | -0.04 | 0.14 | a |
| 1000L/100 | 2 | 0.05 | 0.04487 | 0.09427187 | -0.04 | 0.14 | a |
| 400L/100 | 3 | 0.4 | 0.09629 | 0.20230529 | 0.20 | 0.60 | b |
| 400L/50 | 4 | 0 | 0.00041 | 0.00086141 | 0.00 | 0.00 | a |
| 250L/50 | 5 | 0.15 | 0.07237 | 0.15204937 | 0.00 | 0.30 | ab |
| Nufol 20 | 7 | 0.3 | 0.09099 | 0.19116999 | 0.11 | 0.49 | ab |
| 400L/50 x2 | 9 | 0.1 | 0.06128 | 0.12874928 | -0.03 | 0.23 | ab |

| | | Day 1 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.4 | 0.1609 | 0.3380509 | 0.06 | 0.74 | a |
| 1000L/100 | 2 | 0.5 | 0.1642 | 0.3449842 | 0.16 | 0.84 | a |
| 400L/100 | 3 | 0.15 | 0.1179 | 0.2477079 | -0.10 | 0.40 | a |
| 400L/50 | 4 | 0.55 | 0.1634 | 0.3433034 | 0.21 | 0.89 | a |
| 250L/50 | 5 | 0.45 | 0.1634 | 0.3433034 | 0.11 | 0.79 | a |
| Nufol 20 | 7 | 0.6 | 0.161 | 0.338261 | 0.26 | 0.94 | a |
| 400L/50 x2 | 9 | 0.6 | 0.161 | 0.338261 | 0.26 | 0.94 | a |

| | | Day 1 | Proportion with score M | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.3 | 0.1125 | 0.2363625 | 0.06 | 0.54 | a |
| 1000L/100 | 2 | 0.35 | 0.1165 | 0.2447665 | 0.11 | 0.59 | a |
| 400L/100 | 3 | 0.3 | 0.1125 | 0.2363625 | 0.06 | 0.54 | a |
| 400L/50 | 4 | 0.25 | 0.1067 | 0.2241767 | 0.03 | 0.47 | a |
| 250L/50 | 5 | 0.1 | 0.0754 | 0.1584154 | -0.06 | 0.26 | a |
| Nufol 20 | 7 | 0.1 | 0.0754 | 0.1584154 | -0.06 | 0.26 | a |
| 400L/50 x2 | 9 | 0.25 | 0.1067 | 0.2241767 | 0.03 | 0.47 | a |

| | | Day 1 | Proportion with score H | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.25 | 0.11665 | 0.24508165 | 0.00 | 0.50 | a |
| 1000L/100 | 2 | 0.1 | 0.08139 | 0.17100039 | -0.07 | 0.27 | a |
| 400L/100 | 3 | 0.15 | 0.09659 | 0.20293559 | -0.05 | 0.35 | a |
| 400L/50 | 4 | 0.2 | 0.10795 | 0.22680295 | -0.03 | 0.43 | a |
| 250L/50 | 5 | 0.3 | 0.12329 | 0.25903229 | 0.04 | 0.56 | a |
| Nufol 20 | 7 | 0 | 0.00087 | 0.00182787 | 0.00 | 0.00 | a |
| 400L/50 x2 | 9 | 0.05 | 0.05934 | 0.12467334 | -0.07 | 0.17 | a |

| | | Day 14 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.6 | 0.04898 | 0.10290698 | 0.50 | 0.70 | a |
| 1000L/100 | 2 | 1 | 0.00007 | 0.00014707 | 1.00 | 1.00 | b |
| 400L/100 | 3 | 1 | 0.00007 | 0.00014707 | 1.00 | 1.00 | b |
| 400L/50 | 4 | 1 | 0.00007 | 0.00014707 | 1.00 | 1.00 | b |
| 250L/50 | 5 | 1 | 0.00007 | 0.00014707 | 1.00 | 1.00 | b |
| Nufol 20 | 7 | 0.7 | 0.04667 | 0.09805367 | 0.60 | 0.80 | a |
| 400L/50 x2 | 9 | 0.9 | 0.03208 | 0.06740008 | 0.83 | 0.97 | c |

| | | Day 14 | Proportion with score M | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.4 | 0.04898 | 0.10290698 | 0.30 | 0.50 | a |
| 1000L/100 | 2 | 0 | 0.00008 | 0.00016808 | 0.00 | 0.00 | b |
| 400L/100 | 3 | 0 | 0.00008 | 0.00016808 | 0.00 | 0.00 | b |
| 400L/50 | 4 | 0 | 0.00008 | 0.00016808 | 0.00 | 0.00 | b |
| 250L/50 | 5 | 0 | 0.00008 | 0.00016808 | 0.00 | 0.00 | b |
| Nufol 20 | 7 | 0.3 | 0.04667 | 0.09805367 | 0.20 | 0.40 | a |
| 400L/50 x2 | 9 | 0.1 | 0.03208 | 0.06740008 | 0.03 | 0.17 | c |

| | | Day 28 | Proportion with score No mycelium | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.3 | 0.1427 | 0.2998127 | 0.00 | 0.60 | a |
| 1000L/100 | 2 | 0.4 | 0.1519 | 0.3191419 | 0.08 | 0.72 | a |
| 400L/100 | 3 | 0.45 | 0.154 | 0.323554 | 0.13 | 0.77 | a |
| 400L/50 | 4 | 0.4 | 0.1519 | 0.3191419 | 0.08 | 0.72 | a |
| 250L/50 | 5 | 0.2 | 0.1253 | 0.2632553 | -0.06 | 0.46 | a |
| Nufol 20 | 7 | 0.25 | 0.1352 | 0.2840552 | -0.03 | 0.53 | a |
| 400L/50 x2 | 9 | 0.2 | 0.1253 | 0.2632553 | -0.06 | 0.46 | a |

| | | Day 28 | Proportion with score O | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.35 | 0.1533 | 0.3220833 | 0.03 | 0.67 | a |
| 1000L/100 | 2 | 0.4 | 0.1571 | 0.3300671 | 0.07 | 0.73 | a |
| 400L/100 | 3 | 0.45 | 0.1594 | 0.3348994 | 0.12 | 0.78 | a |
| 400L/50 | 4 | 0.25 | 0.1402 | 0.2945602 | -0.04 | 0.54 | a |
| 250L/50 | 5 | 0.4 | 0.1571 | 0.3300671 | 0.07 | 0.73 | a |
| Nufol 20 | 7 | 0.5 | 0.1601 | 0.3363701 | 0.16 | 0.84 | a |
| 400L/50 x2 | 9 | 0.45 | 0.1594 | 0.3348994 | 0.12 | 0.78 | a |

| | | Day 28 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.35 | 0.1594 | 0.3348994 | 0.02 | 0.68 | a |
| 1000L/100 | 2 | 0.2 | 0.1343 | 0.2821643 | -0.08 | 0.48 | a |
| 400L/100 | 3 | 0.1 | 0.1011 | 0.2124111 | -0.11 | 0.31 | a |
| 400L/50 | 4 | 0.3 | 0.1532 | 0.3218732 | -0.02 | 0.62 | a |
| 250L/50 | 5 | 0.4 | 0.1634 | 0.3433034 | 0.06 | 0.74 | a |
| Nufol 20 | 7 | 0.25 | 0.1449 | 0.3044349 | -0.05 | 0.55 | a |
| 400L/50 x2 | 9 | 0.3 | 0.153 | 0.321453 | -0.02 | 0.62 | a |

| | | Day 28 | Proportion with score M | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0 | 0.00000002 | 4.202E-08 | 0.00 | 0.00 | a |
| 1000L/100 | 2 | 0 | 0.00000002 | 4.202E-08 | 0.00 | 0.00 | a |
| 400L/100 | 3 | 0 | 0.00000002 | 4.202E-08 | 0.00 | 0.00 | a |
| 400L/50 | 4 | 0.05 | 0.00003927 | 8.25063E-05 | 0.05 | 0.05 | b |
| 250L/50 | 5 | 0 | 0.00000002 | 4.202E-08 | 0.00 | 0.00 | a |
| Nufol 20 | 7 | 0 | 0.00000002 | 4.202E-08 | 0.00 | 0.00 | a |
| 400L/50 x2 | 9 | 0.05 | 0.00003927 | 8.25063E-05 | 0.05 | 0.05 | b |

Assessments after Timing 2 application

| | | Day 1 | Proportion with score No mycelium | | | | |
|---|---|---|---|---|---|---|---|
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.1 | 0.0953 | 0.1966992 | -0.10 | 0.30 | a |
| 1000L/100 | 2 | 0.25 | 0.1361 | 0.2809104 | -0.03 | 0.53 | a |
| 400L/100 pre | 3 | 0.35 | 0.1495 | 0.308568 | 0.04 | 0.66 | a |
| 400L/50 | 4 | 0.15 | 0.1129 | 0.2330256 | -0.08 | 0.38 | a |
| 250L/50 pre | 5 | 0.35 | 0.1495 | 0.308568 | 0.04 | 0.66 | a |
| 250L/50 post | 6 | 0.3 | 0.1438 | 0.2968032 | 0.00 | 0.60 | a |
| Nufol 20 | 7 | 0.05 | 0.0696 | 0.1436544 | -0.09 | 0.19 | a |
| 400L/50 post | 8 | 0.1 | 0.0953 | 0.1966992 | -0.10 | 0.30 | a |
| 400L/50 x2 | 9 | 0.1 | 0.0953 | 0.1966992 | -0.10 | 0.30 | a |

| | | Day 1 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.9 | 0.0952 | 0.1964928 | 0.70 | 1.10 | a |
| 1000L/100 | 2 | 0.75 | 0.1361 | 0.2809104 | 0.47 | 1.03 | a |
| 400L/100 pre | 3 | 0.65 | 0.1495 | 0.308568 | 0.34 | 0.96 | a |
| 400L/50 | 4 | 0.85 | 0.1129 | 0.2330256 | 0.62 | 1.08 | a |
| 250L/50 pre | 5 | 0.65 | 0.1495 | 0.308568 | 0.34 | 0.96 | a |
| 250L/50 post | 6 | 0.7 | 0.1438 | 0.2968032 | 0.40 | 1.00 | a |
| Nufol 20 | 7 | 0.95 | 0.069 | 0.142416 | 0.81 | 1.09 | a |
| 400L/50 post | 8 | 0.9 | 0.0952 | 0.1964928 | 0.70 | 1.10 | a |
| 400L/50 x2 | 9 | 0.9 | 0.0952 | 0.1964928 | 0.70 | 1.10 | a |

| | | Day 14 | Proportion with score no mycelium | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.4 | 0.1281 | 0.2643984 | 0.14 | 0.66 | a |
| 1000L/100 | 2 | 0.15 | 0.0946 | 0.1952544 | -0.05 | 0.35 | a |
| 400L/100 pre | 3 | 0.35 | 0.125 | 0.258 | 0.09 | 0.61 | a |
| 400L/50 | 4 | 0.25 | 0.114 | 0.235296 | 0.01 | 0.49 | a |
| 250L/50 pre | 5 | 0.1 | 0.0798 | 0.1647072 | -0.06 | 0.26 | a |
| 250L/50 post | 6 | 0.4 | 0.1281 | 0.2643984 | 0.14 | 0.66 | a |
| Nufol 20 | 7 | 0.35 | 0.125 | 0.258 | 0.09 | 0.61 | a |
| 400L/50 post | 8 | 0.25 | 0.114 | 0.235296 | 0.01 | 0.49 | a |
| 400L/50 x2 | 9 | 0.15 | 0.0946 | 0.1952544 | -0.05 | 0.35 | a |

| | | Day 14 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.6 | 0.1281 | 0.2643984 | 0.34 | 0.86 | a |
| 1000L/100 | 2 | 0.85 | 0.0946 | 0.1952544 | 0.65 | 1.05 | a |
| 400L/100 pre | 3 | 0.65 | 0.125 | 0.258 | 0.39 | 0.91 | a |
| 400L/50 | 4 | 0.75 | 0.114 | 0.235296 | 0.51 | 0.99 | a |
| 250L/50 pre | 5 | 0.9 | 0.0797 | 0.1645008 | 0.74 | 1.06 | a |
| 250L/50 post | 6 | 0.6 | 0.1281 | 0.2643984 | 0.34 | 0.86 | a |
| Nufol 20 | 7 | 0.65 | 0.125 | 0.258 | 0.39 | 0.91 | a |
| 400L/50 post | 8 | 0.75 | 0.114 | 0.235296 | 0.51 | 0.99 | a |
| 400L/50 x2 | 9 | 0.85 | 0.0946 | 0.1952544 | 0.65 | 1.05 | a |

| | | Day 28 | Proportion with score no mycelium | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.45 | 0.2104 | 0.4342656 | 0.02 | 0.88 | a |
| 1000L/100 | 2 | 0.45 | 0.2104 | 0.4342656 | 0.02 | 0.88 | a |
| 400L/100 pre | 3 | 0.5 | 0.2115 | 0.436536 | 0.06 | 0.94 | a |
| 400L/50 | 4 | 0.15 | 0.1515 | 0.312696 | -0.16 | 0.46 | a |
| 250L/50 pre | 5 | 0.05 | 0.0925 | 0.19092 | -0.14 | 0.24 | a |
| 250L/50 post | 6 | 0.25 | 0.1834 | 0.3785376 | -0.13 | 0.63 | a |
| Nufol 20 | 7 | 0.3 | 0.194 | 0.400416 | -0.10 | 0.70 | a |
| 400L/50 post | 8 | 0.4 | 0.2073 | 0.4278672 | -0.03 | 0.83 | a |
| 400L/50 x2 | 9 | 0.3 | 0.194 | 0.400416 | -0.10 | 0.70 | a |

| | | Day 28 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.55 | 0.2104 | 0.4342656 | 0.12 | 0.98 | a |
| 1000L/100 | 2 | 0.55 | 0.2104 | 0.4342656 | 0.12 | 0.98 | a |
| 400L/100 pre | 3 | 0.5 | 0.2115 | 0.436536 | 0.06 | 0.94 | a |
| 400L/50 | 4 | 0.85 | 0.1515 | 0.312696 | 0.54 | 1.16 | a |
| 250L/50 pre | 5 | 0.95 | 0.0916 | 0.1890624 | 0.76 | 1.14 | a |
| 250L/50 post | 6 | 0.75 | 0.1834 | 0.3785376 | 0.37 | 1.13 | a |
| Nufol 20 | 7 | 0.7 | 0.194 | 0.400416 | 0.30 | 1.10 | a |
| 400L/50 post | 8 | 0.6 | 0.2073 | 0.4278672 | 0.17 | 1.03 | a |
| 400L/50 x2 | 9 | 0.7 | 0.194 | 0.400416 | 0.30 | 1.10 | a |

<u>Adjuvants</u>

| | | Day 1 | Proportion with score 0 | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| Untreated | 1 | 0 | 0.00104 | 0.00211744 | 0.00 | 0.00 | a |
| Urea + Silwett-L77 (standard) | 2 | 0.75 | 0.13577 | 0.27642772 | 0.47 | 1.03 | b |
| Urea + Tween 20 | 3 | 0 | 0.00104 | 0.00211744 | 0.00 | 0.00 | a |
| Urea + Activator 90 | 4 | 0.3 | 0.14245 | 0.2900282 | 0.01 | 0.59 | b |
| Urea + X-change | 5 | 0.3 | 0.14245 | 0.2900282 | 0.01 | 0.59 | b |
| Urea + Grounded | 6 | 0.1 | 0.0949 | 0.1932164 | -0.09 | 0.29 | a |
| Urea + Bond | 7 | 0.1 | 0.0949 | 0.1932164 | -0.09 | 0.29 | a |
| Urea + Toil | 8 | 0.5 | 0.15493 | 0.31543748 | 0.18 | 0.82 | b |
| Ammonium nitrate | 9 | 0.1 | 0.0949 | 0.1932164 | -0.09 | 0.29 | a |
| Nuram 37 | 10 | 0.05 | 0.0694 | 0.1412984 | -0.09 | 0.19 | a |
| Nufol 20 | 11 | 0.3 | 0.14245 | 0.2900282 | 0.01 | 0.59 | b |
| Ammonium sulphate | 12 | 0.1 | 0.0949 | 0.1932164 | -0.09 | 0.29 | a |

| | | Day 1 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| Untreated | 1 | 0.15 | 0.09883 | 0.125 | 0.03 | 0.28 | a |
| Urea + Silwett-L77 (standard) | 2 | 0.25 | 0.00081 | 0.151 | 0.10 | 0.40 | a |
| Urea + Tween 20 | 3 | 0.55 | 0.13683 | 0.1723 | 0.38 | 0.72 | b |
| Urea + Activator 90 | 4 | 0.6 | 0.08319 | 0.1696 | 0.43 | 0.77 | b |
| Urea + X-change | 5 | 0.65 | 0.06057 | 0.1652 | 0.48 | 0.82 | b |
| Urea + Grounded | 6 | 0.4 | 0.1375 | 0.17 | 0.23 | 0.57 | a |
| Urea + Bond | 7 | 0.8 | 0.08319 | 0.1388 | 0.66 | 0.94 | b |
| Urea + Toil | 8 | 0.4 | 0.08319 | 0.17 | 0.23 | 0.57 | a |
| Ammonium nitrate | 9 | 0.3 | 0.13479 | 0.1595 | 0.14 | 0.46 | a |
| Nuram 37 | 10 | 0.55 | 0.1348 | 0.1723 | 0.38 | 0.72 | b |
| Nufol 20 | 11 | 0.7 | 0.00081 | 0.1588 | 0.54 | 0.86 | b |
| Ammonium sulphate | 12 | 0.65 | 0.11948 | 0.1652 | 0.48 | 0.82 | b |

| | | Day 1 | Proportion with score M | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| Untreated | 1 | 0.5 | 0.13212 | 0.26899632 | 0.23 | 0.77 | a |
| Urea + Silwett-L77 (standard) | 2 | 0 | 0.00084 | 0.00171024 | 0.00 | 0.00 | b |
| Urea + Tween 20 | 3 | 0.25 | 0.11456 | 0.23324416 | 0.02 | 0.48 | a |
| Urea + Activator 90 | 4 | 0.05 | 0.05781 | 0.11770116 | -0.07 | 0.17 | b |
| Urea + X-change | 5 | 0.05 | 0.05781 | 0.11770116 | -0.07 | 0.17 | b |
| Urea + Grounded | 6 | 0.45 | 0.13147 | 0.26767292 | 0.18 | 0.72 | a |
| Urea + Bond | 7 | 0.1 | 0.07952 | 0.16190272 | -0.06 | 0.26 | ab |
| Urea + Toil | 8 | 0.1 | 0.07952 | 0.16190272 | -0.06 | 0.26 | ab |
| Ammonium nitrate | 9 | 0.55 | 0.13147 | 0.26767292 | 0.28 | 0.82 | a |
| Nuram 37 | 10 | 0.3 | 0.12119 | 0.24674284 | 0.05 | 0.55 | ab |
| Nufol 20 | 11 | 0 | 0.00084 | 0.00171024 | 0.00 | 0.00 | b |
| Ammonium sulphate | 12 | 0.15 | 0.09458 | 0.19256488 | -0.04 | 0.34 | ab |

 

| | | Day 1 | Proportion with score H | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| Untreated | 1 | 0.35 | 0.09323 | 0.18981628 | 0.16 | 0.54 | a |
| Urea + Silwett-L77 (standard) | 2 | 0 | 0.00023 | 0.00046828 | 0.00 | 0.00 | b |
| Urea + Tween 20 | 3 | 0.2 | 0.07847 | 0.15976492 | 0.04 | 0.36 | a |
| Urea + Activator 90 | 4 | 0.05 | 0.043 | 0.087548 | -0.04 | 0.14 | b |
| Urea + X-change | 5 | 0 | 0.00023 | 0.00046828 | 0.00 | 0.00 | b |
| Urea + Grounded | 6 | 0.05 | 0.043 | 0.087548 | -0.04 | 0.14 | a |
| Urea + Bond | 7 | 0 | 0.00023 | 0.00046828 | 0.00 | 0.00 | b |
| Urea + Toil | 8 | 0 | 0.00023 | 0.00046828 | 0.00 | 0.00 | b |
| Ammonium nitrate | 9 | 0.05 | 0.043 | 0.087548 | -0.04 | 0.14 | b |
| Nuram 37 | 10 | 0.1 | 0.05906 | 0.12024616 | -0.02 | 0.22 | a |
| Nufol 20 | 11 | 0 | 0.00023 | 0.00046828 | 0.00 | 0.00 | b |
| Ammonium sulphate | 12 | 0.1 | 0.05906 | 0.12024616 | -0.02 | 0.22 | a |

| | | Day 14 | Proportion with score No mycelium | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| Untreated | 1 | 0 | 0.0008 | 0.0016288 | 0.00 | 0.00 | a |
| Urea + Silwett-L77 (standard) | 2 | 0.99 | 0.0019 | 0.0038684 | 0.99 | 0.99 | b |
| Urea + Tween 20 | 3 | 0 | 0.0008 | 0.0016288 | 0.00 | 0.00 | a |
| Urea + Activator 90 | 4 | 0.65 | 0.1738 | 0.3538568 | 0.30 | 1.00 | bc |
| Urea + X-change | 5 | 0.75 | 0.1602 | 0.3261672 | 0.42 | 1.08 | bc |
| Urea + Grounded | 6 | 0.65 | 0.1738 | 0.3538568 | 0.30 | 1.00 | bc |
| Urea + Bond | 7 | 0.85 | 0.1343 | 0.2734348 | 0.58 | 1.12 | bc |
| Urea + Toil | 8 | 0.8 | 0.1492 | 0.3037712 | 0.50 | 1.10 | bc |
| Ammonium nitrate | 9 | 0.55 | 0.1791 | 0.3646476 | 0.19 | 0.91 | c |
| Nuram 37 | 10 | 0 | 0.0008 | 0.0016288 | 0.00 | 0.00 | a |
| Nufol 20 | 11 | 0.6 | 0.1774 | 0.3611864 | 0.24 | 0.96 | bc |
| Ammonium sulphate | 12 | 0.75 | 0.1602 | 0.3261672 | 0.42 | 1.08 | bc |

| | | Day 14 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| Untreated | 1 | 0.4 | 0.1948 | 0.3966128 | 0.0034 | 0.7966 | a |
| Urea + Silwett-L77 (standard) | 2 | 0.0001 | 0.0023 | 0.0046828 | -0.0046 | 0.0048 | a |
| Urea + Tween 20 | 3 | 0.1 | 0.1231 | 0.2506316 | -0.1506 | 0.3506 | a |
| Urea + Activator 90 | 4 | 0.25 | 0.1746 | 0.3554856 | -0.1055 | 0.6055 | a |
| Urea + X-change | 5 | 0.25 | 0.1746 | 0.3554856 | -0.1055 | 0.6055 | a |
| Urea + Grounded | 6 | 0.35 | 0.1904 | 0.3876544 | -0.0377 | 0.7377 | a |
| Urea + Bond | 7 | 0.15 | 0.1456 | 0.2964416 | -0.1464 | 0.4464 | a |
| Urea + Toil | 8 | 0.2 | 0.1622 | 0.3302392 | -0.1302 | 0.5302 | a |
| Ammonium nitrate | 9 | 0.35 | 0.1904 | 0.3876544 | -0.0377 | 0.7377 | a |
| Nuram 37 | 10 | 0.6 | 0.1929 | 0.3927444 | 0.2073 | 0.9927 | a |
| Nufol 20 | 11 | 0.4 | 0.1948 | 0.3966128 | 0.0034 | 0.7966 | a |
| Ammonium sulphate | 12 | 0.25 | 0.1746 | 0.3554856 | -0.1055 | 0.6055 | a |

| | | Day 14 | Proportion with score M | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| Untreated | 1 | 0.25 | 0.07312 | 0.14887232 | 0.10 | 0.40 | a |
| Urea + Silwett-L77 (standard) | 2 | 0 | 0.00021 | 0.00042756 | 0.00 | 0.00 | b |
| Urea + Tween 20 | 3 | 0.55 | 0.08255 | 0.1680718 | 0.38 | 0.72 | a |
| Urea + Activator 90 | 4 | 0.05 | 0.03761 | 0.07657396 | -0.03 | 0.13 | ab |
| Urea + X-change | 5 | 0 | 0.00021 | 0.00042756 | 0.00 | 0.00 | b |
| Urea + Grounded | 6 | 0 | 0.00021 | 0.00042756 | 0.00 | 0.00 | b |
| Urea + Bond | 7 | 0 | 0.00021 | 0.00042756 | 0.00 | 0.00 | b |
| Urea + Toil | 8 | 0 | 0.00021 | 0.00042756 | 0.00 | 0.00 | b |
| Ammonium nitrate | 9 | 0.1 | 0.05147 | 0.10479292 | 0.00 | 0.20 | a |
| Nuram 37 | 10 | 0.3 | 0.07705 | 0.1568738 | 0.14 | 0.46 | a |
| Nufol 20 | 11 | 0 | 0.00021 | 0.00042756 | 0.00 | 0.00 | b |
| Ammonium sulphate | 12 | 0 | 0.00021 | 0.00042756 | 0.00 | 0.00 | b |

| | | Day 14 | Proportion with score H | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| Untreated | 1 | 0.35 | 0.09252 | 0.18837072 | 0.16 | 0.54 | a |
| Urea + Silwett-L77 (standard) | 2 | 0 | 0.00024 | 0.00048864 | 0.00 | 0.00 | b |
| Urea + Tween 20 | 3 | 0.35 | 0.09252 | 0.18837072 | 0.16 | 0.54 | a |
| Urea + Activator 90 | 4 | 0.05 | 0.04402 | 0.08962472 | -0.04 | 0.14 | b |
| Urea + X-change | 5 | 0 | 0.00024 | 0.00048864 | 0.00 | 0.00 | b |
| Urea + Grounded | 6 | 0 | 0.00024 | 0.00048864 | 0.00 | 0.00 | b |
| Urea + Bond | 7 | 0 | 0.00024 | 0.00048864 | 0.00 | 0.00 | b |
| Urea + Toil | 8 | 0 | 0.00024 | 0.00048864 | 0.00 | 0.00 | b |
| Ammonium nitrate | 9 | 0 | 0.00024 | 0.00048864 | 0.00 | 0.00 | b |
| Nuram 37 | 10 | 0.1 | 0.06003 | 0.12222108 | -0.02 | 0.22 | a |
| Nufol 20 | 11 | 0 | 0.00024 | 0.00048864 | 0.00 | 0.00 | b |
| Ammonium sulphate | 12 | 0 | 0.00024 | 0.00048864 | 0.00 | 0.00 | b |

Water volumes

| | | Day 1 | Proportion with score 0 | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0 | 0.00000002 | 4.262E-08 | 0.00 | 0.00 | a |
| 1000L | 2 | 0 | 0.00000002 | 4.262E-08 | 0.00 | 0.00 | a |
| 800L | 3 | 0.05 | 0.00004005 | 8.53466E-05 | 0.05 | 0.05 | b |
| 600L | 4 | 0 | 0.00000002 | 4.262E-08 | 0.00 | 0.00 | a |
| 400L | 5 | 0 | 0.00000002 | 4.262E-08 | 0.00 | 0.00 | a |
| 200L | 6 | 0.1 | 0.00004906 | 0.000104547 | 0.10 | 0.10 | c |

| | | Day 1 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.2 | 0.1302 | 0.2774562 | -0.08 | 0.48 | a |
| 1000L | 2 | 0.25 | 0.1398 | 0.2979138 | -0.05 | 0.55 | a |
| 800L | 3 | 0.6 | 0.155 | 0.330305 | 0.27 | 0.93 | a |
| 600L | 4 | 0.55 | 0.157 | 0.334567 | 0.22 | 0.88 | a |
| 400L | 5 | 0.4 | 0.1553 | 0.3309443 | 0.07 | 0.73 | a |
| 200L | 6 | 0.7 | 0.1464 | 0.3119784 | 0.39 | 1.01 | a |

| | | Day 1 | Proportion with score M | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.35 | 0.1437 | 0.3062247 | 0.04 | 0.66 | a |
| 1000L | 2 | 0.6 | 0.1475 | 0.3143225 | 0.29 | 0.91 | a |
| 800L | 3 | 0.2 | 0.121 | 0.257851 | -0.06 | 0.46 | a |
| 600L | 4 | 0.45 | 0.15 | 0.31965 | 0.13 | 0.77 | a |
| 400L | 5 | 0.4 | 0.1478 | 0.3149618 | 0.09 | 0.71 | a |
| 200L | 6 | 0.15 | 0.108 | 0.230148 | -0.08 | 0.38 | a |

| | | Day 1 | Proportion with score H | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.45 | 0.09726 | 0.20726106 | 0.24 | 0.66 | a |
| 1000L | 2 | 0.15 | 0.07484 | 0.15948404 | -0.01 | 0.31 | a |
| 800L | 3 | 0.15 | 0.07484 | 0.15948404 | -0.01 | 0.31 | a |
| 600L | 4 | 0 | 0.00045 | 0.00095895 | 0.00 | 0.00 | b |
| 400L | 5 | 0.2 | 0.08285 | 0.17655335 | 0.02 | 0.38 | a |
| 200L | 6 | 0.05 | 0.04674 | 0.09960294 | -0.05 | 0.15 | a |

| | | Day 14 | Proportion with score 0 | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0 | 0.0012 | 0.0025572 | 0.00 | 0.00 | a |
| 1000L | 2 | 0.85 | 0.1381 | 0.2942911 | 0.56 | 1.14 | b |
| 800L | 3 | 0.35 | 0.1843 | 0.3927433 | -0.04 | 0.74 | b |
| 600L | 4 | 0.25 | 0.1674 | 0.3567294 | -0.11 | 0.61 | b |
| 400L | 5 | 0.15 | 0.1381 | 0.2942911 | -0.14 | 0.44 | b |
| 200L | 6 | 0.4 | 0.1892 | 0.4031852 | 0.00 | 0.80 | b |

| | | Day 14 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.75 | 0.09077 | 0.19343087 | 0.56 | 0.94 | a |
| 1000L | 2 | 0.15 | 0.07572 | 0.16135932 | -0.01 | 0.31 | b |
| 800L | 3 | 0.4 | 0.10175 | 0.21682925 | 0.18 | 0.62 | a |
| 600L | 4 | 0.6 | 0.10173 | 0.21678663 | 0.38 | 0.82 | a |
| 400L | 5 | 0.45 | 0.10317 | 0.21985527 | 0.23 | 0.67 | a |
| 200L | 6 | 0.35 | 0.09931 | 0.21162961 | 0.14 | 0.56 | b |

 

| | | Day 14 | Proportion with score M | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.2 | 0.10396 | 0.22153876 | -0.02 | 0.42 | a |
| 1000L | 2 | 0 | 0.00092 | 0.00196052 | 0.00 | 0.00 | a |
| 800L | 3 | 0.2 | 0.10396 | 0.22153876 | -0.02 | 0.42 | a |
| 600L | 4 | 0.15 | 0.09362 | 0.19950422 | -0.05 | 0.35 | a |
| 400L | 5 | 0.3 | 0.11695 | 0.24922045 | 0.05 | 0.55 | a |
| 200L | 6 | 0.15 | 0.09362 | 0.19950422 | -0.05 | 0.35 | a |

| | | Day 14 | Proportion with score H | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.05 | 0.03293 | 0.07017383 | -0.02 | 0.12 | a |
| 1000L | 2 | 0 | 0.00012 | 0.00025572 | 0.00 | 0.00 | a |
| 800L | 3 | 0.05 | 0.03293 | 0.07017383 | -0.02 | 0.12 | a |
| 600L | 4 | 0 | 0.00012 | 0.00025572 | 0.00 | 0.00 | a |
| 400L | 5 | 0.1 | 0.0446 | 0.0950426 | 0.00 | 0.20 | a |
| 200L | 6 | 0.1 | 0.0446 | 0.0950426 | 0.00 | 0.20 | a |

| | | Day 28 | Proportion with score 0 | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.1875 | 0.1753 | 0.3735643 | -0.19 | 0.56 | a |
| 1000L | 2 | 0.2545 | 0.1699 | 0.3620569 | -0.11 | 0.62 | a |
| 800L | 3 | 0.2164 | 0.1675 | 0.3569425 | -0.14 | 0.57 | a |
| 600L | 4 | 0.4547 | 0.1911 | 0.4072341 | 0.05 | 0.86 | a |
| 400L | 5 | 0.0535 | 0.0938 | 0.1998878 | -0.15 | 0.25 | a |
| 200L | 6 | 0.5044 | 0.1919 | 0.4089389 | 0.10 | 0.91 | a |

| | | Day 28 | Proportion with score L | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.7195 | 0.1957 | 0.4170367 | 0.30 | 1.14 | a |
| 1000L | 2 | 0.6017 | 0.1778 | 0.3788918 | 0.22 | 0.98 | a |
| 800L | 3 | 0.6156 | 0.1859 | 0.3961529 | 0.22 | 1.01 | a |
| 600L | 4 | 0.4018 | 0.1783 | 0.3799573 | 0.02 | 0.78 | a |
| 400L | 5 | 0.5826 | 0.1839 | 0.3918909 | 0.19 | 0.97 | a |
| 200L | 6 | 0.4018 | 0.1783 | 0.3799573 | 0.02 | 0.78 | a |

| | | Day 28 | Proportion with score M | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.072 | 0.0959 | 0.2043629 | -0.13 | 0.28 | a |
| 1000L | 2 | 0.1475 | 0.1071 | 0.2282301 | -0.08 | 0.38 | a |
| 800L | 3 | 0.1699 | 0.1211 | 0.2580641 | -0.09 | 0.43 | a |
| 600L | 4 | 0.1475 | 0.1071 | 0.2282301 | -0.08 | 0.38 | a |
| 400L | 5 | 0.2586 | 0.1351 | 0.2878981 | -0.03 | 0.55 | a |
| 200L | 6 | 0.0983 | 0.0902 | 0.1922162 | -0.09 | 0.29 | a |

| | | Day 28 | Proportion with score H | | | | |
|---|---|---|---|---|---|---|---|
| | | | | | | | |
| | Treatment | Prediction | S.E. | Confidence Interval, S.E. x t | LCL (Pred - CI) | UCL (Pred + CI) | |
| UT | 1 | 0.01609 | 0.014689 | 0.031302259 | -0.02 | 0.05 | a |
| 1000L | 2 | 0 | 0 | 0 | 0.00 | 0.00 | b |
| 800L | 3 | 0 | 0 | 0 | 0.00 | 0.00 | b |
| 600L | 4 | 0 | 0 | 0 | 0.00 | 0.00 | b |
| 400L | 5 | 0.08571 | 0.000043 | 0.000091633 | 0.09 | 0.09 | c |
| 200L | 6 | 0 | 0 | 0 | 0.00 | 0.00 | b |

 