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

AUTHENTICATION

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

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GROWER SUMMARY

Headline

- Daffodil rust, a physiological disorder with no known cause, may be associated with high soil water content in the months before flowering.

Background

Starting in the early-1990s UK daffodil growers became concerned about rust-like lesions appearing on daffodil stems. The symptoms occurred sporadically, sometimes presenting as a few insignificant spots, but in more serious cases the lesions were numerous, forming coalescing areas of damage that disfigured stems and caused the product to be downgraded. In severe cases stems showed transverse cracking and became brittle and unmarketable. Despite the convincingly disease-like lesions, initial testing apparently failed to reveal a primary pathogen associated with the lesions, so the condition was classed as a physiological disorder. It soon came to be called 'physiological rust', or more recently 'stem rust' (though similar lesions may occur on leaves). In this report the disorder is called 'daffodil rust'.

To gauge the extent and economic cost of daffodil rust, the HDC organised surveys of growers in 2002 and 2003 and again during the period 2011 to 2013. The responses confirmed that daffodil rust had caused - and continues to cause - commercially significant losses that would justify research into its cause and management. The responses highlighted that growers and advisers had submitted affected (a) plant samples for diagnostic examination and (b) soil and plant samples from affected and 'healthy' crops for mineral analysis. No pathogenic or nutritional cause was found, though neither should be ruled out entirely because sampling had been opportunistic and non-replicated, whereas more structured sampling could have provided more robust conclusions. The surveys also revealed ideas circulating in the industry about other causes of daffodil rust: suspected predisposing conditions often involved rapid changes in temperature, alternating cold and warm periods, and adverse weather.

Daffodil chocolate spot is another physiological disorder of daffodils and has parallels with daffodil rust, though it has not been known to have had any serious effects. As with daffodil rust, pathological and nutritional causes were ruled out, while an association with increasing ambient temperatures was suggested. Several physiological disorders of other horticultural crops are characterised by the appearance of brown or black spotting and have been linked with adverse environmental conditions. Apple leaf spot and drop, for example, has been associated with dry, hot summer weather and a sudden change in temperature, while lettuce dry (or marginal) tip-burn has been linked to water stress when transpiration exceeds water uptake, promoted by sudden checks in growth such as low temperatures.

In the light of this evidence daffodil rust could be a physiological disorder brought about by adverse environmental conditions. The objective of project BOF 76 is to test the proposition that the soil-water environment (soil structure, water availability, soil temperature, nutritional status, etc.) affects the level of daffodil rust. One practical way of assessing the effects of the soil-water environment on the disorder would be to monitor rust development and environmental factors and search for associations between them. Therefore plots of the rust-susceptible daffodil cultivar 'Golden Ducat' were planted in ten daffodil fields at varied locations through west Cornwall, the region where it seemed crops were most prone to the disorder. This scheme should maximise the likelihood that daffodil rust would occur naturally in at least some of the experimental locations, potentially enabling the predisposing factors to be identified through association and regression analysis.

Since the project would supply a structured set of affected and 'healthy' samples, the opportunity will be used (in 2014) to examine further the pathological and nutritional theories of daffodil rust. Samples of plants will be taken for plant clinic examination for fungal or bacterial pathogens, for sequencing viral RNA, and for the analysis of macro- and micro-nutrients. Soil samples will be taken for the analysis of macro- and micro-nutrients.

Summary

Surveys of growers

In 2002 68% of respondents reported having seen daffodil rust on their crops during the previous five years or so, and this figure rose to 86% by 2011, this being cited as the worst 'rust year' so far. In 2002, 36% of respondents had seen some product downgraded and 25% could not supply their preferred customer or had product that was completely unmarketable; in 2011 the corresponding figures had increased to 71 and 57%. For both survey periods the loss of turnover due to daffodil rust was estimated at between 0 and 3% in a good year but up to 15% in a bad year.

Many cultivars were reported as being subject to daffodil rust, though 'Golden Ducat' and 'Mando' were cited a disproportionate number of times. 'Carlton', 'Kerensa' and 'Tamara' were also likely to be affected. Plants showed symptoms of daffodil rust in their first-, second-, third- and subsequent crop-years. Predisposing factors for daffodil rust cited by growers included repeated frost and thaw, alternating mild and cold weather, rapid snow melt with subsequent fast growth, and the end of cold or adverse weather. Where samples had been sent for disease diagnosis or nutrient analysis, no pathogen identification had

been made, nor any associations identified between nutrient levels and the level of daffodil rust.

Field-work

In autumn 2012, 50 kg-plots of bulbs of the rust-prone cultivar ‘Golden Ducat’ were planted in bulb fields at ten sites across west Cornwall, the locations being chosen to represent different situations, topographies, soil types, etc., increasing the likelihood that daffodil rust would occur on at least some of the sites. The locations were (from west to east) Kelynack, St Buryan, Tregiffian, Rosevidney, Roseworthy, Bodilly, Mawla, Penventon, Fourburrow and Goonhavern. At each site a weather station was set up, logging soil water content (SWC) at three depths (0-100, 100-200 and 200-300 mm), soil temperature (at 150 mm depth), air temperature, relative humidity (RH) and precipitation. This would enable the level of daffodil rust to be studied in relation to the soil-water and other features of each site, to identify associations between these factors and the level - of daffodil rust.

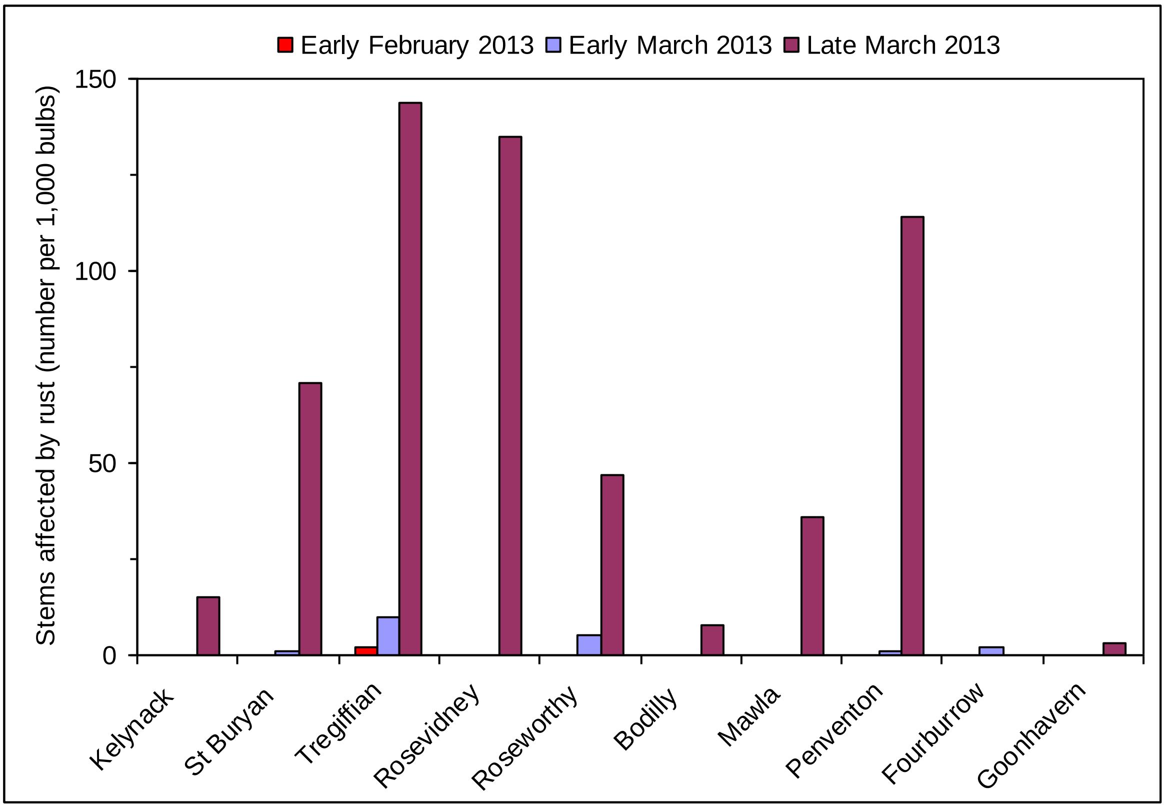
The main assessments of crop development and daffodil rust were carried out about two weeks before, around, and two weeks after flowering (flowers were not picked so that their development could be followed). In spring 2013, at the first assessment (early-February), no characteristic daffodil rust lesions were seen on stems or leaves, with the exception of the site at Tregiffian where two small, rust-like spots were seen on each of two stems. At Penventon inconspicuous depressions and pitting of stems were noted, subsequent observations suggesting these were a widely occurring early-stage of daffodil rust lesions.

At the second assessment (early-March, close to flower-picking at most sites) only infrequent, small and isolated daffodil rust lesions were found, and at only five sites (St Buryan, Tregiffian, Roseworthy, Penventon and Fourburrow). At Tregiffian ten stems bore either a single lesion or several spots. In addition, many stems bore the fainter marks seen earlier at Penventon, with lengthways pitting or blistering, pale or yellowing spots or longitudinal tracks. No characteristic daffodil rust lesions were found on the foliage, though the leaves at several sites had occasional brown, rusty streaks, the occurrence of which did not always correspond with that of the stem lesions.

At the third assessment (late-March) larger numbers of daffodil rust lesions were seen and all were small and typical. Daffodil rust severity varied from one or two small spots or occasionally streaks per stem at Goonhavern, to individual small spots and groups of up to about 15 small spots at Tregiffian. Similarly rust incidence varied from <10 affected stems/plot at Goonhavern to more than 50% affected at Tregiffian. The presumed early lesions were seen at all sites. Rust-like spots or streaks- were also found on leaves at all sites, usually with low severity and low incidence. At Kelynack, St Buryan and Tregiffian the severity of leaf lesions was greater, with more extensive marking and greater incidence (more than 50% of leaves affected).

The progress of daffodil rust development on stems over the three assessment dates is shown in the figure below. Tregiffian had both the earliest appearance of daffodil rust and its highest final incidence, and incidence was also high at St Buryan, Rosevidney and Penventon. Levels were lower at Roseworthy and Mawla, while the plots at Kelynack, Bodilly, Fourburrow and Goonhavern remained virtually free of the disorder.

Different severity of rust symptoms. Top: increasing rust severity with blistering (left), a few rust lesions (middle) and larger, coalescing lesions (right). Bottom: close-up of blistering (left) and rust lesions with cracking (right).



Incidence of daffodil rust at ten sites assessed on three dates in winter-spring 2013.

Associations between daffodil rust and soil-water and other factors

The levels of daffodil rust seen in this first crop-year were low in both incidence and severity, and would not have constituted a 'rust problem' in a commercial sense. Nevertheless, daffodil rust levels at the ten sites varied considerably, allowing them to be assessed in respect of the various differences between sites. The flowering stage was over at all sites by the end of March 2013 and, by implication for a picked crop, the maximum expression of daffodil rust on stems would have been reached by that time. SWC and weather data had been logged from 1 November 2012, providing five months' logged soil and weather data to explore for associations with daffodil rust levels. The plots showed that the values for soil and air temperatures, RH and precipitation were rather uniform across the ten sites, making it unlikely that these factors could be associated with the varying levels of daffodil rust across sites. However, there were substantial differences between sites in SWC measured at individual depths in the soil (0-100, 100-200 and 200-300 mm) or averaged or totalled across all depths (0-300 mm). Comparisons of these data with rust levels showed that three of the four sites with the highest incidence of daffodil rust were associated with the highest SWC – St Buryan, Tregiffian and Rosevidney (the exception being Penventon). The effect was most clearly seen by plotting SWC averaged over the three depths, perhaps because this draws on a larger number of individual data-points. If this association can be confirmed, the rust-promoting effect of high SWC could be a short-term effect (say, high SWC affecting stem growth at the time of rapid shoot growth immediately preceding flowering) or a longer-term one (perhaps an accumulating effect of high SWC over the previous months, or at some earlier key-stage of development). To investigate these possibilities, SWC was examined across shorter periods (months and weeks). The results showed that the key-period of the putative SWC effect was November to December, where the effect was significant, rather than January to March.

Potential associations between the levels of daffodil rust and other factors were sought through regression analysis. Examining geographical data- failed to show any associations with rust levels. Soil structural factors – a Visual Soil Structure Quality Assessment -, ADAS soil texture assessment, soil depth and the proportions of clay, silt, sand and stone particles - likewise failed to show any association with the levels of daffodil rust.

Some husbandry factors were also assessed. As a result of continuing wet weather in autumn 2012 when the plots were planted, planting was delayed at three sites, which might have affected plant growth. However, there was no association between planting date and rust levels. Before bulb planting, fertilisers were applied at most sites, generally P and K, but also N at Roseworthy and organic fertiliser at Penventon. At seven sites brassicas (which leave high N residues) were the previous crop. There was no evidence that high or low levels of daffodil rust were associated with the type of fertiliser applied or with the previous cropping of brassicas. Correlation analysis confirmed there was no evidence for associations between levels of daffodil rust and the concentrations of soil N, P, K or Mg or soil pH in pre-planting, autumn and spring analyses. The same was true for micro-nutrient concentrations (analysed in spring 2013 only), except for a weak effect of aluminium concentration whereby higher rust levels occurred at lower aluminium concentrations. To investigate the possibility that daffodil rust levels might be associated with the rate of plant development and growth in the run-up to flowering, rust levels were examined in relation to shoot/leaf and stem length in early-February and their increase in length during February. The only significant association, was between rust levels and shoot/leaf growth, with higher rust levels where growth was greater.

There is a trend in these data to support a positive association between high SWC in the months before flowering with relatively higher levels of daffodil rust at flowering. However, this held at only three of the four high-rust sites, so there may be an interaction with another factor or factors. The non-conforming site, Penventon (low SWC but high rust level), is notable as being on a steep and freely-draining south-facing slope. These results cover the first year of the project only, with low levels of daffodil rust, and so should be treated with caution. The work is continuing, and the indications from the second year of the crop are that higher rust levels are developing across most sites. These rust levels will be examined with the benefit of a longer run of weather data and further soil and plant nutrient analysis.

Financial Benefits

On the basis of information provided by growers, daffodil rust results in a 3% average annual loss of revenue from cut-flowers (spread across all years), or losses of 10% in one year in three (with negligible losses in the intervening years). A 3% annual loss is estimated to amount to about £0.7 m annually to UK growers, or around £2.3 m every three years. These are direct monetary losses resulting from daffodil rust, from reduced flower yields and (or) downgraded product, and in reality there would be additional cost savings associated with eliminating unpredictable yields and poor quality due to rust. These losses could be largely eliminated if the project succeeds in understanding the causes of daffodil rust and leads to the provision of solutions, avoidance or risk management. Perhaps more importantly, solving the rust problem would remove the likelihood of a gross loss of markets through lowered customer perception of the product.

Action Points

- Forward planning should take account of avoiding planting rust-prone varieties in poorly draining sites.
- At this early stage in the project no other action points can be suggested.

SCIENCE SECTION

Introduction

Daffodil rust – a problem in contemporary cut-flower production

The physiological disorder called daffodil rust degrades stem quality, making some cut-flowers unmarketable. With commercial daffodil growing in the UK more dependent on the sales of cut-flowers than of bulbs, it is vital to avoid anything that could harm customers' perception of product quality. We do not know the cause of daffodil rust.

From the early-1990s the minutes of the HDC Bulbs and Outdoor Flowers (BOF) Panel recorded the concerns of bulb growers over rust-like lesions being found on daffodil stems. The severity of the symptoms varied from a few insignificant spots, through larger groups of mildly disfiguring lesions, to prominent rusty lesions along much of an increasingly brittle stem. Similar lesions were seen on leaves, though attracting less attention because it is the cut-flowers that are the marketed commodity. References to the problem were initially sporadic, but soon increased, with some reports of batches that were so severely affected as to be unmarketable. The condition soon became known as 'physiological rust' and was regarded as a physiological disorder (non-parasitic disorder or abiotic disease) since 'rust' did not appear to be associated with a pathogen. More recently the term 'stem rust' has been adopted, though this ignores the foliar component. For convenience, for the rest of this report the disorder will be referred to simply as 'daffodil rust', except where that term could make its meaning ambiguous.

In anecdotal information daffodil rust was reported on many cultivars in both Cornwall and eastern England. Daffodil rust was observed on crops in their second and subsequent years of growth, but there were differences of opinion as to the occurrence of daffodil rust on first-year crops, though this may have been simply due to a lack of observation of the first-year crops from which flowers are not usually picked. Daffodil rust does not appear to have been reported in daffodils forced in glasshouses. Perhaps the most predictable feature of rust is its unpredictability. As would be expected, growers and others developed their own theories about the conditions that led to the appearance of daffodil rust lesions, such as the "crop grows too fast after a cold frosty spell", or that it was seen mainly in waterlogged areas.

In 2002, prior to considering an experimental approach, the HDC BOF Panel instigated a survey of growers to establish the economic importance of daffodil rust, to seek possible relationships between cultural practices and the prevalence of daffodil rust, and to collect the industry's theories about the its predisposing factors. The survey was repeated in 2003. In all 62 responses were received, and they confirmed the view that daffodil rust was leading to a substantial reduction in quality and output. The survey was repeated in 2011, 2012 and 2013 and, although the response rate was much lower, the daffodil acreage represented by the respondents was nevertheless substantial and the replies confirmed there had been no diminution of the negative economic effect of daffodil rust on cut-flower production. The data from all five years' questionnaires were summarised as part of the present project and are given below under 'Surveys of growers'.

Daffodil rust appears to have been the subject of little, if any, research, other than sporadic individual attempts at disease diagnosis or correlation with soil or plant nutrient levels (anecdotal information and personal communications). These have uncovered no causal organisms or links to high or low levels of macro- or micro-nutrients (some examples are given below under 'Surveys of growers'). The lack of a causal pathogen or nutrient deficiency for daffodil rust should dissuade growers from applying unnecessary fungicides or additional fertilisers in an attempt to prevent or cure the disorder. Daffodil rust does not appear to have been described in the key advisory literature or research reviews in either the UK (such as or Rees (1992)), the Netherlands (e.g. Langeslag (1990) or van Aartrijk *et al*., 1995)) or the USA (e.g. Gould & Byther (1979), Chastagner & Byther (1985) or De Hertogh & le Nard (1993)). Literature searches were conducted by the author, using the CABI 'Horticultural Abstracts' database covering world horticultural R&D from 1973 to date, and no references to daffodil rust were found. This may imply that daffodil rust is not a problem in other producer countries.

Possible causes of daffodil rust

<u>Fungal disease</u>

First, it should be stated that a true (fungal) rust disease ***is*** found on daffodils, though not (as far as we know) in the UK. In the Netherlands daffodils can be infected by the rust *Aecidium narcissi* which spreads from the reed-grass *Phalaris arundinacea* used as a covering material to protect bulbs and shoots from frost and wind-blown sand (van Aartrijk *et al*., 1995). Covering materials are not used on daffodil crops in the UK. Apart from that, reports of true rust pathogens on daffodils are very rare, though *Puccinia schroeteri*, *P. narcissi* and *Coleosporium narcissi* have been seen (Moore *et al*., 1979; Chastagner & Byther, 1985). The diagnosis of a true rust infection should be relatively easy. Finding a pathogen associated with daffodil rust has been more difficult.

Some parallels can be drawn between daffodil rust and another physiological disorder of daffodils, chocolate spot (which is unconnected with chocolate spot of beans caused by *Botrytis fabae*). Chocolate spot of daffodils has been known to growers and advisors much longer than rust, though it does not appear to have caused any serious concern and so is judged of little economic or other consequence. The symptoms consist of chocolate-coloured spots or streaks that appear sporadically on leaves. 30 years ago it would have been relatively easy to find a professional who could undertake such investigational work as part of an advisory role. It appears that some effort was expended in looking for a causal organism, according to the account of Moore *et al*. (1979):

> "In two successive seasons (1967/8) several hundreds of cultivars [of daffodils] in the collection at Rosewarne EHS [Experimental Horticultural Station] were examined for signs of [chocolate spot]. Most examples were from Groups 1A and 2A [basically non-white trumpet and large-cup cultivars] but not all these cultivars had the markings in both seasons. Histological preparations showed that the colour is caused by the death of the epidermal cells but no fungus or bacterium has been isolated consistently from the streaks. Scanning electron micrographs have shown that the cuticle is intact which seems to exclude a causal agent. A viral cause seems to be eliminated because no virus particles have been detected or isolated from narcissus seedlings with symptoms. Casual observations suggest that the appearance of the [chocolate] spots is associated with increasing ambient temperatures."

For daffodil rust there seems to have been no study comparable to this one on chocolate spot. It is known, however, that from time to time growers and advisors have sampled affected stems for examination at plant clinics (anecdotal reports, personal communications), and some examples are included under 'Surveys of growers'. No primary pathogen appears to have been isolated from these samples, though it is unfortunate that samples have been taken in an opportunistic way rather than as part of a structured programme. And it is, of course, almost impossible to prove a negative. Therefore, there is lingering doubt about entirely dismissing a pathogenic cause for daffodil rust (or, for that matter, daffodil chocolate spot). Attempting to carry out further systematic isolations from stems affected by daffodil rust seems to be justified, and in this project 'rusty' and 'healthy' samples from the ten sites will be examined by a diagnostics laboratory.

<u>Viral disease</u>

From a study of chocolate spot in the Netherlands it was reported that an unknown filamentous virus had been found both in apparently healthy daffodil plants as well as in those "with [viral] symptoms of yellow stripe in the leaf-tips, light green leaf discoloration, chocolate spotting or silver streak" (Kamerman *et al*., 1975). In Scotland "chocolate spot was observed in virus-tested [daffodil] clones, and it is suggested that it may be a physiological disorder induced by environmental conditions" (Mowat *et al*., 1983).

Could viruses be implicated in daffodil rust? From studies of the physiological disorders tip-burn and cigar-burn of stored cabbage, it was suggested that waterlogged or other adverse conditions contributed to the disorders through interfering with water uptake, with a consequent shortage of calcium in the rapidly growing leaves. It was shown that Turnip Yellows Virus (formerly called Beet Western Yellows Virus) and Turnip Mosaic Virus were primary factors in the development of tip-burn and cigar burn, respectively, both disorders being exacerbated by the presence of Cauliflower Mosaic Virus (Walsh *et al.,* 2004; Walsh, 2008). These three aphid-transmitted viruses have not been reported from daffodils, though most daffodil stocks are highly infested with several other viruses (Brunt, 1995). The peach potato aphid, *Myzus persicae*, is a common vector of both the viruses implicated in tip-burn and cigar burn and of some important daffodil viruses (Walsh *et al.*, 2004; Brunt, 1995).
As part of the current project, in 2014 tissue samples from stems with and without rust lesions will be frozen in liquid nitrogen and stored at -70°C at Warwick Crop Centre (WCC), University of Warwick. If required at a later stage, these samples will be used to screen for viral RNA.

Other pathogens

Diseases caused by bacteria and mycoplasma-like organisms are rare in daffodils (Hanks, 2013, p.44).

Nutritional problems

If the pathogen theory of daffodil rust is rejected, it seems reasonable to consider next a nutritional cause, since nutritional issues are known to be important in many physiological disorders. Deficiencies could be caused directly by the low level of a nutrient, by its low mobility (e.g. in the case of boron or calcium), by a pH effect (e.g. for boron, calcium or molybdenium), or indirectly by levels of another nutrient (e.g. of calcium by K, ammonium nitrate, Mg or boron). Several physiological disorders that are characterised by the production of darker brown or black spots on the surface or internally have been related to boron and (or) calcium deficiency (e.g. bitter pit of apples, internal rust spots of apple and 'five o'clock shadow' of carrot) (Swain, 1985). Similarities to other disorders related to nutrient deficiencies led to the suggestion by Andrew Tompsett (personal communication) that daffodil rust might result from boron deficiency, though no clear benefit was observed in boron spray trials. Ruamrungsri *et al*. (1996) reported the results of growing daffodil 'Garden Giant' in hydroponic culture with various nutrients omitted: the only physiological disorders reported were water-soaked areas near the base of the leaf when boron was omitted, and root-tip browning and death when calcium was missing.

It is known that occasional nutrient analysis of soil and tissue samples from daffodils with and without rust has been carried out for growers, in much the same way as disease diagnostics. No association between nutrient levels and the development of daffodil rust has been reported. In spring 2003, for example, soil and tissue samples were taken from examples of healthy and rusty daffodil crops grown by a Lincolnshire grower and analysed under the auspices of the HDC (HDC BOF Panel, personal communication). These results (described under 'Surveys of growers') showed no clear trend indicating a link between nutrient levels and the occurrence of daffodil rust. As for disease diagnostics, in 2014 a full set of plant and soil samples will be taken from each site for the analysis of macro- and micro-nutrient concentrations to re-examine this question.

Following analysis at WCC of nutrients in plants and soil of 'rusty' and 'healthy' daffodils, John Hammond (personal communication, 2010) suggested that daffodil rust was not directly a result of deficient nutrient levels, but "more an issue with rooting ability or development which is restricting uptake of nutrients overall, e.g. soil structure, which can explain why some crops grow out [of the daffodil rust symptoms] and the differences between cultivars".

Adverse environmental conditions

Some physiological disorders characterised by the production of darker brown or black spotting have been related to adverse environmental conditions (Swain, 1985). For example, dry hot summer weather and a sudden change in temperature have been implicated in apple leaf spot and drop, and water stress when transpiration exceeds water uptake, promoted by sudden checks in growth such as low temperatures, in lettuce dry (marginal) tip-burn. Rejecting a pathological cause for daffodil chocolate spot, Moore *et al*. (1979) suggested that the appearance of the spots is associated with increasing ambient temperatures. As well as rust and chocolate spot, daffodils can show a number of physiological disorders of flower development, and although little is know of their causes, temperature and adverse water relations have often been implicated (Moore *et al*., 1979; Rees, 1972).

- Bud death can occur shortly before spathe-splitting and flower opening, resulting in a dried-out bud atop the stem resembling a 'drumstick'. This is particularly a problem of double-flowered cultivars when forced under glass, and 'Golden Ducat' – our test cultivar for the present daffodil rust project because of its susceptibility – is very prone to it, even when field-grown. This disorder was investigated in 'Golden Ducat' and other double cultivars in HDC project BOF 17 (Hanks, 1992). Dissections of the stem and bud during development in the glasshouse suggested the plants were unable to translocate sufficient water to support the rapid growth of such dense bud tissues. Misting, calcium, growth retardant and fungicide treatments did not prevent bud death.

- Bud death in *Narcissus poeticus* 'Flore Pleno' ('Tamar Double White') is a specific instance of the disorder. This cultivar was at one time a speciality crop of the Tamar Valley and was studied intensively at the time (Tompsett, 1972; Rees, 1972; Moore *et al*., 1979). Growing on warm slopes, extreme temperatures, hot, dry growing seasons, wet autumns and adverse water relations and a poor root system were suggested as predisposing factors. However, irrigation, mulching and shading treatments were unsuccessful in preventing bud death.
- 'Bullhead' is a physiological disorder resulting in abnormal flowers and abnormal flower opening in another double cultivar, 'Cheerfulness' (Tompsett, 1979; Moore *et al*., 1979). High temperatures were also linked with this disorder.

A possible link between daffodil rust, soil temperature and water availability was investigated in HDC project BOF 62 (Fellows & Hanks, 2007). Pot-grown bulbs of six cultivars were placed in a polyethylene/mesh 'thermo-gradient tunnel' at Warwick HRI, Wellesbourne (Wurr *et al.,* 1996). This facility maintained a temperature gradient from *ca* 4°C above outside temperature at the warm end to *ca* 0.5°C above outside temperature at the cool end. The treatments consisted of high, medium and low temperatures (plants placed at the warm or cool end of the tunnel or at the mid-point), combined with four levels of soil moisture from near-dry to very wet (42, 56, 69 and 82% of maximum water capacity, respectively). However, there was no development of daffodil rust symptoms in any of the treatments, so the hypothesis that conditions linked to temperature and water availability cause the disorder through the disturbance of normal water relations could not be tested. The range of treatments used in this project may have been too mild to be effective.

Research approach

The continuing unpredictable losses and down-grading of cut-flowers due to daffodil rust encouraged the HDC to commission the present project in 2012. Ultimately the aim of research would be to discover the cause of daffodil rust and propose strategies for reducing or eliminating it. Project BOF 76 aims specifically to test the proposition that daffodil rust develops as a consequence of adverse soil/water relations. Despite the failure of an earlier project (BOF 62) to provoke daffodil rust development through applying a matrix of temperature and soil water content treatments, it is possible that the treatments in that project were simply not sufficiently extreme to do so. A different and more strategic approach was taken in the present project, which involved aiming to maximise the chance of daffodil rust occurring in experimental plots. Bulbs of a stock of the daffodil rust-prone cultivar 'Golden Ducat' were planted on ten diverse sites across west Cornwall to provide a range of natural soil, water and other environmental conditions, and variations in husbandry, that might lead to the development of daffodil rust to different extents across the sites. Soil, water and other environmental variables were monitored and regression analysis used to identify any association between daffodil rust development and specific conditions. The emphasis on soil and water conditions was not seen as excluding making use of the opportunity of a multi-site experiment to carry out systematic soil and plant nutrient analysis and disease diagnostics to test further the possible pathological and nutritional origins of the disorder.

The body of this report covers, first, the surveys of growers, and to bring all the survey information together both the 2003-2004 and 2011-2013 responses are included. Secondly, the first year of field-work (2012-2013) and its preliminary data analysis are described.

Surveys of growers

A survey of daffodil growers was undertaken by the HDC in 2002 and 2003 to discover the impact of the disorder on production. The responses were analysed as part of the earlier HDC daffodil rust project (BOF 62). Subsequently, to assess whether the grower perception of daffodil rust had changed over the intervening decade, the HDC carried out a follow-up survey in 2011, 2012 and 2013, to be summarised as part of the present HDC project (BOF 76). For easier cross-referencing, all five years of the surveys are summarised in the present report.

Survey methodology

Questionnaires were developed by HDC technical staff and BOF Panel members. They followed a similar pattern over the two survey periods, though details varied as experience was gained. In 2002 and 2003 the questionnaires were posted to all daffodil growers on the HDC data-base, while in 2011 to 2013 they were sent by broadcast email. In 2013 the process was further adapted and two questionnaires were sent out, first asking for the "dates of first occurrence of [daffodil rust] symptoms", and secondly, requesting a "summary for 2013". In addition, in 2013 growers were asked if they had previously submitted daffodil rust-affected plant and soil samples for disease diagnostics or nutrient analysis and, if so, whether they would be prepared to share this information with the researchers. Responses were encouraged through reminders in the broadcast HDC Weekly Newsletter and by personal contacts. Examples of the blank questionnaires are shown in Appendix 1.

In addition to these surveys of growers, in 2013 a flower trader was able to make available a record of the total number of flower 'lots' received that season, and the number of them that were free of, or affected by, daffodil rust. A lot was defined as one variety received from one grower, irrespective of the number of deliveries of each received. For each lot, a random sample of 160 to 200 bunches (two full flower trays or boxes) was examined and scored for rust severity (Table 1), recording the maximum score seen.

A key element of all these assessments was the scale of 0 to 5 used for scoring the severity of daffodil rust. In 2002 and 2003 the scores and guideline illustrations used were those provided by Andrew Tompsett (personal communication, 2002) and shown in Table 1. In 2011, 2012 and 2013 these descriptions were adapted in the hope of providing clearer criteria for defining 'very slight' or 'slight' daffodil rust (Table 2). Both scales were intended for recording daffodil rust from a commercial viewpoint, with a score of 3 indicating daffodil rust severe enough to downgrade the stems, and scores of 4 and 5 indicating stems that were unmarketable because of more extreme damage.

Table 1. Daffodil rust severity scores developed by Andrew Tompsett and used in the HDC surveys of 2002 and 2003.

Score	**Description**
0	No symptoms
1	Slight blistering of epidermis only - "not really rust"
2	Slight cracking and rusting
3	Moderate cracking and rusting – becoming disfiguring
4	Severe daffodil rust, very disfiguring, flowers unmarketable
5	Very severe daffodil rust, cracking and stem bending, flowers unmarketable

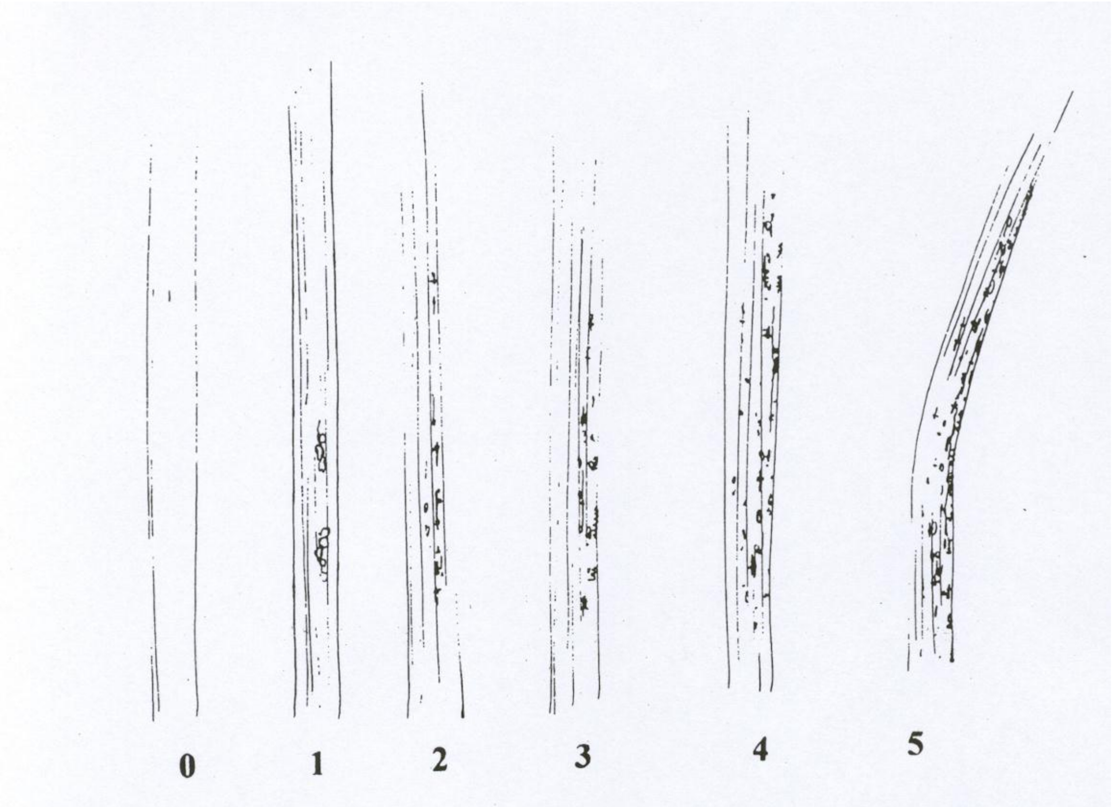


Table 2. Daffodil rust severity scores modified for use in the HDC surveys of 2011 to 2013.

Score	Description
0	No symptoms or slight blistering of epidermis only
1	Very slight daffodil rust (only one or two small lesions per stem, or small lesions that are not fully rust-coloured)
2	Slight daffodil rust, not disfiguring, no effect on sales
3	Moderate daffodil rust, downgrade or find other buyers
4	Severe daffodil rust, some cracking across stems, unmarketable
5	Very severe daffodil rust, obvious cracking and bending of stems, unmarketable

While efforts were made to make these surveys representative, and to interpret the data fairly, it should be stated that they were essentially limited and random spot-checks limited by commercial accessibility. The results should therefore be interpreted with caution. Unfortunately the surveys did not provide much information on stocks or cultivars that did not show daffodil rust symptoms.

Results of 2002 survey

One hundred and eighty-three survey forms were posted by the HDC in January 2002. Forty-four replies were received, a response rate of 24%. The respondents accounted for 1715ha of daffodil production, some 43% of the total area grown in England and Wales at the time. The main findings of the 2002 survey are shown here.

- Sixty-eight per cent of respondents had seen daffodil rust in the previous four or five years.
- Respondents estimated their loss of turnover due to daffodil rust varied between 4 and 15% in their worst year, and between 0 and 3% in their best year.
- In 2002, 36% of respondents had seen flower sales reduced through daffodil rust, 36% had seen flowers down-graded, 25% had been unable to supply their preferred customers or had had to seek alternative outlets, and 25% had experienced totally unmarketable flowers.
- Twenty-three cultivars were included as exhibiting daffodil rust in the returns for 2002. ‘Golden Ducat’ was mentioned 13 times, ‘Carlton’ and ‘Golden Harvest’ each nine times, and ‘Mando’, ‘Saint Keverne’ and ‘White Lion’ each five times. ‘Golden Ducat’ and ‘Mando’ were cited a disproportionately large number of times compared with the relatively small areas of these cultivars grown.
- Nine of these cultivars were reported to have shown daffodil rust in their first crop-year, though these did not include the apparently rust-prone ‘Golden Ducat’. All 23 cultivars were reported to have shown daffodil rust to some level in their second crop-year.

- The numbers of cultivars showing daffodil rust scores of 1 through 5 were 14, 8, 6, 9, and 5 out of 23, respectively (different sources or stocks of a variety would have different scores, so the numbers did not add to 23). The cultivars with the highest score, 5, were ‘Carlton’, ‘Ice Follies’, ‘Saint Keverne’, ‘Kerensa’ and ‘White Lion’ – but not ‘Golden Ducat’ or ‘Mando’.

Some respondents mentioned that flower sales were of increasing importance at a time of falling bulb prices, so research on the problem would be welcome. Other comments made included the following.

- “Symptoms usually occur with repeated mild & damp/cold/mild & damp conditions. The belief is that the crop grows too fast after a cold frosty spell and that the flower stems are brittle. Signs of daffodil rust can be traced on the stems almost to the first day of the cold period.”
- “The 2002 season was good considering it was a wet mild winter.” “Rust not a problem in 2002 (in Scotland) as weather has been dry and cool.”
- “Rust does not appear to affect the whole field – often only parts and not the same part each year.”
- “Suggest that the problem is due to boron deficiency.”
- “Only seen rust in a two year down crop, ex forced stock and mainly in waterlogged areas of the field.”
- “If rust is present in the first year, it always occurs in the second year.” “Crops more prone to stem rust in third year down.”
- “Gannilly [*sic*], St Keverne, Karenza [*sic*] and Golden Ducat show problems almost every year.” “Rust is a severe problem on Golden Ducat…” “Golden Ducat seems to get rust every year.” “Golden Ducat is the only variety that causes concern.” “Generally find that Golden Ducat and Carlton are worst affected.” “Tamara… is susceptible.”
- “Not really a problem on Isles of Scilly although there was one case reported for Soleil d’Or.”

Results of 2003 survey

A similar number of survey forms were posted in early-2003. Eighteen responses were received; a response rate of only 10%, and two of the respondents grew daffodils exclusively in pots so were excluded from further assessments. The remaining 16 growers accounted for 872ha of daffodil production, some 22% of the total area grown in England and Wales. The main findings of the 2003 survey are shown here.

- Seventy-five per cent of respondents had seen daffodil rust in the previous four or five years.

- The proportion of respondents, who had seen daffodil rust in 2001, 2002 or 2003, were 69, 75 and 63%, respectively.
- Of the respondents providing actual figures for the periods before 2001, or in 2001, 2002 or 2003, their average percentage area affected by daffodil rust varied between 6 and 9% across the four periods, average area where flowers were downgraded between 3 and 5%, and average area where flowers were unmarketable between 1 and 3%, with an average reduction in turnover between 2 and 3%.
- 'Golden Ducat' was again the cultivar with most incidences of daffodil rust recorded.

Results of 2011 survey

In 2011 seven responses were received in response to the broadcast email (Table 3). The respondents were growing between 15 and 950ha of daffodils each, a total area of 1771ha, representing about 38% of the total area of daffodils grown in England and Wales at that time. Despite the low response rate, the fact that two of the respondents were together growing 1350ha of daffodils, about 34% of the total grown, means that these observations can hardly be dismissed. In 2011 growers were also asked about their overall impressions of daffodil rust, and how these had changed since the previous surveys. The main findings are listed here.

- Six of the seven growers replied they had seen more daffodil rust in the past 6 years than before.
- Five out of seven had had to downgrade flowers, of whom four had seen overall flower sales reduced, had not been able to supply their preferred customer, or had had to seek alternative markets, and three had experienced completely unmarketable flowers. Some had been able to find alternative and less particular markets for poorer or downgraded stems, while others thought this practice was unacceptable and considered it better not to pick poor stems; one commented that "stems can split after picking…"
- Estimated losses in revenue due to daffodil rust had varied from 1 to 15%, or £100k to £221k, in their worst year, while in the best years there were no losses from daffodil rust.
- Some respondents mentioned that 2010 or 2011 were the worst years for daffodil rust so far encountered.

Grower comments included ideas on the factors thought to favour the development of daffodil rust, including those listed below.

- "In years when rust is a problem, it can usually be traced back to a gap in the spray programme."
- "The winter of 2011 started very cold and was 4 weeks late [resulting in] the crop growing very fast once it got moving, to the point that we were cropping 4 weeks of

flowers in just over 2 weeks. Winters prior to this were considerably milder and although we see rust every year… the actual losses were considerably less. However, I also believe that post 2001, we started to use more advanced fungicides on the crop than in the 1990s and I think that the use of Amistar and in particular Folicur, which has cell wall shortening properties has had an effect on the levels of rust. Prior to 2001 my memory tells me that outbreaks were more frequent and severe (similar to this year), and were certainly linked to the prevailing weather patterns. In particular, repeating periods of frost, followed by rapid thaw and heavy rain would induce outbreaks. There was a theory… that you could estimate the date of the outbreak by measuring the position of the rust on the stem/leaf compared to the growing tip and dividing the amount by 1cm. This would give you a date in time that would invariably relate to a hard frost being broken by a rapid rise in temps and heavy rain."

- "Rust can appear after a first picking where the crop has been clean. Second and third picks can be more affected following a clean first pick. Second and third picks can be stunted and twisted particularly in Ganilly. Golden Ducat can exhibit symptoms from the outset. Seemingly good flowers can deteriorate in the tray and rust marks lead to brittle stems and cracking. Varieties that have two sharp spines to the flower stem [are] very badly affected e.g. Dellan and St Keverne."
- "Rust seems to be worst during/after a period of adverse weather e.g. cold/frosts, winds."
- "Carlton and Golden Ducat are the two varieties which show symptoms of rust on an annual basis. The severity of the symptoms [is] very dependant on climatic conditions. The expression of the disorder is usually with distortion to the flower stems and leaf in Golden Ducat, and with brittle breaking flower stems in Carlton. The symptoms are often seen after a cold spell being followed by a milder [growing] spell."
- "We have trialled various trace element treatments, inc copper, boron and manganese - with no obvious reduction of rust."
- "The rust in 2011 seemed to develop after rapid growth after the snow melt. Later varieties grew slower while it was dryer and we saw no rust. The Year 3 is more susceptible[,] I think we never see it much in 2 year down crops."

Results of 2012 survey

In 2012 only three responses were received (included in Table 3). The respondents were growing between 10 and 135ha of daffodils each, a total of 156ha, and, as this represents only about 1% of the total area of daffodils grown in England and Wales, the data should be treated with caution. Of the seven stocks reported on, the highest daffodil rust scores were

2, occurring in stocks of ‘Martinet’ and ‘Golden Ducat’, and 1.5, occurring in one stock of ‘Saint Keverne’; the other four stocks scored 1.

Grower comments included:

- “…very little rust this season in complete contrast to 2010/11”;
- “…the worst variety was ‘Golden Ducat’… [but this] did not cause problems this year as the doubles were wanted [i.e. strong demand ignored the rust symptoms] but in some years the extent of rust on the last 2 picks could [have] created problems”;
- “[rust] seemed to start after rapid growth following two cold weeks”.

Results of 2013 survey

A questionnaire asking about the dates of first daffodil rust occurrence in 2013 was sent by broadcast email on 8, 15 and 22 February and 1 and 8 March to levy-payers in the BOF sector (1,418 recipients, including 380 daffodil growers). The broadcasts were opened by 266 recipients (19% of the total) and 20 recipients (1% of the total) also clicked on the link in the broadcast. Only three growers – corresponding to <1% of the daffodil growers - returned completed questionnaires. A second questionnaire, asking for a summary of daffodil rust damage, was sent by broadcast email on 24 May specifically to daffodil growers (380 recipients); it was opened by 89 recipients (23%) and just one returned a completed questionnaire. The four respondents were growing between 10 and 900ha of daffodils, a total of 1160ha, representing about 29% of the area grown in England and Wales. As in the 2011 survey the fact that one of the respondents was growing 900ha of daffodils, about 22% of the total daffodil area, means that the observations, though derived from so few growers, cannot be dismissed (included in Table 3). A panel member, who telephoned other growers over this survey period to seek further responses, concluded there was reluctance to discuss the issue, perhaps because of commercial sensitivities and the possibility of giving the product a bad name.

Of the 27 cultivars or stocks reported on (some cultivars were represented by more than one stock), the highest daffodil rust scores were 4 to 5 which occurred in one stock (third-year-down ‘Dellan’) and 3 which occurred in two cultivars, one of which worsened substantially over the observation period and was then downgraded to a lower specification. The other cultivars showed only mild daffodil rust symptoms, mostly scoring 1. The following comments were made.

- “[There was] low level rust from our own stocks [and locally bought-in stocks] across the board… all crops exhibiting very brittle stems when cropping… [There was] very high rainfall… mild January… early season… The incidence of rust was very widespread this winter but at a low level and certainly not at a level to affect sales apart from one incidence [where the product] was withdrawn from the packing line but subsequently sold to a lesser specification customer…"

- "[Rust was] terrible in 3rd-year-down 'Dellan' [but] not a problem in others". This grower estimated that losses ranged from 2% to 30% across the different stocks.

Summary of grower surveys

- Despite a poor response to the later surveys, the data received indicated that daffodil rust has remained a problem over the whole period 2002 to 2013, and has probably increased. The decline in responsiveness to surveys may have been a result of administrative overload, commercial sensitivity (including the possibility of giving the product a bad name) or perhaps acquiescence that "rust happens" but is severe only in some years and in certain varieties and "nothing can be done about it even if you knew what caused it".
- Daffodil rust can occur in many cultivars, but only a few cultivars are especially susceptible to it; unfortunately these include two varieties very much in demand, 'Golden Ducat' (a popular, late-flowering double daffodil) and 'Carlton' (out-dated and base rot-susceptible, but nevertheless popular and classic variety).
- Daffodil rust can occur in the first, second and subsequent crop-years: there is no evidence that it does not appear in first-year-down crops.
- Theories about the causes of daffodil rust appear to centre around (a) wetness and (b) marked changes in temperature.
- Growers may think of daffodil rust as a fungal disease and therefore implicate spray programmes as affecting its severity.
- Samples sent by growers for analysis do not appear to have suggested that a pathogen or nutrient imbalance is responsible for the occurrence of daffodil rust.

An example of between-year differences in daffodil rust severity is illustrated for 2011 to 2013 in Figure 1. Daffodil rust severity scores varied greatly across the three years, with 2011 showing the highest severity scores. Among the 18 stocks reported on in this period, scores of 4 or 5 (meaning the flowers were virtually unmarketable) were reached by nine stocks, with four more reaching 3 (meaning downgrading the flowers). These high scores covered eight cultivars and five of the six growers who had provided varietal information. In cases where crops of a cultivar in different crop-years were included, not all crop-years exhibited daffodil rust to the same degree, and, generally, it was less severe in the younger crops.

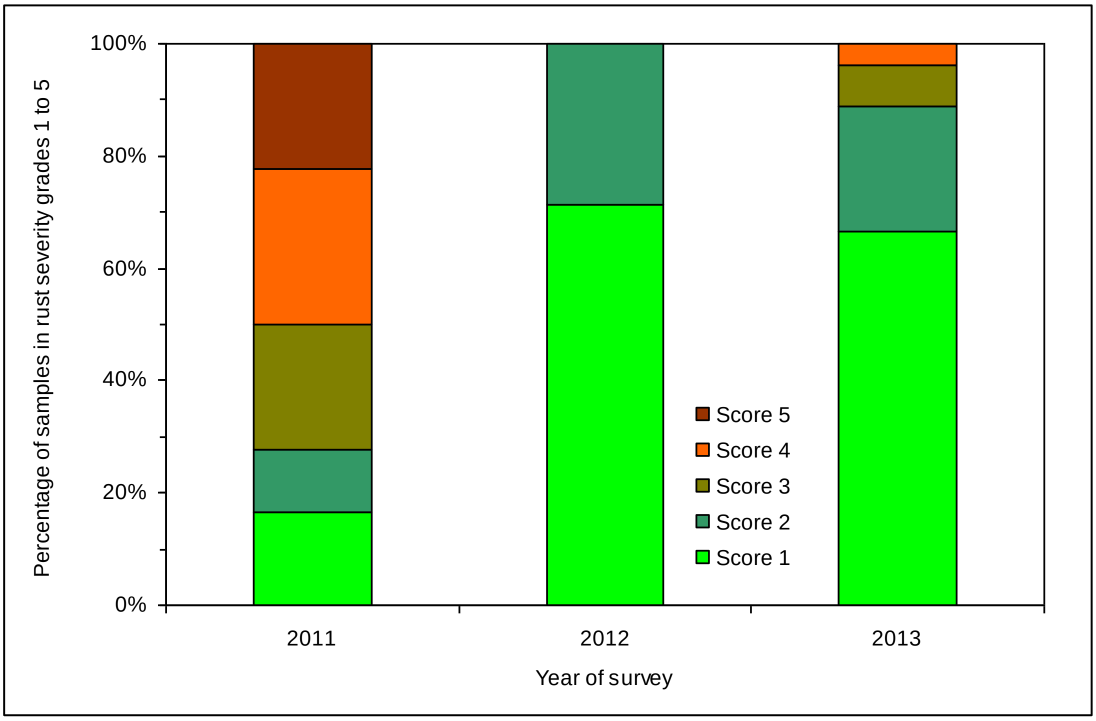


Figure 1. Frequency of daffodil rust severity scores in 2011, 2012 and 2013 surveys.

Flower trader's survey 2013

The survey covered a year's intake of flowers, comprising 165 'flower lots' (flowers of one variety from one grower). The severity of daffodil rust symptoms of each lot was assessed on a scale of 0 to 5 (Table 1), and, overall, 135 lots of the 165 (82%) were free of the symptoms. The results, based on the percentage of lots having each score and classified according to the county of origin, are shown in

Figure **2**. For lots from Lincolnshire, Norfolk and Cornwall, 75, 81 and 91% of stocks, respectively, were free of daffodil rust, contrary to the expectation that the disorder was more frequent in Cornwall than in other bulb-growing areas. Only six lots were not from these three counties, and these data have been excluded. As this survey includes widely divergent numbers of lots per county, these results should be used with caution.

Classified by cultivar, the following had positive scores a disproportionate number of times: 'Golden Ducat', 'Kerensa', 'Mando' and 'Tamara'. These cultivars are known to show the symptoms regularly, but the results were probably skewed because, apart from 'Kerensa', they are widely grown. 'Golden Ducat' probably has the most economic significance because its longer stems and heavier buds are favoured, though this probably contributes to more stem breakages during handling, accentuated by its susceptibility to rust. It is hoped it will be possible to repeat this survey in 2014.

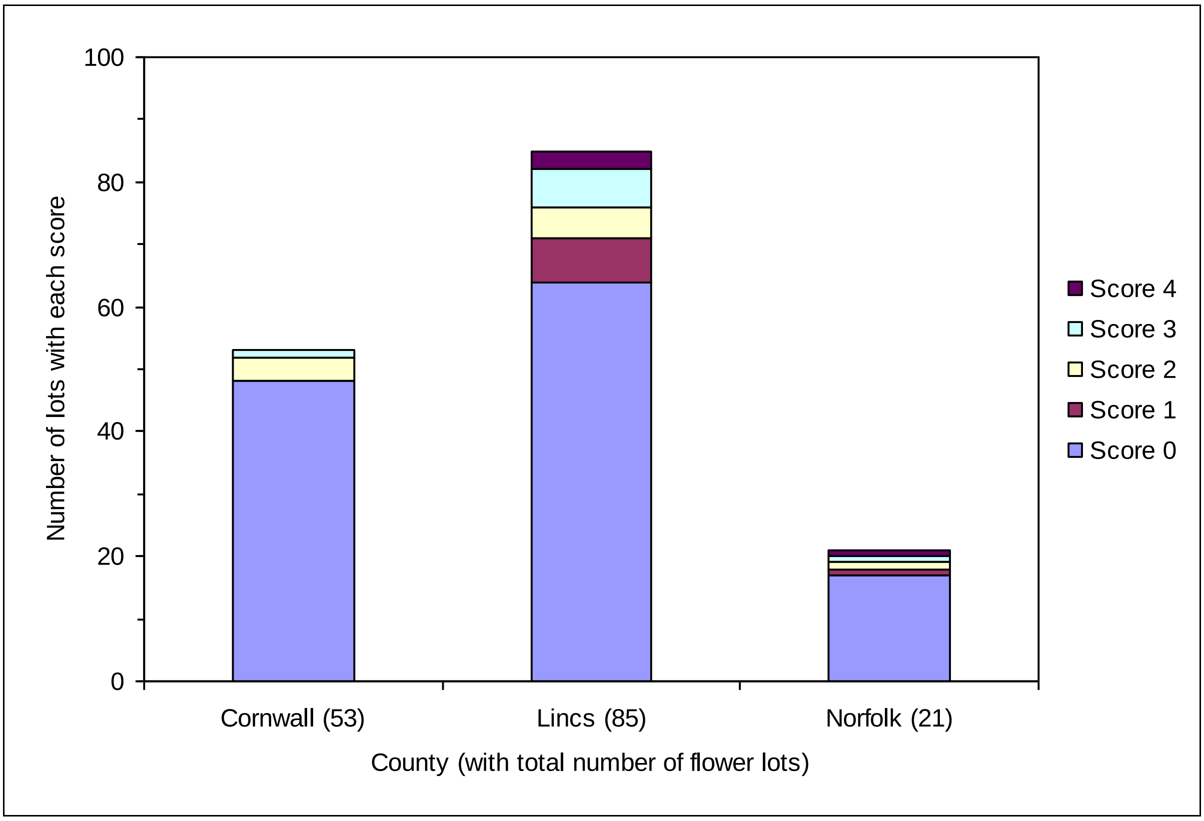


Figure 2. The number of flower lots having maximum daffodil rust scores of 0 to 5, classified by county of origin, from a flower trader's survey of sendings received in 2013.

Table 3. Incidence and severity of daffodil rust reported in the 2011 to 2013 surveys of growers.

Grower number and total area grown	Number of stocks and crop-years having rust [1]	Dates when rust first seen	Numbers of stocks and cultivars with different rust severity scores [3]				
			1 or 1.5	2	3	4 or 4.5	5
Survey year 2011							
1 na	1 All	na [2]	1 'Grand Soleil d'Or'	0	0	0	0
2 950ha	4 3rd, 4th	na	0	0	0	4 'Golden Touch'; 'Golden Ducat'; 'Standard Value'; 'Saint Patrick's Day'	0
3 30ha	8 2nd upwards	na	2 6th-year 'Saint Patrick's Day'; 2nd- and 4th-year 'Camelot' but not some other years	1 6th-year 'Carlton' but not younger crops	2 2nd-year 'Ganilly' and 3rd-year 'Golden Ducat' but not some other years	1 3rd-year 'Ganilly'	2 2nd-year 'Dellan' and 6th-year 'Golden Ducat' but not 1st-years
4	2	na	0	0	0	0	2

400ha	1st to 3rd						2nd-year 'Golden Ducat' and 2nd- and 3rd-year 'Carlton' but not 1st-year crops
5 22ha	2 2nd	na	0	1 'Carlton'	1 'Golden Ducat'	0	0
6 15ha	na	na	-	-	-	-	-
7 100ha	1 1st to 3rd	na	0	0	1 3rd-year 'Tamsyn' but not 1st- or 2nd-year	0	0
Survey year 2012							
8 10ha	3 3rd and 4th	December ('Martinet') to 2 February ('Saint Keverne')	2 3rd-year (but not 2nd-year) 'Mando'; 'Saint Keverne'	1 'Martinet', with some down-graded	0	0	0
9	3	3 March	2	1	0	0	0

12ha	2nd	('Carlton') to 20 March ('Standard Value')	'Carlton'; 'Standard Value'	'Golden Ducat'			
10 135ha	1 3rd	27 February	1 'Tamsyn'	0	0	0	0

Survey year 2013

11 900ha	11 2nd and 3rd	8 January ('Tamara') to 5 February ('Golden Harvest' and 'Investment')	9 (wide range of cultivars	1 3rd-year 'Jedna' (score 1 for 2nd-year 'Jedna')	1 'Mando', (score in-creased from 1 (15 January) to 3 (3 March) when product down-graded)	0	0
12 10ha	3 3rd and 4th	"At flowering"	1 'Standard Value'	1 'Saint Keverne'	1 'Martinet'	0	0
13 100ha	3 2nd and 3rd	10 January (3rd year 'Tamsyn') to 18 March ('Dellan')	0	3	0	0	0

14 150ha	10 1st to 3rd	na	8 including 1st-year ‘Dellan’	1 2nd-year ‘Dellan’	0	1 3rd-year ‘Dellan’	0

[1] Other inspected stocks were unaffected by daffodil rust (or were assumed to be unaffected); some participants listed several unaffected cultivars

[2] na, not available

[3] See Table 2

Soil and plant analysis undertaken by growers

In the 2011 survey three growers commented on sending soil or plant samples associated with daffodil rust symptoms for mineral analysis or disease diagnostics. One commented "we have sent many samples for analysis including soil, bulb and leaf tissue… all of the tissue and soil samples have all come back from the lab over the years showing normal or near normal levels of nutrient and no deficiencies. There have never been any pathogens found in any of the samples sent that would/could cause the deformities that are associated with rust…" The other comments were "I have had flower stems and the soil sampled in the past… Results were inconclusive and remedial action taken did not solve the problem", and "Sample sent to Netherlands for analysis, no organisms present…"

Two cases were reported in further detail.

Case 1

In March 2003 a Lincolnshire grower experienced severe daffodil rust in some areas of his crop and sampled plants and soil for analysis. Samples were taken from normal crops of 'Golden Ducat' and 'Saint Keverne' and from crops of the same cultivars that had severe daffodil rust with unmarketable stems. In the case of the normal 'Golden Ducat', samples were taken from (a) a uniformly normal area of the crop and (b) by selecting normal plants from an area of mixed quality, these being referred to as 'random' and 'selected' samples, respectively. The crops were in their second or third crop-years and none had received fertilizer in the base before planting or subsequently as top-dressing.

Soil samples were analyzed for pH, P, K, Mg and, because it has been associated with physiological browning and tissue breakdown in a wide range of crops, calcium. The results are given in Figure 3. The graph showed that normal and unmarketable samples were not differentiated by having distinctly grouped pH or P concentrations, i.e. the values for normal and unmarketable samples were intermingled. However, in the case of K and calcium concentrations the unmarketable samples had lower levels than the normal samples, whereas for Mg the reverse was true. According to the recommended levels given in 'RB209', both soil samples that yielded unmarketable samples were deficient in K.

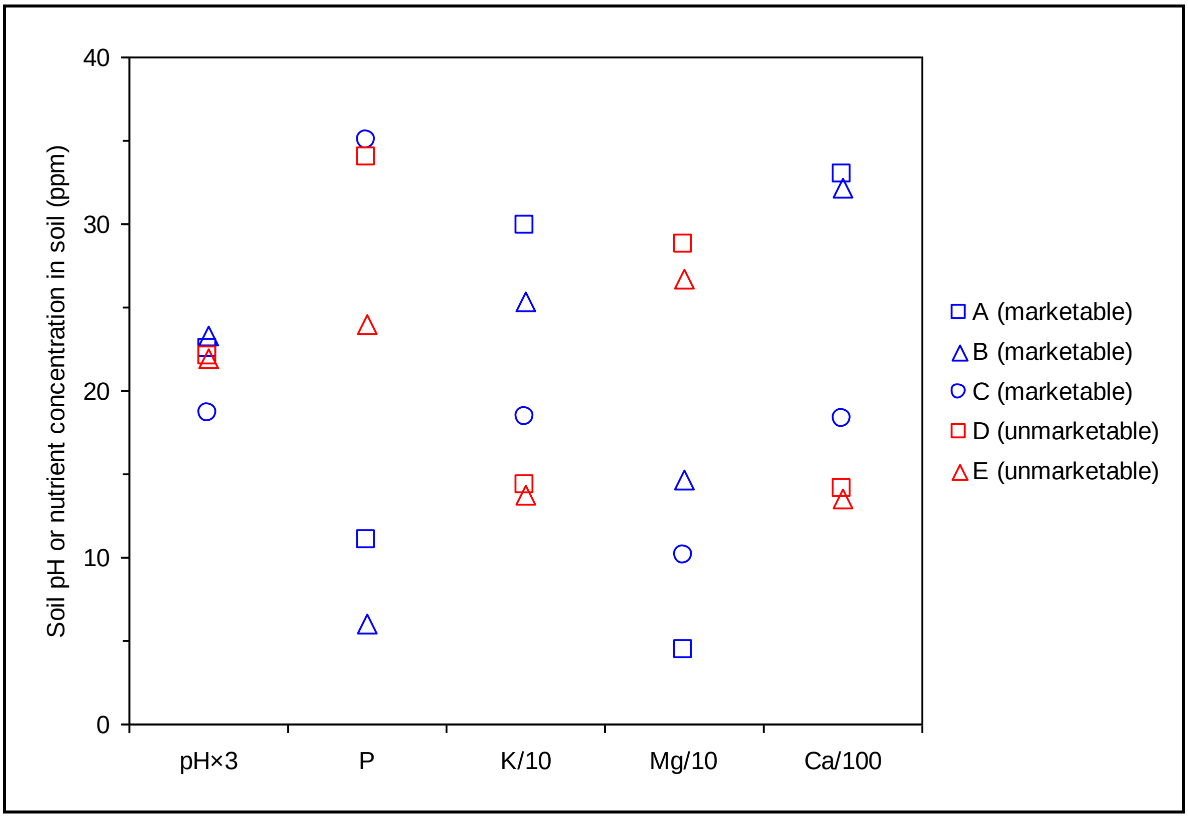


Figure 3. Soil pH and concentrations of P, K, Mg and Ca at three sites producing rust-free, marketable stems and two sites yielding unmarketable stems due to daffodil rust. Samples A, B and D are 'Golden Ducat' (A is 'random' sample, B is 'selected' sample, see text) and samples C and E are 'Saint Keverne'. For convenience, the values for some variables have been scaled to bring them on-scale (indicated by ×3, /10, etc.).

Bulb and stem samples were analyzed for major and trace nutrients, with results shown in Figure 4 and Figure 5, respectively. In all but three cases the values for normal and unmarketable samples were intermingled, showing no characteristic high or low levels dependent on the daffodil rust status of the plants. The exceptions occurred in bulb samples only, with unmarketable samples having lower concentrations of calcium and iron and higher concentrations of Mg.

It is tempting to highlight the lower levels of soil and bulb calcium in unmarketable plants (and perhaps the antagonism with Mg), but there were no indications of any other associations. However, the results were based on five samples and the normal and unmarketable stems were from crops in different locations. Further, some calcium-related disorders are due to the lack of mobility of the calcium ion and the availability of soil water, not simply to actual concentrations; and finally, early experiments on bulb nutrition showed that nutrient reserves within the bulb could delay the onset of nutrient deficiencies due to any inadequacies of the growing medium for a period of years. Interestingly, the analyst in the current case noted that the results of analysis might be more meaningful if soil moisture variations at the sites were also taken into account.

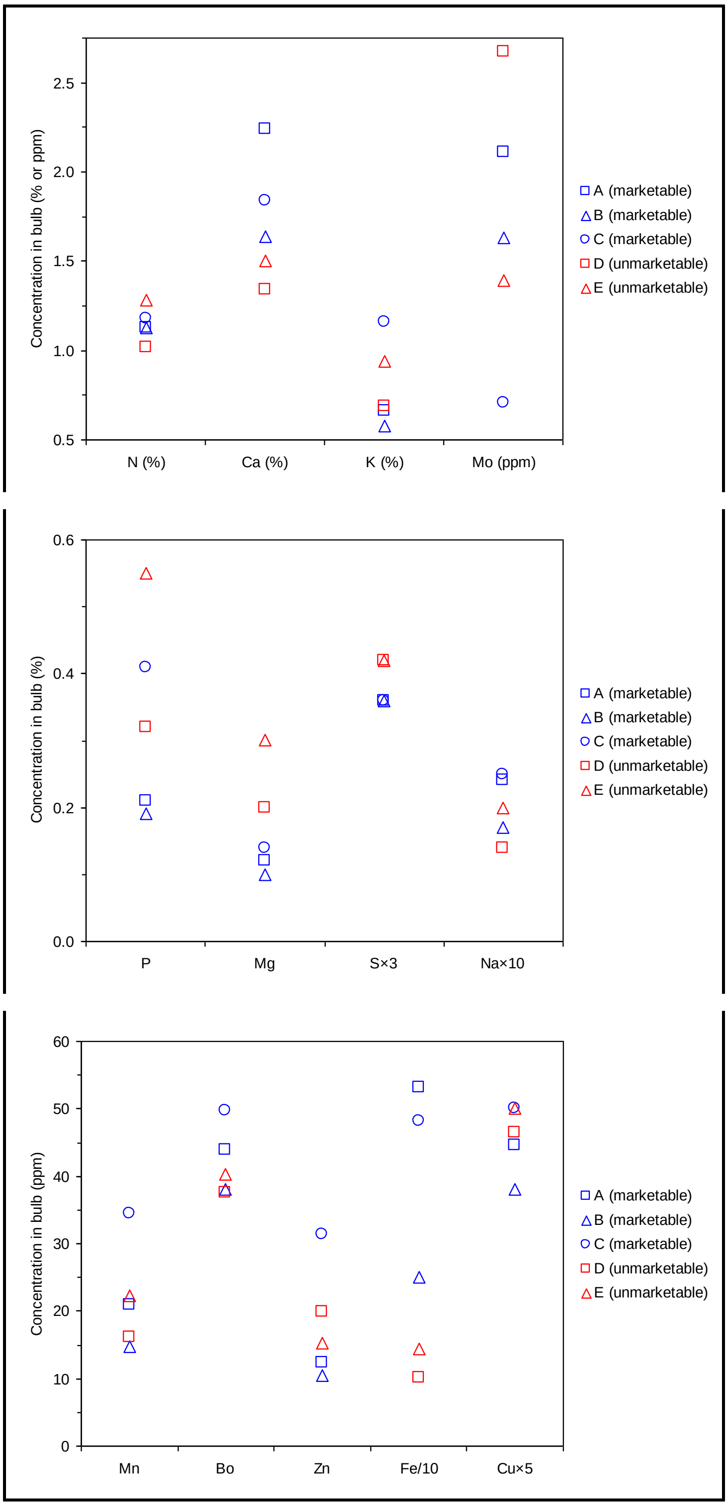


Figure 4. Concentrations of nutrients in bulbs at three sites producing rust-free, marketable stems and two sites yielding unmarketable stems due to daffodil rust. Samples A, B and D are 'Golden Ducat' (A is 'random' sample, B is 'selected' sample, see text) and samples C and E are 'Saint Keverne'. For convenience, the values for some variables have been scaled to bring them on-scale (indicated by ×5, /10, etc.).

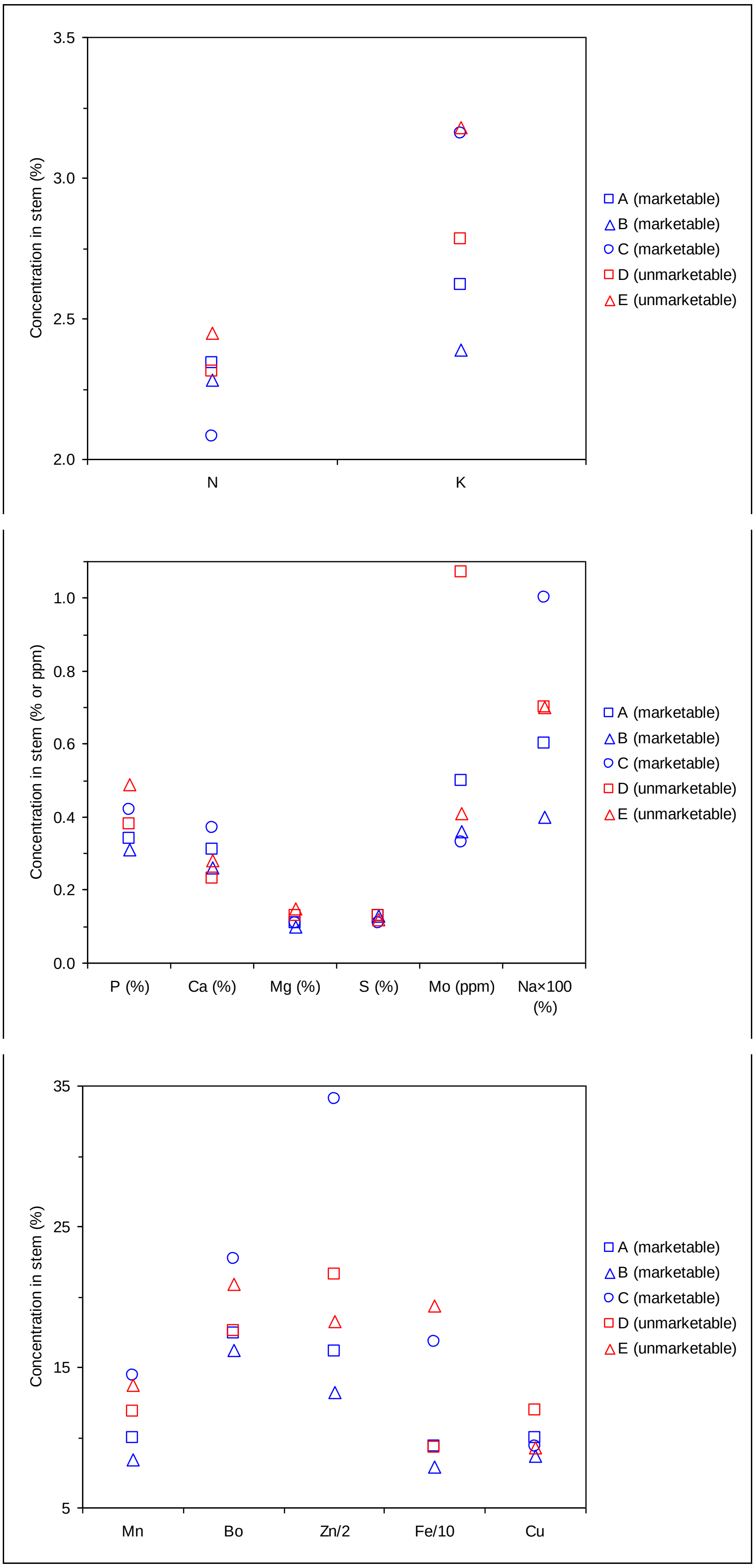


Figure 5. Concentrations of nutrients in stems at three sites producing rust-free, marketable stems and two sites yielding unmarketable stems due to daffodil rust. For explanations of A to E and scaling of values (e.g. ×100, /2, etc.) see Figure 4.

Case 2

In June 2012 a grower in Cornwall experienced damaging levels of daffodil rust in one area of a field, though stems were of a high quality in the remainder of the same field. Soil samples were therefore taken and analysed from both the ‘good’ and ‘rusty’ areas.

The only specific problem with the soil from the rusty area was a high calcium content: high calcium would be expected to restrict Mg availability. On the other hand the peculiarity of the good site was a very high level of iron (suggested as possibly the result of poor drainage). Both sites were low in boron and manganese and high in copper, zinc and molybdenum. There were no other substantial differences between samples in the levels of macro- or micro-nutrients or soil pH.

These results might appear to suggest ***low*** Mg availability as a possible cause of daffodil rust (though in Case 1, above, soil Mg levels were ***higher*** in the rusty areas), and also that low levels of boron or manganese are unlikely to be a cause of daffodil rust. However, this is a report of a single example which may mean little in general: it would be useful to see the results of similar analyses which growers are likely to have obtained from time to time.

Field-work: materials and methods [1]

Objective

The aim of the field-work was to test the proposition that the soil-water environment (e.g. soil structure, water availability, temperature or nutritional status) affects the occurrence, incidence or severity of daffodil rust. The occurrence of daffodil rust damage is unpredictable, so the work was structured to increase the likelihood that the disorder would occur in at least some of the experimental plots: the cultivar used, 'Golden Ducat', is very susceptible to daffodil rust (see survey results above), ten sites with a variety of soil types and topography were used, and the work was located in west Cornwall, the region of the UK where it was believed daffodil crops seem most prone to daffodil rust.

Bulb material

A suitable stock of narcissus cv 'Golden Ducat' was sourced following consultation with the HDC Industry Representative and others. Two hundred and fifty kg of bulbs of each of two grades, 10-12 and 12-14cm circumference were obtained. The bulbs had received standard hot-water treatment. From each grade 25kg of bulbs were allocated for planting at each of the ten sites. Twenty-five kg of bulbs is equivalent to *ca* 610 and 425 bulbs of the smaller and larger grades, respectively (for convenience "per 1,000 bulbs" is used in the text).

Sites

Following discussion with bulb growers about suitable sites, ten commercial daffodil fields were selected, taking into account the requirement to locate them throughout west Cornwall and to include varied soil types, topography and husbandry. The names used for the sites reflected current usage locally and are not necessarily definitive or unique.

- Site locations are shown in Figure 6 and Figure 7.
- Site locations, topography and aspect are in Table 4.
- Information supplied by the growers on pre-planting soil analysis are provided in Table 5 and details of previous cropping, fertiliser and lime applications and dates of bulb planting in Table 6.
- Grower assessments of soil types and soil information from the Soil Map of England and Wales (Soil Survey of England and Wales, 1983) are in Table 9 (under the 'Field-work: results' section), along with some other soil quality assessments.

[1] For accurate reporting materials may be referred to by the names of commercial products. No endorsement of the products mentioned is intended, nor criticism of products not mentioned.

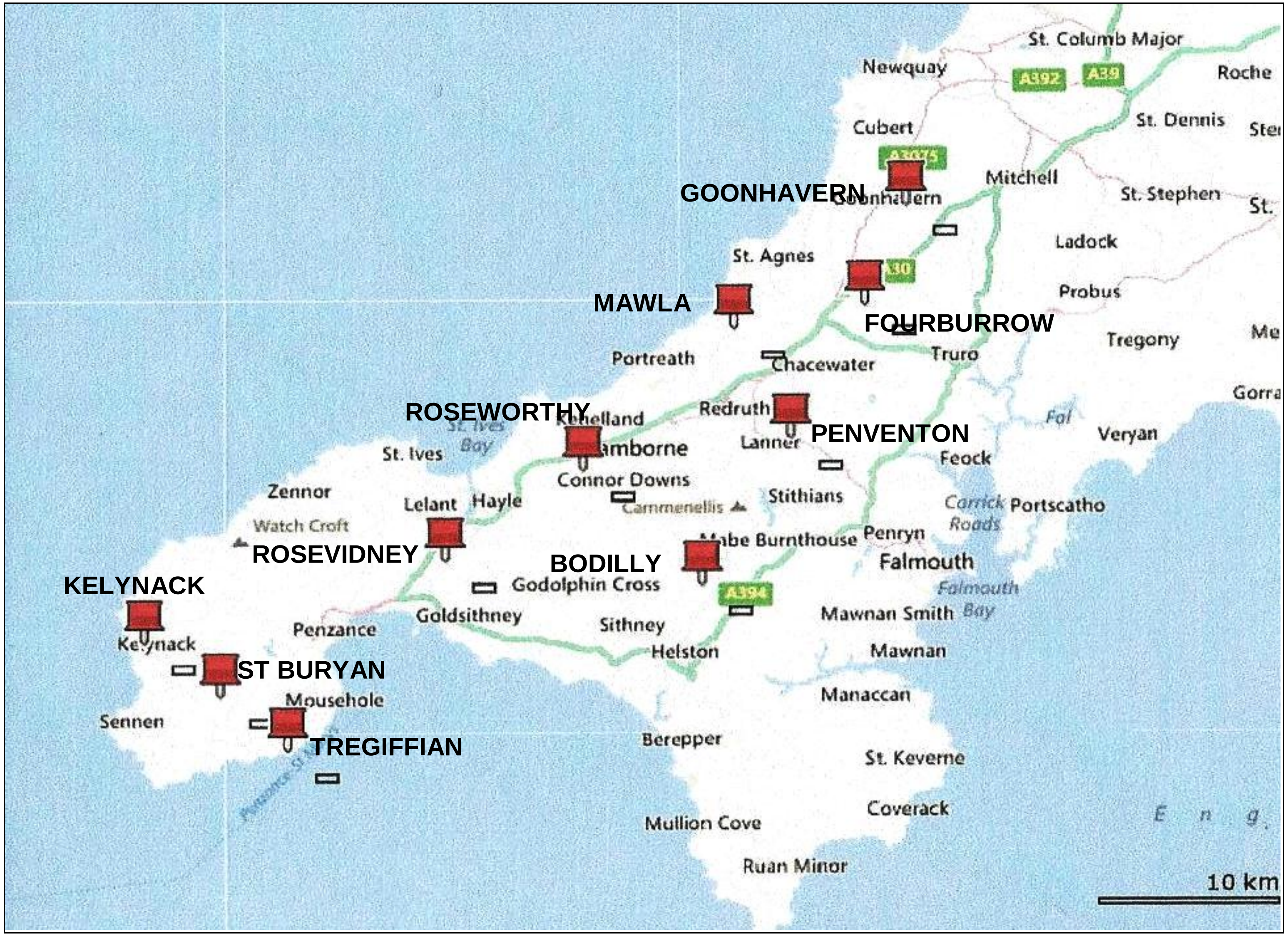


Figure 6. The locations of the ten experimental plots of 'Golden Ducat', indicated by red pins with place names in block capitals.

Figure 7. Some of the varied locations of the 'Golden Ducat' plots.

Table 4. Location, elevation and aspect of the ten sites, listed in standard order from west to east

Site reference and name	O.S. grid reference	Latitude (°)	Longitude (°)	Elevation (m)	Aspect, shelter and plot position
A Kelynack	SW 36413 29971	50.111180	-5.688128	107	In level part of S-W facing field. Close to sea (0.7km). End of outside rows E-W.
B St Buryan	SW 40491 26646	50.083135	-5.628959	121	Relatively level site, plot at lower end of field sloping gently to E. End of middle row E-W.
C Tregiffian	SW 44071 23250	50.054204	-5.576742	73	S-facing field. Near cliff edge (0.2km from sea). Large, very exposed field. Middle of outside headland rows E-W.
D Rosevidney	SW 53346 33870	50.153462	-5.454283	77	Site gently sloping S. End of outside headland rows SW-NE.
E Roseworthy	SW 61155 38796	50.200885	-5.348260	77	Site sloping gently N. Open, exposed site. End of middle rows N-S.
F Bodilly	SW 67519 31844	50.140991	-5.254984	138	Gentle slope, S-W facing. Some broken shelter (trees and buildings) to W. End of outside rows SW-NE.

G	Mawla	SW 69899 46588	50.274311	-5.230672	102	On NW-SE sloping ridge. Some shelter from S and W (hedgerow). Old mining area. End of outside rows SW-NE.
H	Penventon	SW 72769 40130	50.217433	-5.186588	91	Strong SE slope. Broken shelter (hedgerow) to NW. End of middle rows SE-NW.
I	Fourburrow	SW 77193 47636	50.286518	-5.129086	96	SW-facing steep slope. Very open site. Middle of outside rows NE-SW.
J	Goonhavern	SW 79638 53300	50.338305	-5.098118	83	W-facing slope. Some shelter (trees) except on S. End of outside rows E-W.

Husbandry

At each site the two grades of bulbs were planted in two adjacent lengths of ridge each *ca* 20m long (except at Rosevidney and Roseworthy where they were each *ca* 30m long). The inter-ridge distance varied according to the growers' usual practices and was between 0.76 and 1.06m; a typical arrangement would be planting ridges in pairs at 0.76m centres using wheelings of 1.82m, giving an inter-ridge distance between adjacent ridges across the wheelings of 1.06m. This gave a planting density of *ca* 14t/ha with 20m-long plots (or *ca* 9t/ha with 30m-long plots). Unavoidably, due to the prolonged and exceptionally wet autumn in 2012, some bulb planting had to be delayed until the advent of better conditions, so planting dates ranged from 12 September to 5 November 2012 (Table 6).

Bulb planting and subsequent husbandry at the sites followed each grower's usual practice. However, it was requested that flowers were not picked but left *in situ* to allow the full development of any daffodil rust symptoms for assessment, so the trial plots were marked (e.g. with high-visibility tape) to emphasise this requirement. (This was broadly successful, though a better protocol might be to arrange for the plots to be surrounded by varieties that flower substantially earlier or later than 'Golden Ducat'.) The markers also served to draw tractor drivers' attention to the location of the plots and their monitoring stations (see below), though at Kelynack one-half of one ridge, and at Mawla *ca* 1m at one end of each ridge, were damaged by vehicles 'cutting corners' (these damaged portions were not assessed and counts from the remaining plot were scaled-up to take account of these losses). At St Buryan one end of each ridge appeared to be planted with a different bulb stock and these areas too were excluded from assessments. Each grower was asked to provide details of sprays applied and other field operations, information which will be collated for the Final Report.

Environmental monitoring

After planting the bulbs an 'Active Irrigation Scheduling' monitoring station, with added air temperature and humidity sensors, was set up in the plot at each site by Plantsystems[2] who continued to monitor and maintain them. Each station included a sensor to measure percentage soil water content (SWC) at 0-100, 100-200 and 200-300mm depth, soil temperature at 150mm depth, rainfall, air temperature and relative humidity (Figure 8). The soil sensors were inserted in the ridge centre, while air temperature and humidity sensors were positioned 200 to 300cm above the ridge tops, roughly corresponding to mid-canopy height for the fully-grown crops. The measure 'percentage SWC' is equivalent to the amount of water in mm per 100mm depth (1% = 1mm of water in 100mm of soil), and could also be expressed as the average across the three depths (i.e. mm of water in 100mm of soil) or the total of the three depths (i.e. mm of water in 300mm of soil). All measurements were logged at 15-minute intervals, accumulated on Plantsystems' data-base, and checked and downloaded at appropriate intervals for analysis.

Weather and soil data for the period 1 November 2012 to 31 March 2013 are summarised in Figure 9 to Figure 13.

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[2] Plantsystems Ltd (now Agrovista UK Ltd trading as Plantsystems); http://www.plantsystems.co.uk/

Figure 8. Plantsystems (Agrovista) ‘Active Irrigation Scheduling’ monitoring station on site.

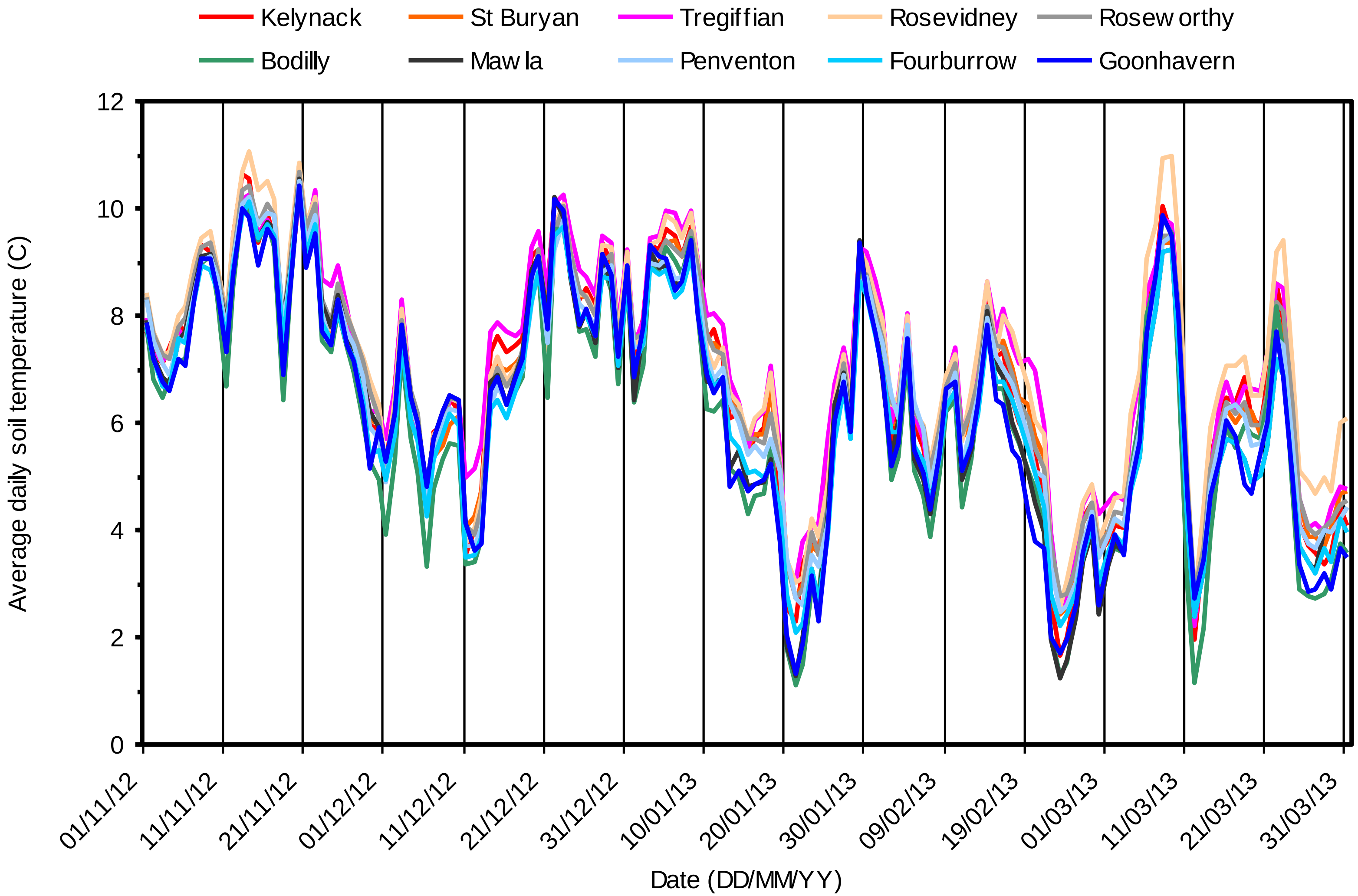


Figure 9. Average daily soil temperature for 1 November 2012 to 31 March 2013.

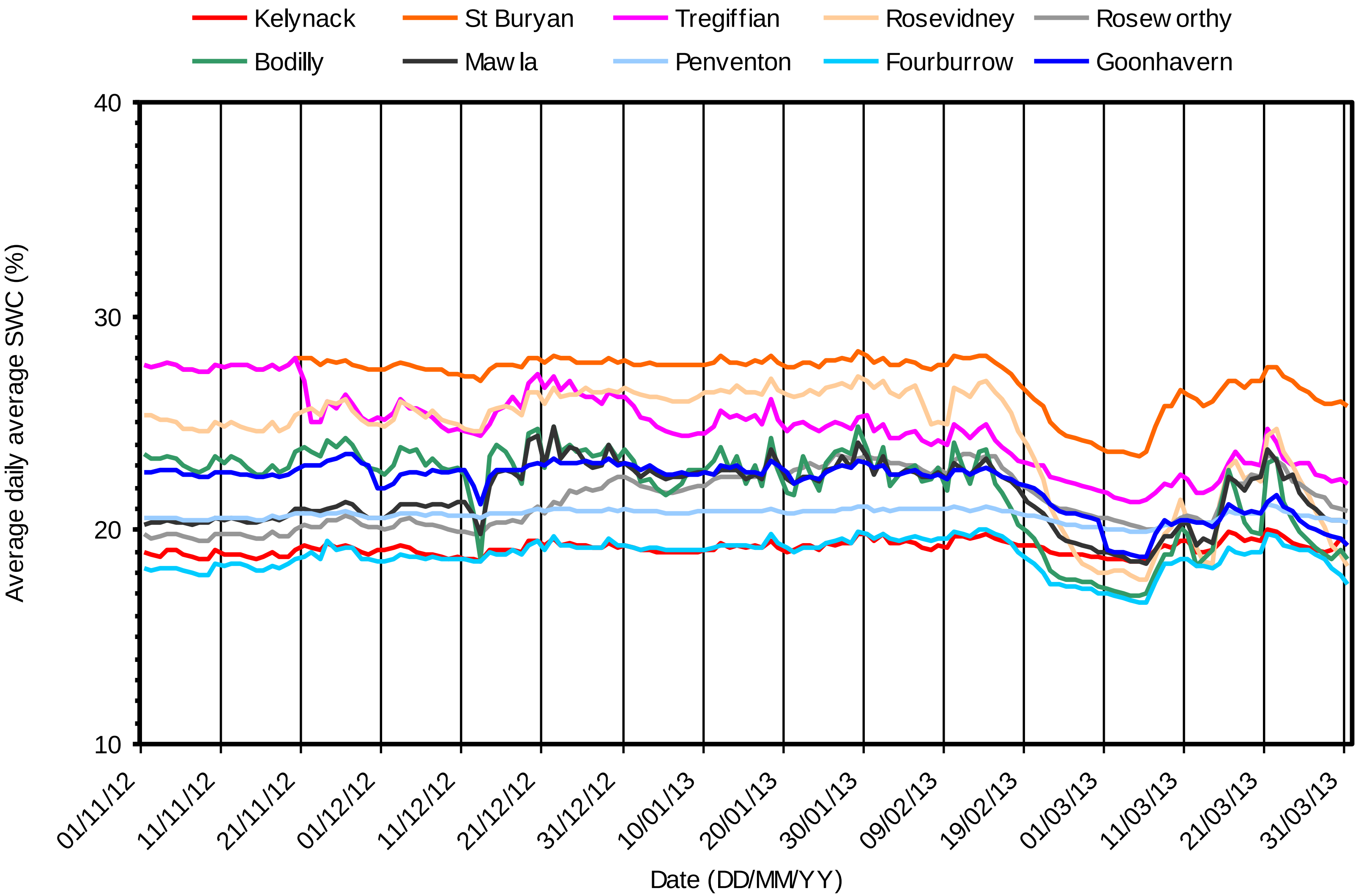


Figure 10. Average daily average SWC for 1 November 2012 to 31 March 2013.

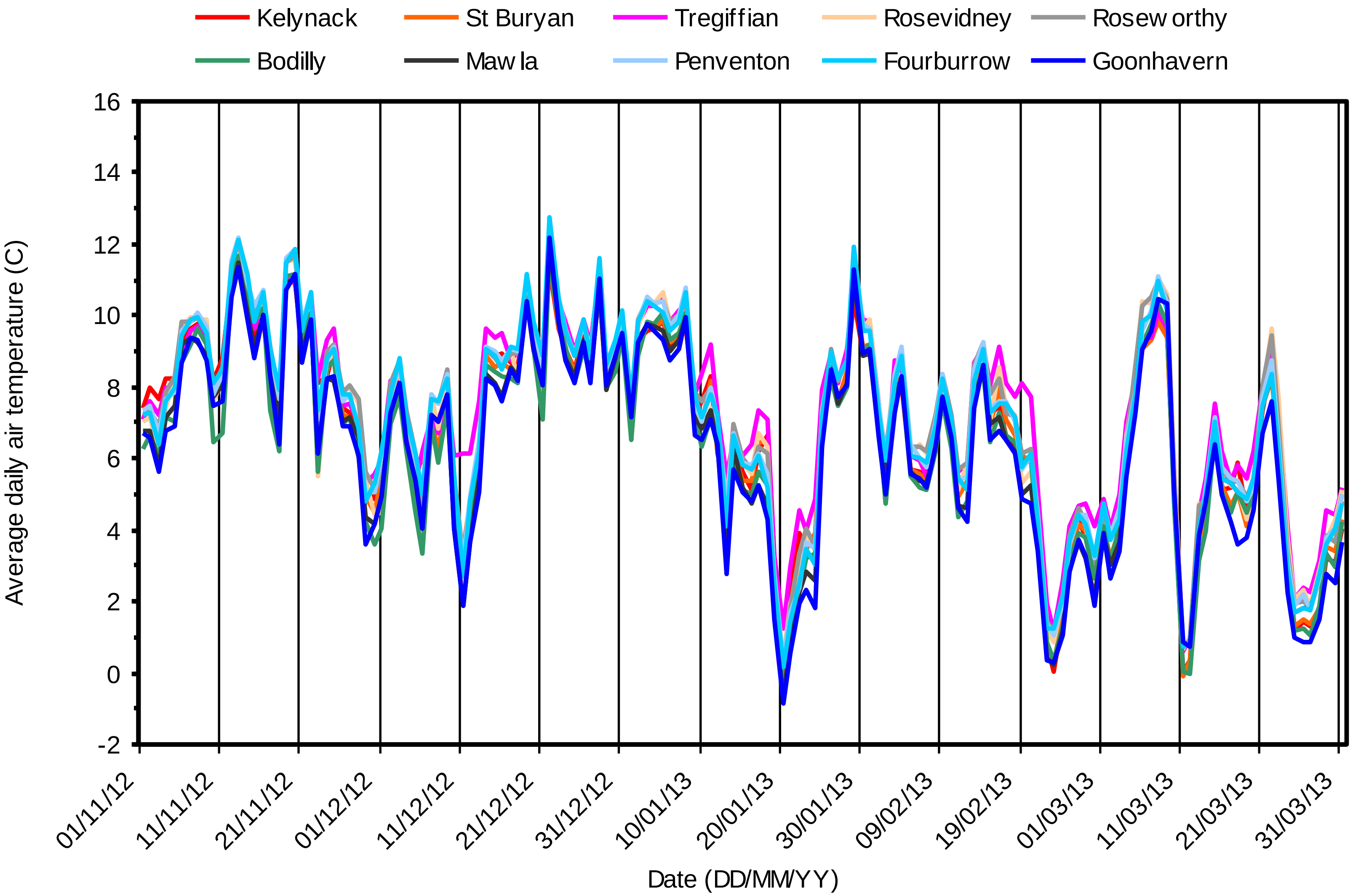


Figure 11. Average daily air temperature for 1 November 2012 to 31 March 2013

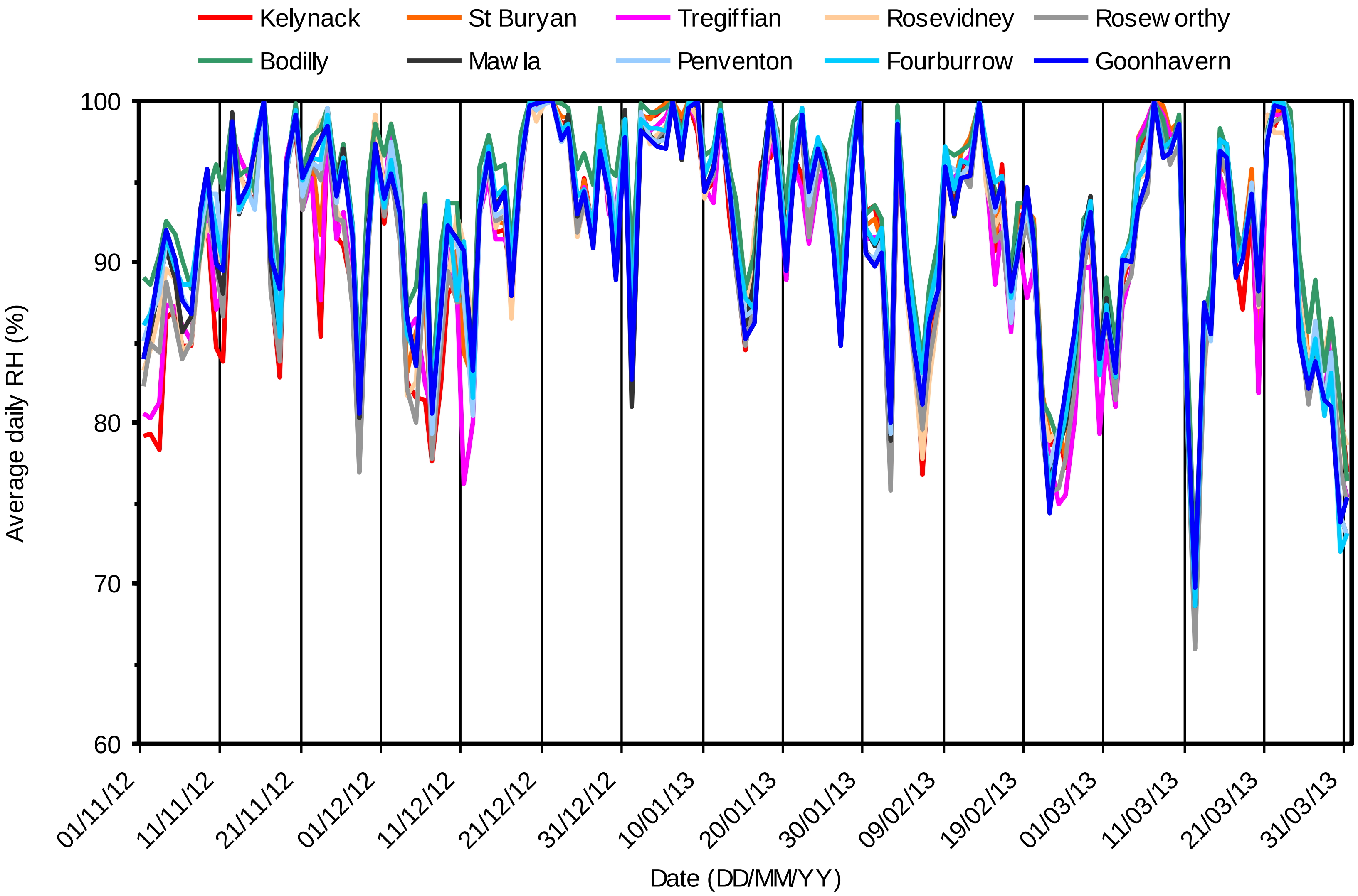


Figure 12. Average daily RH for 1 November 2012 to 31 March 2013.

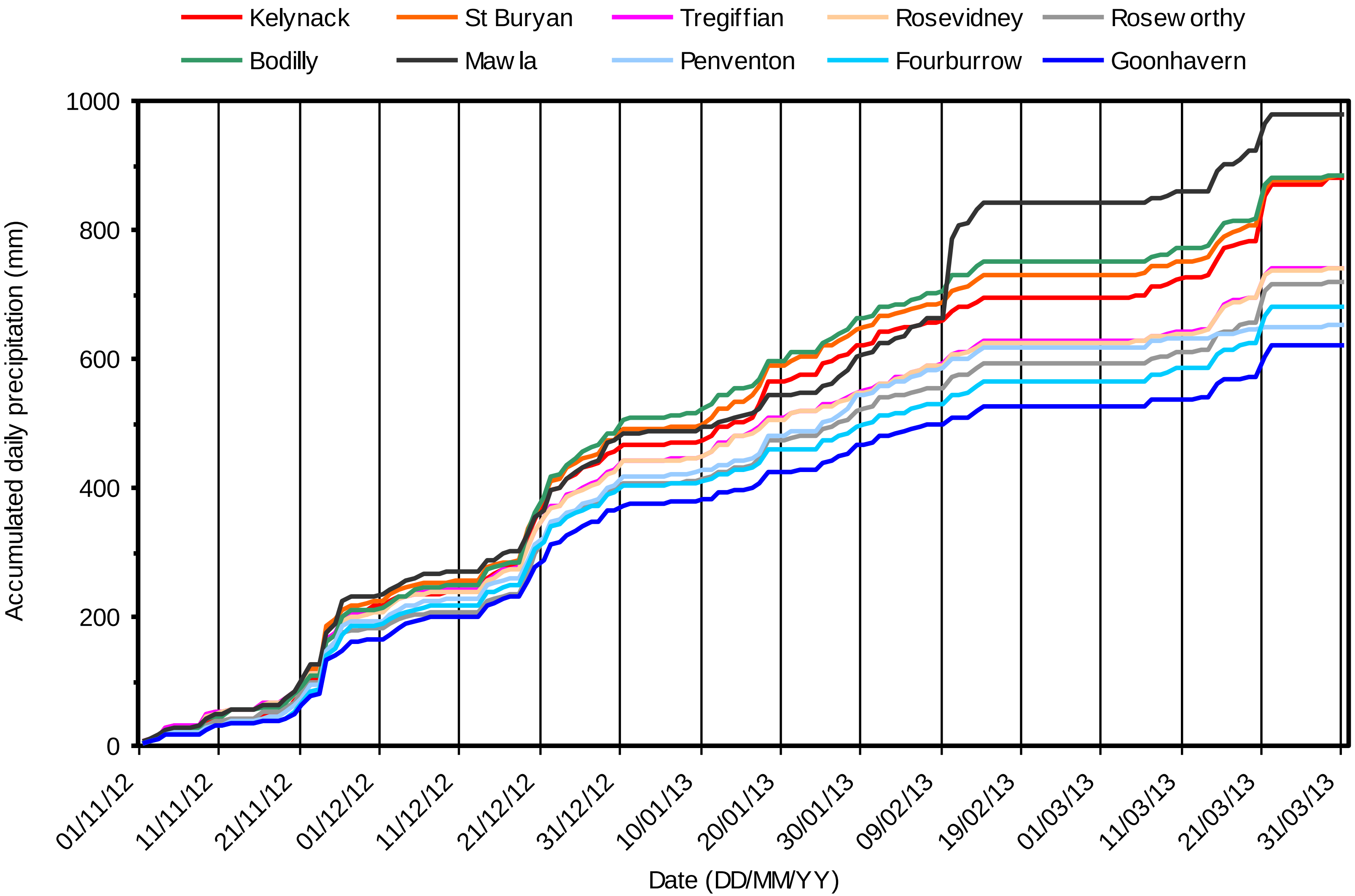


Figure 13. Accumulated daily precipitation for 1 November 2012 to 31 March 2013.

Soil sampling

In autumn 2012 (25 to 27 October) the soil in each of the ten plots was examined and samples taken for analysis. Soil sampling and examination techniques were guided by the standard text 'RB209' (Defra and Welsh Assembly Government, 2010). For consistency, regular samples were taken from half-way up the ridges rather than from the furrows.

- Using a 'cheese corer' (2.5cm diameter) a soil sample 0 to 20cm deep was taken by bulking at least ten cores from across a whole plot. Samples were analysed for the concentrations of major nutrients (nitrate-N, P, K and Mg) at WCC.
- Using a 90cm soil auger (2.5cm diameter), soil samples were taken from 0 to 30, 30 to 60 and (as far as practical) 60 to 90cm deep. The maximum practical auger depth was 45 to 50cm except at Roseworthy where it was 60cm. Soil from each layer was examined. A bulked sample of three to five 0 to 30cm cores were subjected to mechanical (particle size) analysis by Anglian Soil Analysis Ltd, and the results were used to define the soil texture at each site.

In addition a 15cm-thick soil block was removed to full spade-depth, placed on a tray and each principal layer was examined and described (a) by assessment of soil texture by hand (following 'RB209') and (b) using a 'Visual Soil Structure Quality Assessment' (VSSQA) which defines soil structure as an easily quantifiable score from 1 (friable) to 5 (very compact) (Ball *et al*., undated).

In spring 2013 (26 to 28 March), after the bulk of the winter rain and well after any fertilisers had been applied, further soil samples were taken at each site using the methods described above.

- Samples 0 to 20cm-deep were analysed for pH and the concentrations of macro-nutrients (N, P, K and Mg) and micro-nutrients (S, Ca, Na, Fe, Mn, Cu, Zn, B, Mo and Al) by WCC.
- Samples were taken from as deep as practical (to 45cm at Fourburrow, Roseworthy, Mawla and Goonhavern and to 50cm at the other sites) and analysed for total mineral N by WCC.

Soil and plant sampling and disease diagnostics

The project calls for further sampling across all plots when commercial levels of daffodil rust are apparent on some plots. Since only low levels of daffodil rust were present on the plots in 2013, these samples will be taken in 2014.

- Soil and leaf samples will be taken for full nutrient analysis to be carried out at WCC.
- Stem samples will be taken for disease diagnostics at a commercial plant clinic.

- Small stem tissue samples will be taken, frozen in liquid nitrogen, and stored at -70°C at WCC for the determination of viral RNA.

Table 5. Soil chemical analysis for August 2012 provided by growers.

Site reference and name		pH	Nutrient index		
			P	K	Mg
A	Kelynack	6.3	4	2+	2
B	St Buryan	5.5	3	2+	3
C	Tregiffian	6.2	3	1	3
D	Rosevidney	6.6	3	2-	2
E	Roseworthy	na [2]	na	na	na
F	Bodilly	5.9	3	1	2
G	Mawla	7.3	4	3	2
H	Penventon	- [1]	-	-	-
I	Fourburrow	na	na	na	na
J	Goonhavern	5.4	0	1	2

[1] Soil analysis not done [2] Not available

Table 6. Previous cropping, and fertilizer and lime application for the ten sites.

Site reference and name		Last crop and previous brassicas or grass (if any)	History of organic fertiliser	Fertiliser applied in 2012	Lime applied in 2012	Planting date (2012)
A	Kelynack	1-year ley, silage taken Previously brassicas	No	300kg/ha sulphate of potash	Nil	25 Sep
B	St Buryan	Potatoes Previously brassica	No	450kg/ha 0:11:34	7.2t/ha	5 Oct
C	Tregiffian	Long-term pasture	No	450kg/ha 0:11:34	3.5t/ha	25 Sep
D	Rosevidney	Spring barley Previously brassicas	No	450kg/ha 0:11:34	Nil	20 Oct
E	Roseworthy	Brassicas for last 3 years	No	500kg/ha 5:10:30 [1]	na	12 Sep
F	Bodilly	Barley Previously brassicas	No	500kg/ha 0:11:34	4.8t/ha	5 Nov
G	Mawla	Barley Previously brassicas	No	450kg/ha 0:11:34	Nil	5 Nov

H	Penventon	3 to 4-year ley, mainly grazed, some silage taken	FYM, cattle	Nil	Nil	28 Sep
I	Fourburrow	Winter wheat	No	600kg/ha 0:18:36	na	17 Sep
J	Goonhavern	Brassicas	No	400kg/ha 0:11:34	9.1t/ha	1 Nov

[1] The N applied at this site was unlikely to have been necessary

Crop and rust assessments

Plots were assessed as described below at three stages of growth each year, early stem extension, around picking and post-cropping. In 2013 the assessment dates were 7 to 8 February, 27 February to 1 March, and 26 to 28 March.

- The minimum, most usual and maximum crop growth stages (GS,

- Table **8**) were recorded, including (where applicable) stem and foliage heights.
- All emerged stems were checked individually on both surfaces for symptoms of daffodil rust (see Figure 14). Where present its incidence and severity were scored on a standard 0 to 5 scale (Table 7) and the numbers of stems per plot with daffodil rust were recorded. Although the numbers of stems affected was not a strictly quantitative measure of incidence, *ca* 1000 bulbs were planted at each site and it was felt that using this measure was a clearer way of seeing differences in incidence. The stems were also checked for any signs of early daffodil rust lesions.
- Foliage was checked for daffodil rust symptoms (clump-by-clump rather than by individual leaves) and similarly scored, if appropriate.
- The general occurrence of symptoms of pests, diseases and other disorders and damage were noted.
- Similar observations will be made over the second (and preferably third) years of the crops. If appropriate, the root profile of fully-grown crops will be examined.

Table 7. Daffodil rust severity and incidence scores used in plot assessment (note: the severity scores are modifications of those given in Table 1 and Table 2).

Severity	**Scores**	**Incidence**
None seen	0	None seen
Slight markings that may not be rust-coloured, or typical spots but restricted to one or two small spots only ("not really rust"); almost unnoticeable	1	Up to 1% of stems affected
Sparse but typical lesions; no commercial significance (but worth watching)	2	Up to 5% of stems affected
Moderate lesions that are becoming disfiguring; commercially might lead to down-grading	3	Up to 10% of stems affected
Severe daffodil rust, some cracks, very disfiguring; flowers un-marketable	4	Up to 50% of stems affected
Very severe daffodil rust with very obvious cracking and stem bending; flowers un-marketable	5	Up to 100% of stems affected

Table 8. A scale of growth stages (GS) for daffodils.[1]

Period	GS	Description	Notes
Unplanted bulb (GS 0)	0.1	'Dormant' bulb in storage	Bulbs would normally be planted at GS 0.1 or 0.2
	0.2	Root initial development evident close to the surface of the bulb	
	0.3	Shoot and/or roots emerging from stored bulb	Apply only to stored bulbs
	0.4	Bulb becoming desiccated with loss of skin, emerging roots or shoots becoming moribund	
	0.5	Bulb shrivelled, light in weight, or rotted	
Planted bulb (GS 1)	1.1	No clear emergence of shoot and/or roots	
	1.2	Roots and/or shoot emerging, <1cm in length	
	1.3	Roots and shoot elongating	
	1.4	Shoot tip close to soil surface	
Emergence (GS 2)	2.1	First shoots starting to emerge	Foliage height nominally 0
	2.2	Shoots elongating, but no buds obviously visible	Record foliage height (and stem height for 2.3 and 2.4) [2]
	2.3	Shoots elongating, tips of flower buds visible without pulling shoots apart	
	2.4	Full length of buds visible ('upright pencils')	
Anthesis (GS 3) [3]	3.1	Flower buds still 'upright pencils' with no colour showing, but becoming clear of the foliage; flower cropping could have begun if a very tight stage is required and stem length is adequate	Record foliage and stem heights
	3.2	Flower buds are 'fat pencils' with no colour showing, flower cropping should have begun	Record stem height
	3.3	Pedicels bending and/or spathes splitting, colour may be showing; a very late picking stage	
	3.4	Pedicels fully 'goose-necked' but flowers not open	This stage may pass quickly and variably

	3.5	Flowers (or florets) starting to open	
	3.6	Flowers fully open	For multi-headed types, 50% of florets open, senescing or senescent
	3.7	Flowers at least starting to senesce (petal tips dying) but not fully senescent	
	3.8	Flowers (or florets) fully senescent, leaves still fully green and upright	
Post-flowering (GS 4)	4.1	Leaves still fully green, but at least some leaves starting to bend to ground	
	4.2	As 4.1, but some leaves bending conspicuously and at least some leaves with senescent (yellowing and dying) tips	
	4.3	Most leaves almost flat, with general incidence of senescence at the leaf ends	
	4.4	Some 50% of leaf area senescent	
	4.5	Less than 10% leaf area remaining green	
	4.6	None (or a trace) of leaf area remaining green	
'Summer dormancy' (GS 5)	5.1	Small amounts of green foliage remaining attached to bulbs	
	5.2	Any foliage attached to the bulbs now dead	
	5.3	Dead foliage lost or removed	
Lifted bulb (GS 6)	6.1	Bulb surface damp and/or not cleaned	
	6.2	'First stage' drying (surface drying) complete	
	6.3	'Second stage' drying complete	
	6.4	Bulbs cleaned (and graded if appropriate)	

[1] Avoid the following when recording: plot or row ends; obvious rogues, off-types and atypically damaged/diseased plants; late flowers from lateral bulbs; and the most advanced plants if these are about 1% or less of the total.

[2] Record shoot height from the point of emergence from the soil to the uppermost tip of foliage, and stem height from the point of emergence from the soil to the topmost tip if the bud, spathe or flower.

[3] If flowers cropped and no remnants left to estimate exact GS, record as '3.C' (cropped).

Figure 14. Top: increasing rust severity with blistering (left), a few rust lesions (middle) and larger, coalescing lesions (right). Bottom: close-up of blistering (left) and rust lesions with cracking (right).

Associations between daffodil rust levels and soil, water and other factors

In preliminary data assessments in Year 1 the potential associations at each site of the severity and incidence of daffodil rust with soil water content and weather data (collected by the monitoring stations) was assessed using graphical summaries in the form of box-and-whisker plots, which provide easy visualisation of the means and ranges where there are many values. Using the data collected at 15-minute intervals for the factor and period being considered (say, air temperature during December 2012), the central thick line in the plots indicates the median value, the box covers values falling in the second and third quartiles, and the 'whiskers' extend to minimum and maximum values excluding any extreme values (or 'outliers', defined as those which fall outside a value of 1.5 × the inter-quartile range, shown on the plots by the circles). A box-and-whisker plot covering a one-month period is therefore derived from almost 3,000 data points at each site for any one factor.

In addition, simple scatter plots were used to look for potential associations at each site of the severity and incidence of daffodil rust with other data collected - soil nutrient levels, geographical and topographical factors, crop growth, etc. These plots are shown with best-fit lines and correlation coefficients (r) with their statistical significance. An association between the level of rust and, say, SWC, would be indicated by the points having a distinct pattern (e.g. tending to fall on a straight line) and a value of r approaching 1. Following convention, the symbols (*), *, ** and *** are used to indicate significance at the 0.1, 0.05 ('significant'), 0.01 ('highly significant') and 0.001 ('very highly significant') levels of probability (*p*), meaning that the result could have be expected to have occurred by chance once in 10, once in 20, once in 100 or once in 1000 instances, respectively, with NS indicating 'not significant'.

In Year 2 more formal analyses will be carried out using multiple regression, considering either linear or non-linear models (for any developmental data that can be considered as continuous responses) or logistic regression models (for data in the form of the presence or absence of daffodil rust symptoms or where the response is measured as the proportion of plants showing symptoms). Analyses will take appropriate account of the temporal nature of the data, considering the accumulated weather conditions over fixed periods prior to the observations on daffodil rust symptoms, and considering increases in symptoms between consecutive field observations. These approaches should allow the identification of the key weather and soil factors associated with daffodil rust development, and potentially to a preliminary predictive model that can be validated with further controlled experiments.

Field-work: results

Soil analysis

<u>Autumn 2012</u>

Particle size analysis (Figure 15 and Figure 16) showed that the soil at all ten sites contained <20% v/v clay but variable amounts of sand, from <10% sand at Rosevidney (classified as a sandy clay loam), 10 to 30% sand at Fourburrow, Mawla and Roseworthy (silt loams) and 30 to 50% sand at the remaining six sites (sandy silt loams). The soil textures quoted by the growers were also recorded (Table 9), but these differed somewhat and may not have been derived through using standard methods.

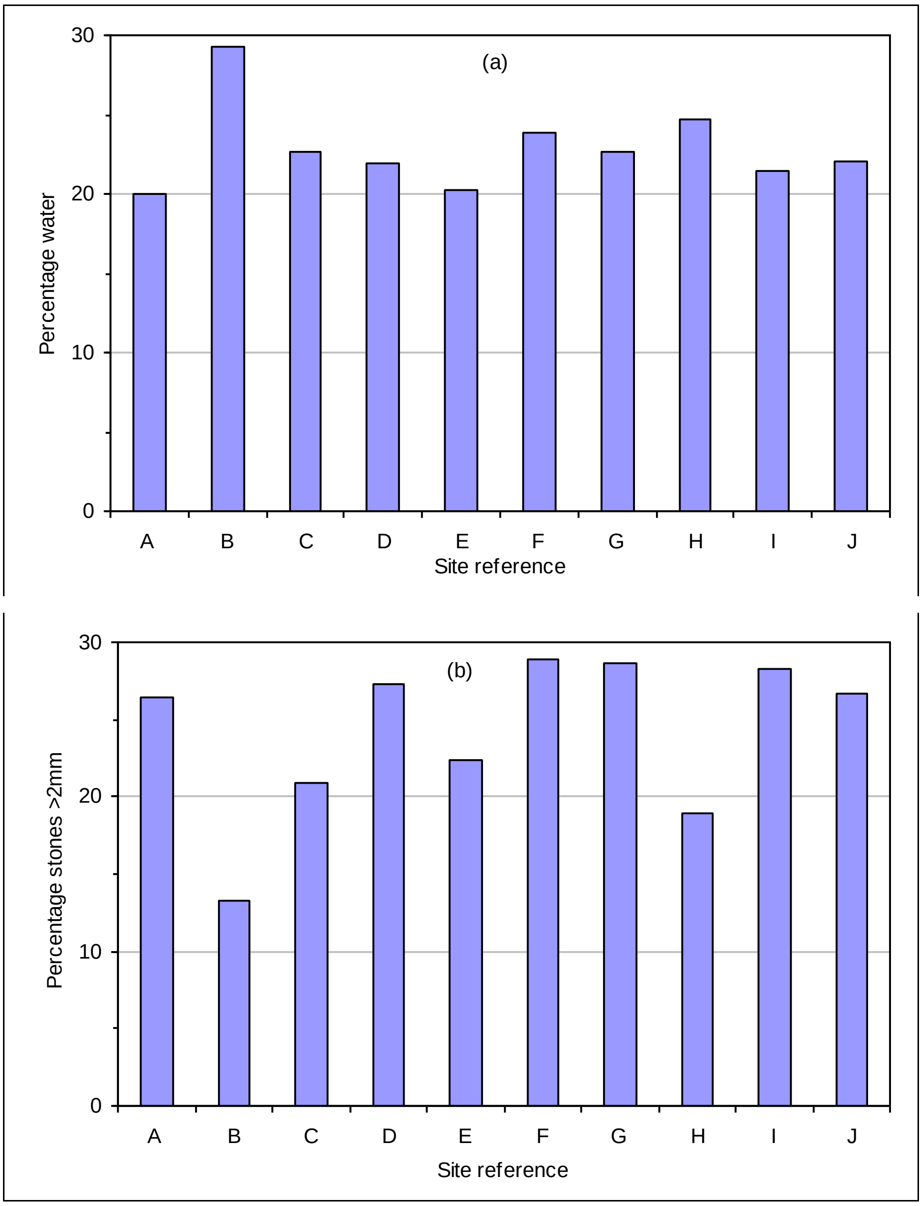
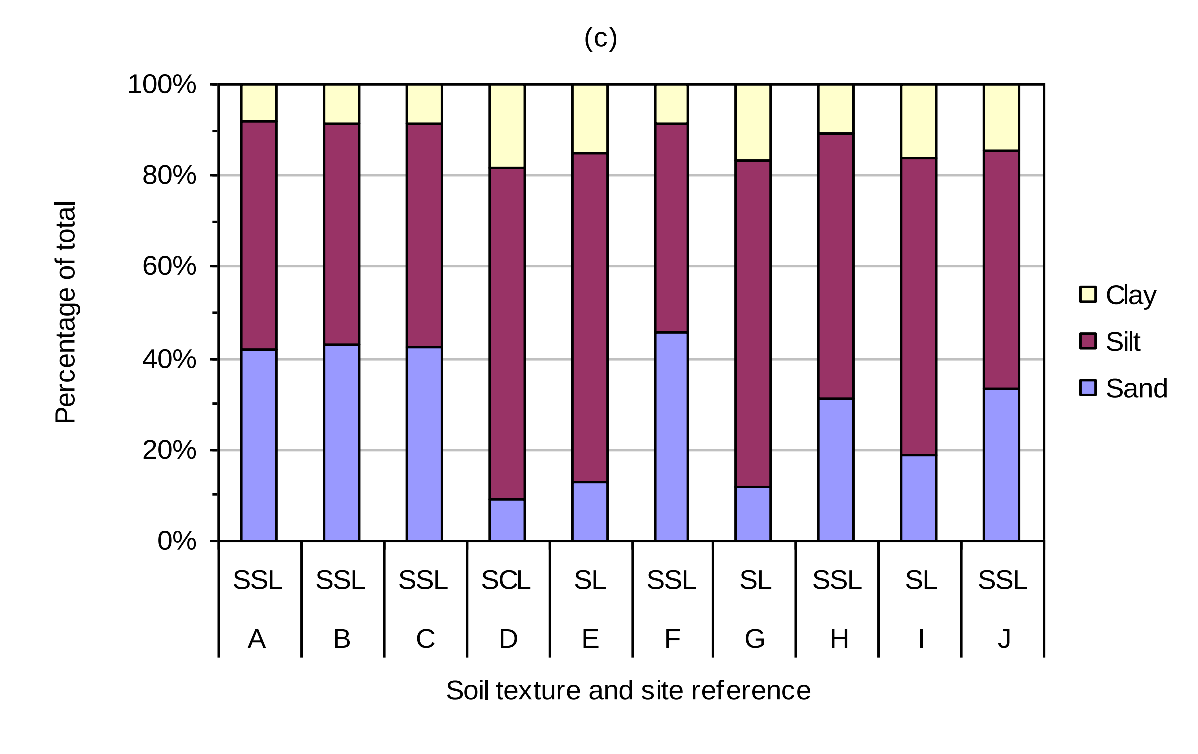


Figure 15. Soil particle size analysis to 30cm for the ten sites: (a) soil percentage water (w/w); (b) stones >2mm as percentage of dry soil. Data continued in Figure 16.



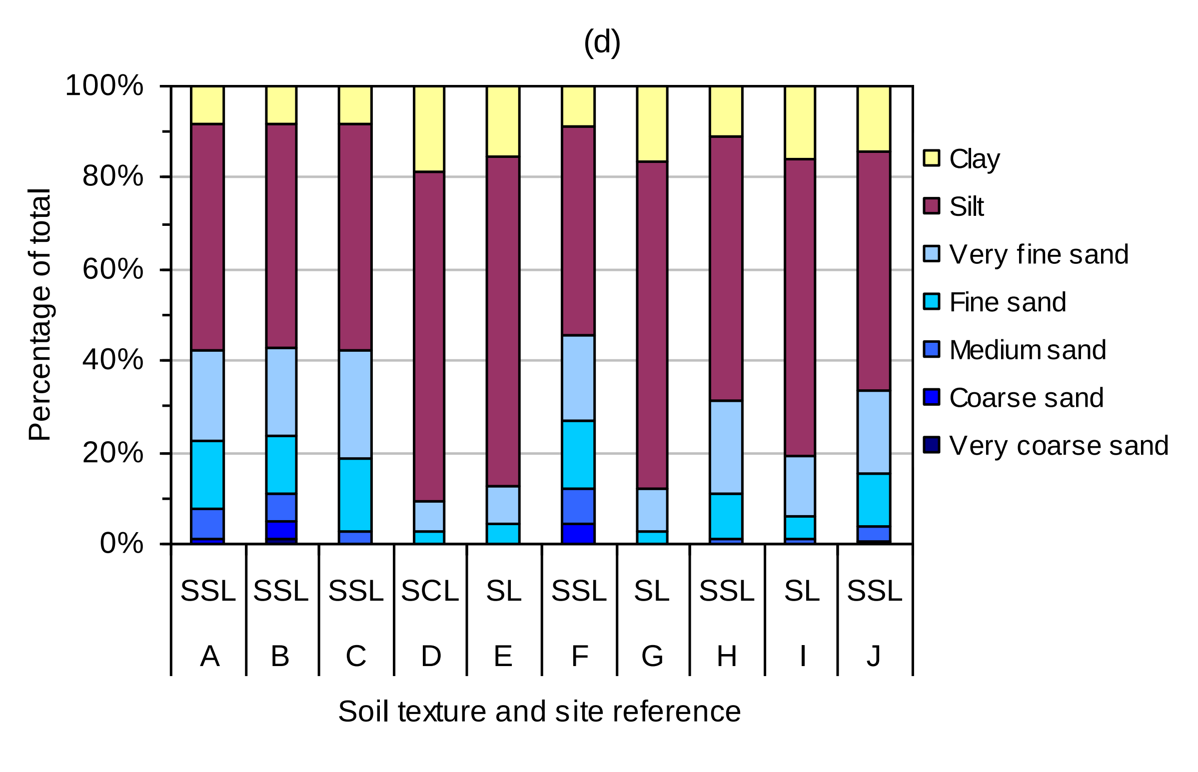


Figure 16. Soil particle size analysis to 30cm for the ten sites (data continued from Figure 15): (c) clay, silt and sand as percentage of dry soil after stones >2mm removed; (d) as (c) but including the

breakdown to grades of sand. The soil textures indicated by the particle size analysis are also shown.

Soil texture was also assessed using a 'Visual Soil Structure Quality Assessment' (VSSQA) which produces an easily quantifiable score from 1 ('friable') through 'intact', 'firm' and 'compact' to 5 (very compact) (Table 9). Most sites scored 1.0 to 1.5 (friable or friable/intact), with Goonhavern scoring 2.0 (intact) and Rosevidney 2.5 (intact/firm).
Examination of the soil horizons showed that most sites had a relatively shallow, well worked upper layer 20 to 30cm-deep (Figure 17 and Table 9). Bodilly and Rosevidney had a shallow upper layer (12 to 15cm-deep), while Roseworthy had a uniform upper layer almost 60cm deep. Other than at Roseworthy, the soil then became progressively more compact and clayey, sandy, gritty or stony. Below 40 to 50cm the soil was difficult to penetrate, except at Roseworthy where the hard layer did not start until about 60cm. The surface contained gravel, stones or rock chips at all sites, and liberally so at Tregiffian, Rosevidney, Roseworthy, Mawla and Fourburrow. The soil appeared particularly prone to capping at St Buryan. All sites appeared to drain well down slopes, though at St Buryan and Rosevidney water tended to stand in the furrows of the plots, these being sited in lower parts of the field.

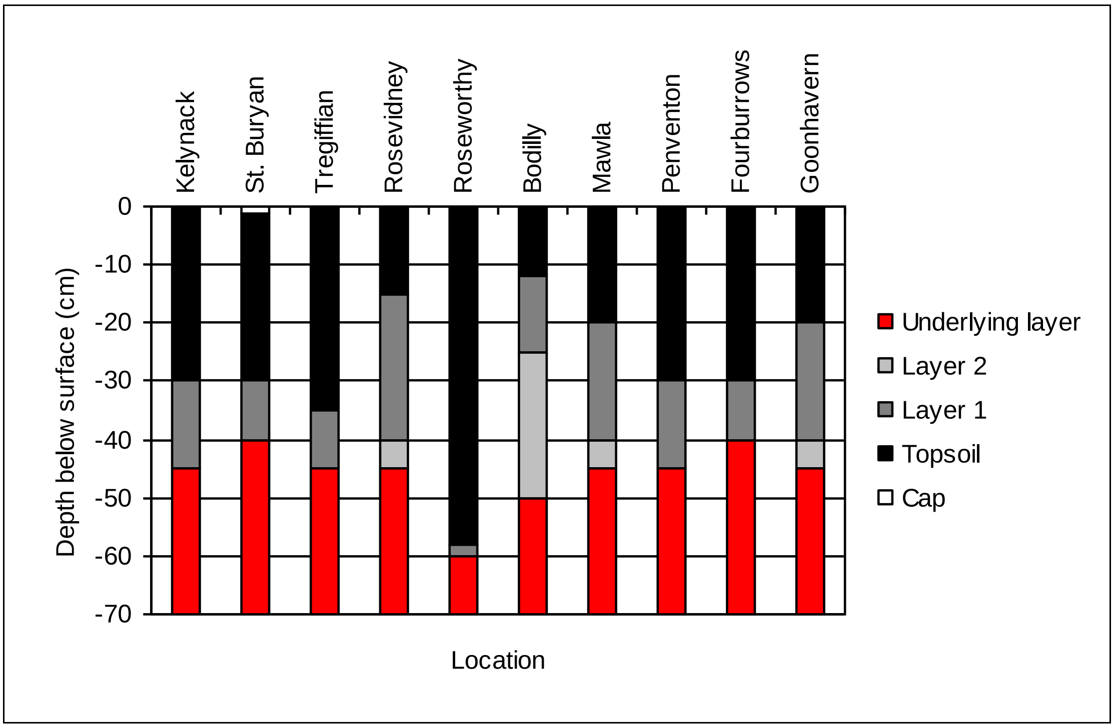


Figure 17. Soil horizons at the ten test sites.

The soil associations at most sites were based on typical brown earths as used for horticultural crops in west Cornwall (Table 9). However, at St Buryan, Tregiffian and Bodilly there were typically dark brown podzolic soils resulting from acidic weathering, also used for horticultural crops in west Cornwall. At Kelynack the soil was similar, but with a humose or peaty topsoil, a soil not usually used for horticultural crops.

Table 9. Soil texture assessments and soil descriptions (assessed October 2012), drainage (assessed February 2014) and soil associations for the ten sites.

Site reference and name		Soil texture [1]	Soil texture [2]	VSS QA [3]	Soil surface state, horizons (see Figure 17) and drainage	Soil association [4]
A	Kelynack	Sandy silt loam	Loamy fine sand	1.00	Surface: many stones and rock chips up to 15cm across, occasionally more. 0-25cm: uniform, friable dark brown soil with small stones 25-45cm: becoming increasingly compacted, lighter in colour, clayey >45cm: stony layer not easily penetrable Draining well except in wheelings where compacted	Moor Gate (612b)
B	St Buryan	Sandy silt loam	Medium sandy loam	1.25	Surface: many stones and rock chips up to 15cm across, occasionally more; extensive effect of precipitation with fines washed into furrows, now capped. 0-30 to 0-50cm: uniform, friable dark brown soil with small stones, becoming increasingly compacted >30 to >50cm: yellow, sandy and stonier >50cm: stony, not easily penetrable Standing water in some furrows including those of the plot	Morton-hampstead (611b)
C	Tregiffian	Sandy silt loam	Medium sandy loam	1.00	Surface: liberally strewn with quartz or blue-grey angular stones up to 20cm across; windward sire of ridges dried out. 0-25 to 0-40cm: uniform, friable dark brown soil with small stones >25 to >40cm: clayey, stonier but still more or less friable >45cm: stony, not easily penetrable	Morton-hampstead (611b)

					Furrows can flood with heavy rain but drain well down steep slope	
D	Rosevidney	Sandy clay loam	Silty clay loam	2.50	Surface: liberally strewn with cobbles up to 2.5cm across, occasionally to 15cm. 0-15cm: uniformly dark brown with small stones, friable 15-40cm: clayey with small stones 40-45cm: hard, sandy layer >45cm: not easily penetrable Furrows can flood with heavy rain but drains down, only furrows at the lower end of field (including plot) with standing water.	Denbigh 2 (541k)
E	Roseworthy	Silt loam	Medium sandy loam	1.00	Surface: liberally strewn with quartz or blue-grey angular stones up to 20cm across. 0-58cm: soil uniform, brown 58-60cm: soil lighter in colour, gritty >60cm: stony layer not easily penetrable Much fines washed into furrows, drains reasonably well but some standing water in furrows.	Trusham (541n)
F	Bodilly	Sandy silt loam	Medium sandy loam	1.00	Surface: much gravel evident on surface, and strewn with quartz or brown/pink stones up to 10cm across 0-12cm: uniform, brown, friable soil 12-25cm: sticky clay 25-50cm: clay with sand and grit >50cm: sandy or gritty layer not easily penetrated Patches of standing water in furrows.	Morton-hampstead (611b)
G	Mawla	Silt	Silty	1.50	Surface: liberally strewn with quartz or blue-grey stones or chips mostly	Denbigh 2

		loam	clay loam		up to 15cm across. 0-20cm: soil uniform, dark brown and friable 20-40cm: soil becoming increasingly clayey and compacted 40-45cm: brick-red compacted layer >45cm: not easily penetrable Water appears to drain down slope well, but still some standing water in furrows.	(541k)
H	Penventon	Sandy silt loam	Silt loam	1.00	Surface: some small stones to 3cm (rarely 10cm) across. Ridge tops gravely and draining. 0-30cm: uniform dark-brown soil 30-45cm: increasingly stony and clayey >45cm: stony or sandy layer not easily penetrable Water drains down slope well, but still some standing water in furrows in places.	Denbigh 2 (541k)
I	Fourburrow	Silt loam	Silty clay loam	1.00	Surface: very liberally strewn with quartz or blue-grey angular stones up to 15cm across. 0-30cm: soil uniform, light brown 30-40cm: soil becoming increasingly clayey and pink-coloured >40cm: stony layer not easily penetrable Well drained down steep slope.	Denbigh 2 (541k)
J	Goonhavern	Sandy silt loam	Silty clay loam	2.00	Surface: strewn with quartz or blue-grey stones or chips up to 15cm across; ridge top with many cobbles up to 1cm across and clods up to 10cm across.	Denbigh 2 (541k)

	0-20cm: loose brown soil with cobbles and small stones 20-40cm: soil becoming increasingly clayey and compacted 40-45cm: sandy compacted layer >45cm: not easily penetrable Water drains down well but some water standing at lower end of field.

[1] Soil texture (0-30cm) as determined by particle size analysis (see Figure 16)

[2] Soil texture as provided by growers

[3] ‘Visual Soil Structure Quality Assessment’ (Sq1 friable, Sq2 intact, Sq3 firm, Sq4 compact and Sq5 very compact) (Ball, Batey & Munkholm, undated)

[4] Soil association and map symbols (Soil Survey of England and Wales, 1983) as follows:

541: typical brown earths – non-alluvial loamy soils with a non-calcareous subsoil without significant clay enrichment. The 541k subgroup (the Denbigh 2 soil association) represents well drained, fine loamy soils over slate or slate rubble, typical of early potato and broccoli growing in west Cornwall, while 541n (Trusham) are also well drained, fine loamy soils, but over deeply weathered rock, some shallow and some deeply sloping and with bare rock locally and typically used for “horticultural crops in drier districts.”

611: typical dark brown podzolic soils resulting from pedogenic accumulation of iron and aluminium or organic matter or some combination of these, normally formed as a result of acidic weathering conditions and, under natural or semi-natural vegetation, having an unincorporated acidic layer at the surface. The 611b subgroup (Moretonhampstead) represents well drained, gritty loamy soils with a humose surface horizon in places, some with steep slopes and with boulders and rocks locally, some used for early potato and broccoli growing in west Cornwall.

612: humic podzolic soils, as 611 but with a humose or peaty topsoil. The 612b (Moor Gate) subgroup represents well drained, humose gritty loamy soils, occasionally with a thin iron-pan, many with steep slopes, often with boulders or rocky, and not traditionally used for horticultural crops.

The results of macro-nutrient analysis are shown in Table 10 and are summarised here.

- pH values varied from slightly acid (pH 6.3 at Fourburrow and Penventon) to 7.7 (at Roseworthy, indicating a trend to over-liming or a naturally lime-rich soil), with the remainder neutral at 7.0±0.4, all values suitable for growing daffodils.
- Nitrate-N levels varied from 6mg/kg of dry soil (Goonhavern) to 40 to 606mg/kg (Bodilly, Tregiffian, St Buryan and Kelynack). The additional N applied at Roseworthy in 2012 (Table 6) did not result in a high nitrate-N level in this analysis.
- P levels were adequate at all sites, varying from about 60mg/L (Fourburrow and Goonhavern) to >160+mg/L (Penventon, St Buryan and Kelynack).
- K levels varied from 117mg/L (Penventon) to 233mg/L (St Buryan), indicating a requirement for potash at some sites.
- Mg levels were all adequate, varying from 56mg/L (Fourburrow) to 233mg/L (St Buryan).

Table 10. Soil chemical analysis (0-20cm) in October 2012 for the ten sites.

Site reference and name		**pH**	**NO_3-N (mg/kg)**	**P (mg/L)**	**K (mg/L)**	**Mg (mg/L)**
A	Kelynack	7.4	42.5	164.8	212.8	133.1
B	St Buryan	6.7	41.7	163.6	460.8	233.0
C	Tregiffian	7.0	50.7	128.0	171.2	143.6
D	Rosevidney	6.8	14.8	102.4	442.5	98.5
E	Roseworthy	7.7	17.7	115.2	317.9	102.1
F	Bodilly	7.4	55.5	148.5	171.2	155.6
G	Mawla	7.3	23.7	82.4	314.9	100.9
H	Penventon	6.3	20.0	166.8	117.8	112.8
I	Fourburrow	6.3	13.5	56.5	274.4	57.5
j	Goonhavern	6.8	5.9	57.3	164.4	117.9

<u>Spring 2013</u>

The results of soil analysis are shown in Table 11 and are summarised here.

- Soil pH remained lowest at Fourburrow and Penventon (5.9 to 6.1) but the high pH at Roseworthy had moderated (to 7.4); the other sites remained at 7.0±0.7.
- Nitrate-N varied from 7.5ppm (at Fourburrow) to 17.0ppm (at Bodilly); ammonium-N varied between 4.6 or 4.7 (Fourburrow and Rosevidney) and 6.7 (Mawla).
- P remained low at Fourburrow and Goonhavern (22 to 23ppm), increasing to 62ppm at Penventon.
- K varied between 105 (Penventon) and 401ppm (Rosevidney).
- Mg varied from 58 (Fourburrow) to 245ppm (St Buryan).

- Al varied from 14 (Rosevidney) to 35ppm (Bodilly).
- Ca varied from 1709 (Fourburrow) to 4134ppm (Roseworthy).
- Fe varied from 1 (Fourburrow) to 3ppm (Bodilly).
- Mn varied from 0.5 (Roseworthy) to 4.8ppm (Fourburrow).
- Na varied from 771 (Mawla) to 1107ppm (Bodilly).

Table 11. Soil chemical analysis (0-20cm) in March 2013 for the ten sites.

Site reference and name	pH	Total mineral N (ppm)		Nutrient concentration (ppm)							
		NO_3-N	NH_4-N	P	K	Mg	Al	Ca	Fe	Mn	Na
A Kelynack	7.0	10.6	6.0	58.3	199.9	150.9	21.3	4116	1.4	0.5	920
B St Buryan	6.6	11.0	5.5	50.9	384.5	245.4	16.4	4090	1.9	1.1	866
C Tregiffian	6.8	16.7	6.0	38.3	185.8	148.9	16.7	3101	2.9	1.4	922
D Rosevidney	6.7	12.5	4.6	49.1	400.7	104.3	14.3	3011	1.1	3.5	976
E Roseworthy	7.4	10.8	6.4	52.5	302.5	106.9	15.1	4134	2.9	0.5	985
F Bodilly	7.0	17.0	5.9	48.6	170.4	140.6	35.2	4097	3.8	1.6	1107
G Mawla	7.3	11.3	6.7	41.5	329.7	78.3	17.8	3911	1.4	0.9	772
H Penventon	6.1	9.0	6.2	62.3	104.7	112.7	20.1	2803	1.6	5.1	780
I Fourburrow	5.9	7.5	4.7	23.2	208.5	58.4	24.5	1709	1.0	5.8	887
J Goonhavern	6.9	11.7	6.2	22.2	176.1	119.9	19.6	2898	1.2	2.1	799

Crop and daffodil rust assessments

<u>Spring 2013</u>

Early stem extension stage The first crop and daffodil rust assessment was carried out on 7 to 8 February 2013 when plants at most sites were at an early stem extension stage.

Crop GS and shoot/leaf and stem lengths are shown in Figure 18. Most plants at most sites had reached GS 2.4 (full length of bud visible), but at four sites – Fourburrow and the three late-planted sites, Goonhavern, Mawla and Bodilly – crop development was late, with a smaller proportion of plants having reached GS 2.4 and most at GS 2.2 (shoots elongating, no buds visible) or 2.3 (tips of buds visible). Crop GS were mirrored by shoot/leaf and stem lengths.

No characteristic daffodil rust lesions were seen on either stems or leaves, with the exception of Tregiffian, where, over the whole plot, two small rust-like spots were seen in each of two stems. At Penventon inconspicuous depressions or pitting of the stems was noted, and subsequent observations suggested these may be a widely occurring early-stage of daffodil rust lesions.

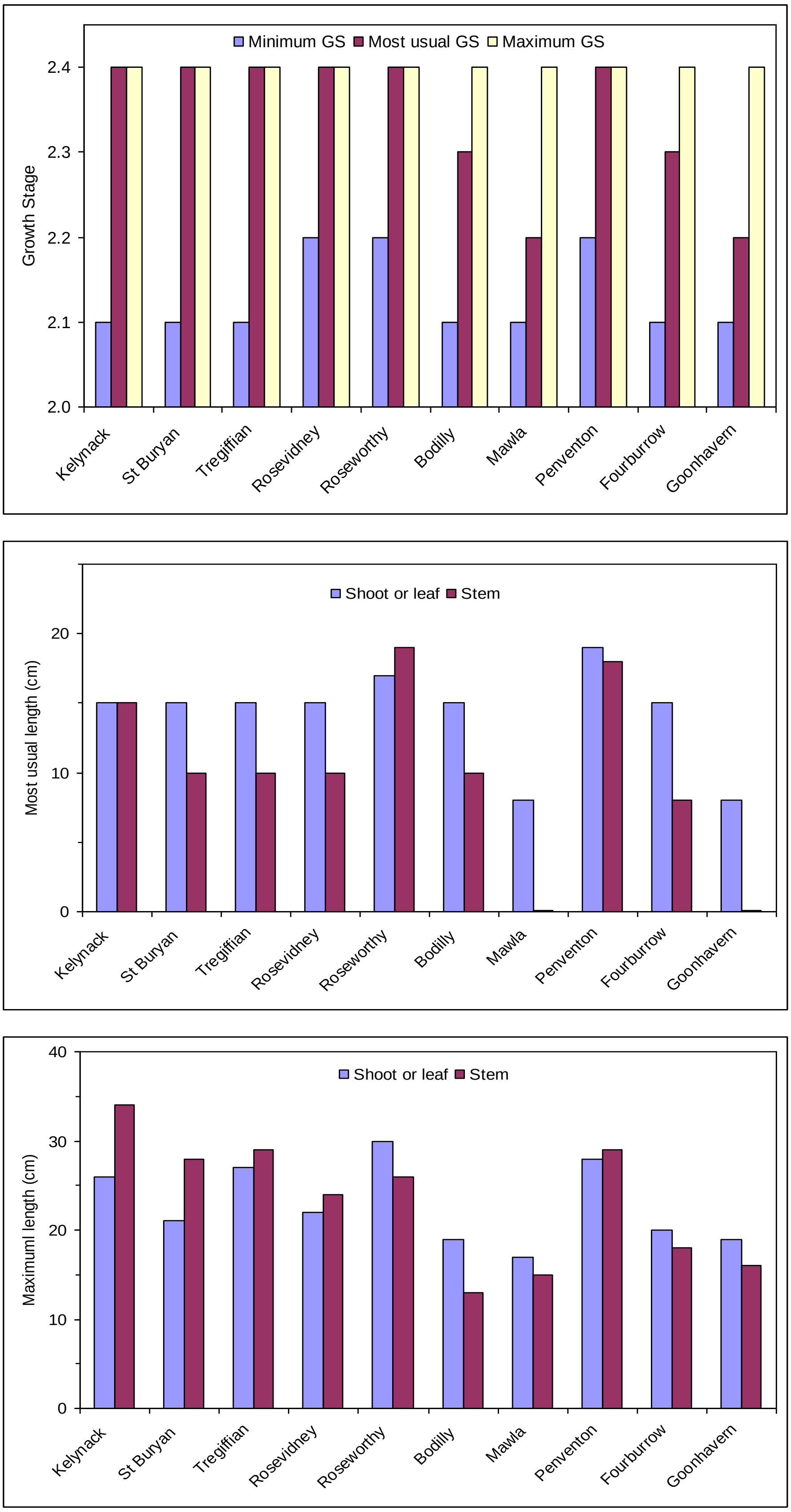


Figure 18. Crop development at ten sites assessed 7 to 8 February 2013: (top) minimum, most usual and maximum GS, (middle) most usual shoot/leaf and stem lengths and (bottom) maximum shoot/leaf and stem lengths.

Picking stage The second crop and daffodil rust assessment was carried out on 27 February to 1 March 2013, when most plants were at about the flower picking stage. Crop GS and shoot/leaf and stem lengths are shown in Figure 19. Most plants at most sites, including Fourburrow (which had been relatively late at the previous assessment), showed uniform and simultaneous development and had reached at least GS 3.1 (early cropping stage) and a few, GS 3.5 (flowers starting to open). However, at the three late-planted sites (Mawla, Bodilly and Goonhavern) crop development remained late, with shorter shoots/leaves or stems and fewer plants at cropping stage; most plants here were still at GS 2.1 (many shoots still emerging from the soil) to GS 2.4 (full length of bud visible).

Only infrequent, small and isolated daffodil rust lesions were found and these at only five of the ten sites. At Penventon and St Buryan a single stem with one daffodil rust lesion was seen at each site, at Fourburrow one stem bore an elongated lesion and a second two small spots, at Roseworthy there were five stems with one to three lesions each, and at Tregiffian (where very slight symptoms had been recorded at the previous assessment) there were ten stems with either a single lesion or several spots near ground level (Figure 20).

It seems likely that the sparse, small lesions found represented an early stage in the development of daffodil rust. In an *ad hoc* test attempting to follow daffodil rust development, a sample of ten stems with lesions was picked and the pattern and size of lesions recorded, and the stems were transported and stored (2 days) before being placed in vases of plain water in ambient room conditions. The lesions showed no further development up to the end of vase-life 5 days later. The impression that daffodil rust lesions do not develop further following cropping has been confirmed by comments from growers and merchants, though it should be confirmed through more formal testing.

In addition to these few distinct lesions, numerous stems throughout bore the fainter marks alluded to in the first assessment at Penventon. The markings resembled slight lengthways pitting or blistering, pale or yellowing spots or longitudinal tracks, none having the characteristic appearance of daffodil rust lesions. While always mild in severity, the incidence of these markings varied between sites (Figure 21).

Leaves were also examined for rust-like lesions. Although no characteristic daffodil rust lesions were found, foliage at Tregithian, Penventon, Bodilly, Roseworthy and St Buryan bore occasional brown, rusty streaking, usually near the leaf-tip. The observations of rust lesions on leaves did not always correspond with those of rust lesions on stems.

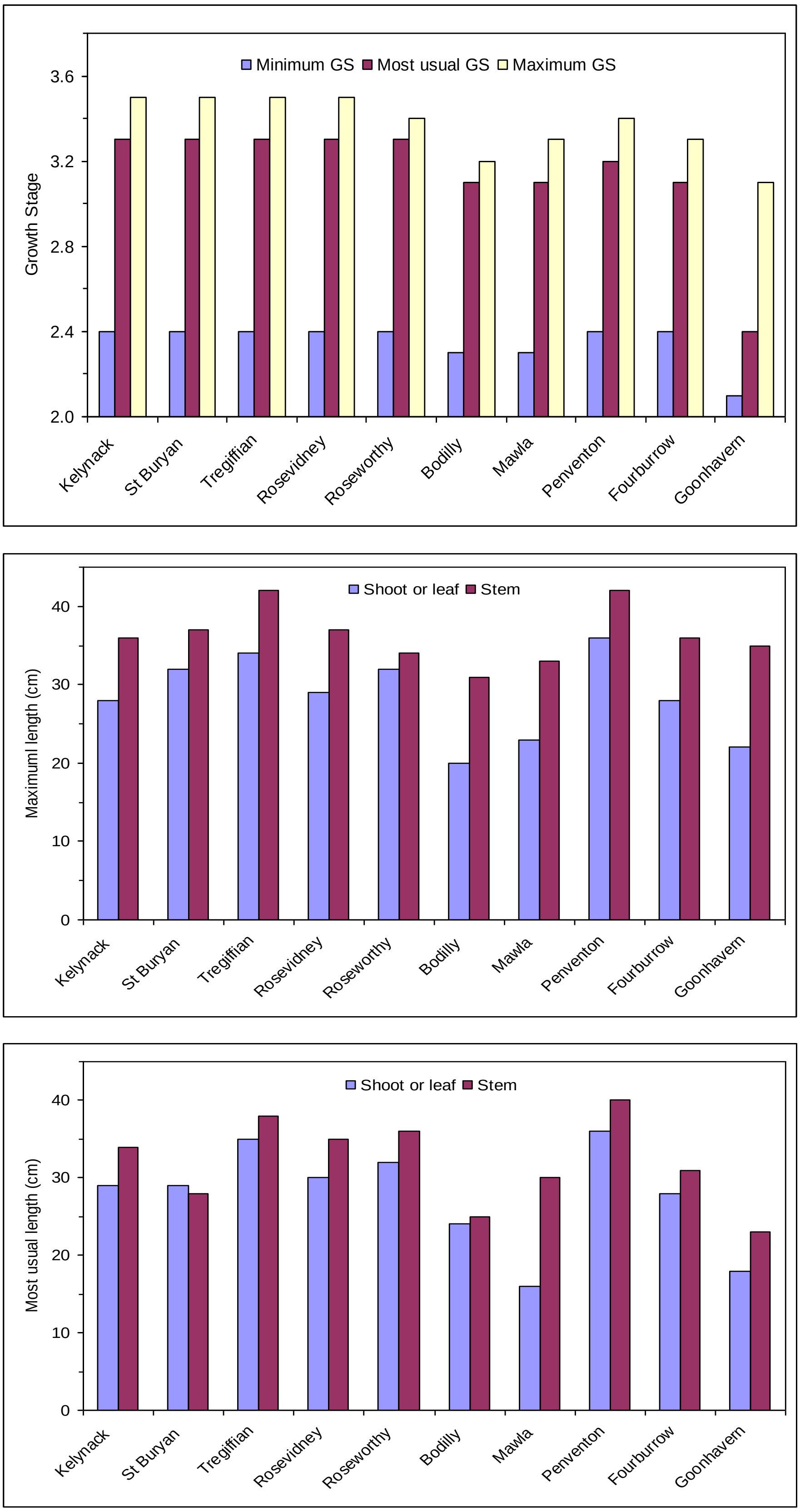


Figure 19. Crop development at ten sites assessed on 27 February to 1 March 2013: (top) minimum, most usual and maximum GS, (middle) most usual shoot/leaf and stem lengths, (bottom) maximum shoot/leaf and stem lengths.

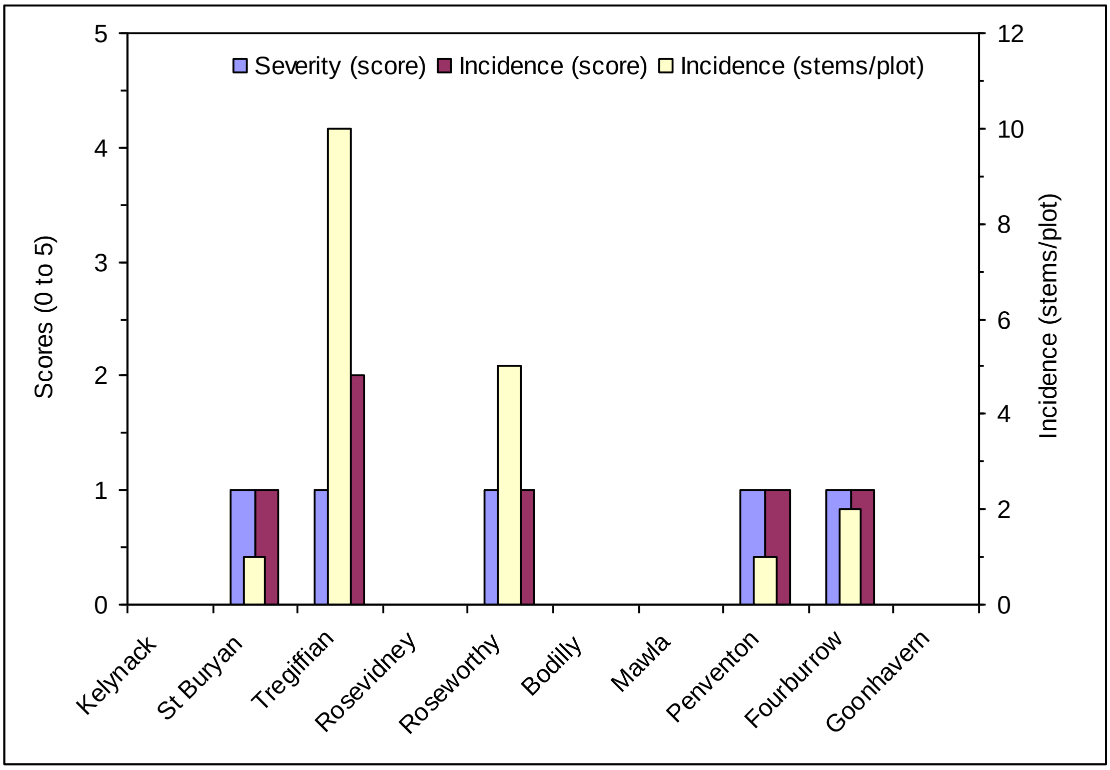


Figure 20. Severity and incidence scores for daffodil rust at ten sites assessed on 27 February to 1 March 2013. Daffodil rust incidence is also shown as the number of stems affected per plot of 1,000 bulbs.

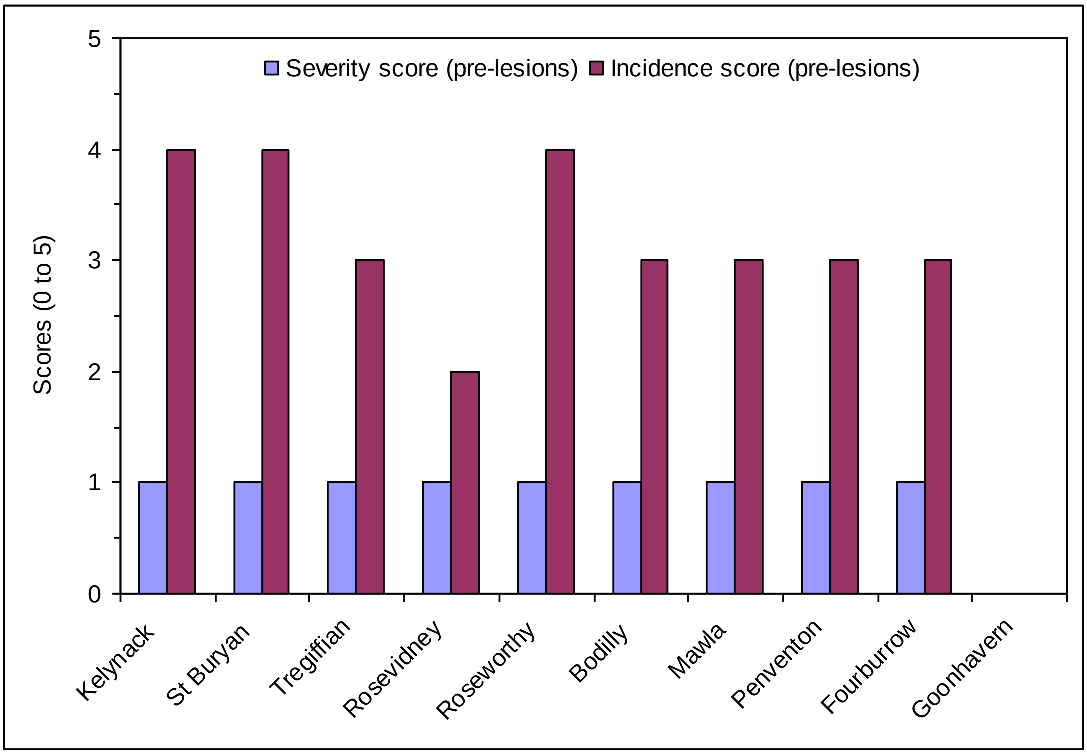


Figure 21. Severity and incidence scores for ‘pre-lesions’ at ten sites assessed on 27 February to 1 March 2013.

Post-picking stage The third crop and daffodil rust assessment was carried out on 26 to 28 March 2013. At this date the earliest crop, at Kelynack, was largely at GS 3.8 (flowers fully senescent), while the latest, Goonhavern, showed a wide variation in development between GS 3.1 (stems elongating, early picking stage) and GS 3.6 (flowers fully open, post-cropping) (Figure 22). Overall, the five most westerly crops were at a post-cropping stage and the five most easterly crops were later, though this may have been partly due to late planting at Goonhavern, Mawla and Bodilly.

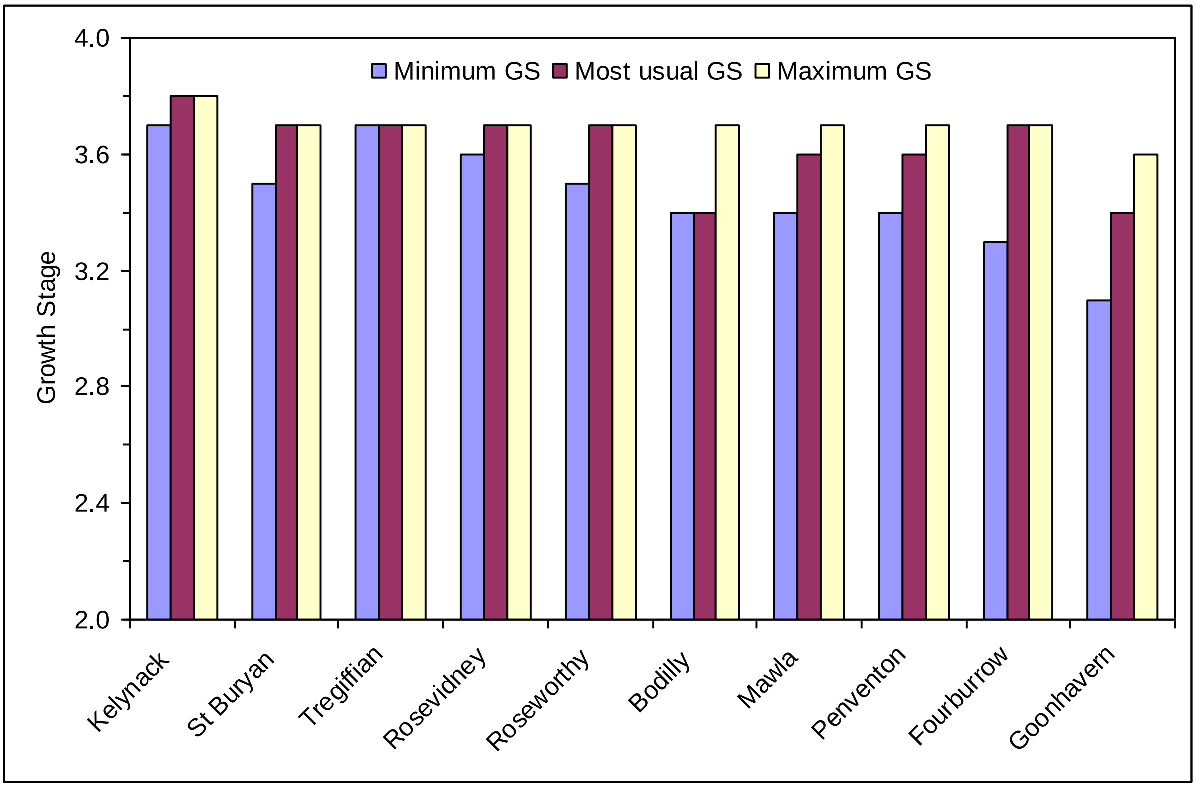


Figure 22. Crop development at ten sites assessed on 26 to 28 March 2013: minimum, most usual and maximum GS.

Small but characteristic daffodil rust lesions were seen at all sites except Fourburrow, one of the sites where its prior incidence had been very low. The severity of daffodil rust varied from one or two small spots (or occasionally streaks) per stem at Goonhavern, to individual small spots and groups of up to *ca* 15 small spots at Tregiffian, the earliest site to have show daffodil rust. Likewise, incidence varied from <10 stems/plot affected at Goonhavern to >100 at Tregiffian, Rosevidney and Penventon. The severity and incidence scores and the number of stems affected are shown in Figure 23. 'Pre-lesions' were seen at all sites though no attempt was made to record their severity or incidence.

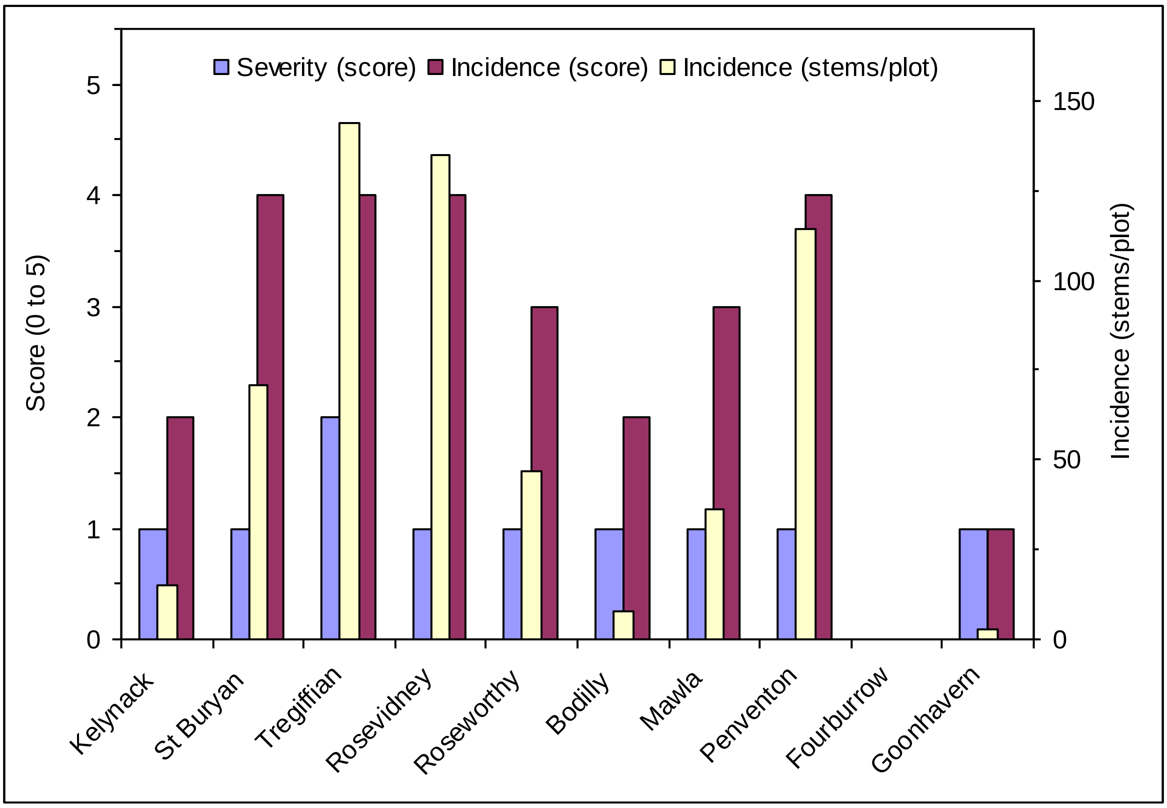


Figure 23. Severity and incidence scores for daffodil rust at ten sites assessed on 26 to 28 March 2013. Daffodil rust incidence is also shown as the number of stems affected per plot of 1,000 bulbs.

Rust-like spots or streaks were also found on leaves at all sites, many streaking down from the leaf-tip, and usually with a low severity (a single or a few small spots or streaks per leaf) and incidence (a few up to about 10% of leaves affected) (Figure 24). At Kelynack, St Buryan and Tregiffian the severity of leaf lesions was greater, with more extensive markings and greater incidence (more than 50% of leaves were affected). However, the relationship of these foliar, rust-like lesions to the stem lesions needs further investigation.

The presence of other pests, diseases and disorders was noted when other records were taken (Table 12). Frost-damaged leaf-tips were common, and there were occasional flower-opening disorders, leaf-tips damaged by *Stagonospora*, and smoulder symptoms. Yellow streaking on the leaves was common, possibly the result of cold weather or virus infections. These issues were not site-specific and appeared to be a stock problem. The crop foliage appeared pale at Kelynack, while at Mawla and Goonhavern the crops were late and uneven and some foliage was yellowish-green.

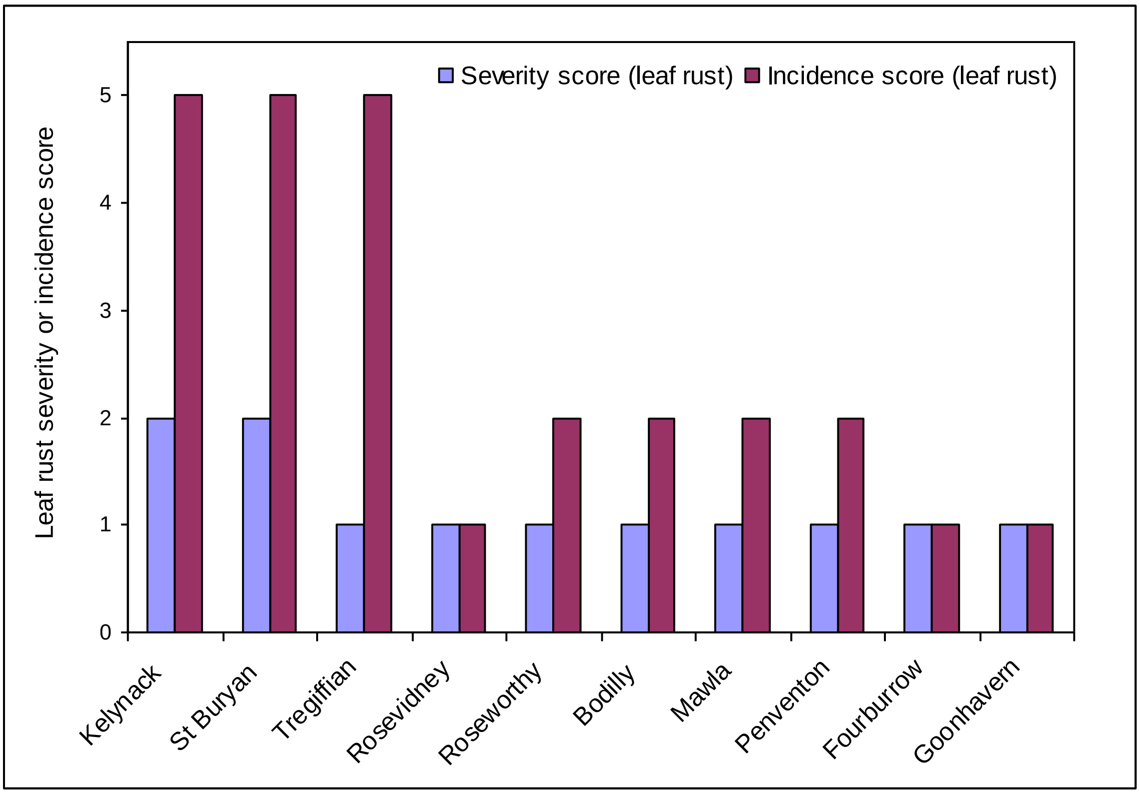


Figure 24. Severity and incidence scores for daffodil rust leaf lesions at ten sites assessed 26 to 28 March 2013.

Table 12. Notes on other pests, diseases and disorders observed at the ten sites during the 2013 growing season.

Site and name		Pests, diseases and disorders seen
A	Kelynack	Numerous yellow leaf-tips and bands. Several dead buds and *Stagonospora* leaf-tip lesions. Rare nibbled buds, flowers with *Botrytis spotting* and prematurely open, distorted buds. Leaves pale.
B	St Buryan	Numerous yellow leaf-tips and bands. Several *Stagonospora* leaf-tip lesions. Rare brown non-splitting spathes, dead buds, prematurely opening flowers and smoulder primaries.
C	Tregiffian	Occasional yellow leaf-tips and bands. Occasional *Stagonospora* leaf-tip lesions. Rare brown non-splitting spathes, dead buds, prematurely opening flowers and smoulder/bulb-scale mite damage.
D	Rosevidney	Rare dried spathes, prematurely opening flowers, *Stagonospora* leaf-tip lesions and dead buds.
E	Roseworthy	Occasional fungal leaf-tip lesions. Rare distorted flowers and dead buds.
F	Bodilly	Occasional smoulder primaries, smoulder/bulb-scale mite damage and dead buds.
G	Mawla	Numerous yellow leaf-tips and bands. Rare nibbled buds. Weak, late and

		uneven growth, pale foliage and much wind damage. Some plants sickly yellowish-green.
H	Penventon	Rare smoulder primaries, smoulder/bulb-scale mite damage, prematurely opening flower, *Stagonospora* leaf-tip lesions and dead buds.
I	Fourburrow	Several yellow leaf-tips and bands and *Stagonospora* leaf-tip lesions. Rare curved leaves (smoulder symptom) and bulb-scale mite damage. Two patches near row ends damaged by tractor.
J	Goonhavern	Several stems broken by wind. Shoots late, sparse, uneven and some plants sickly yellowish-green.

Summary of daffodil rust occurrence in 2013

The progress of daffodil rust development over the three dates is shown in Figure 25. Tregiffian had both the earliest appearance of daffodil rust and its highest final incidence. Fourburrow, Goonhavern, Bodilly and Kelynack remained virtually free of the disorder. Daffodil rust levels were not related their position on the west-east axis.

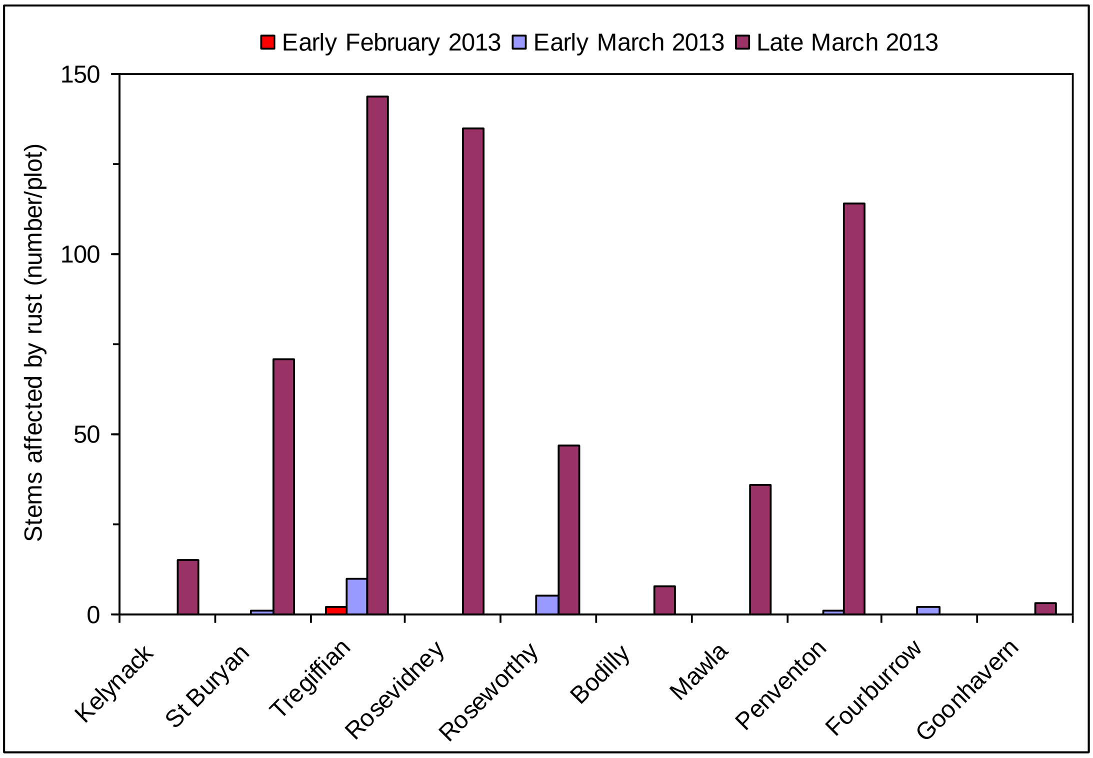


Figure 25. Incidence of daffodil rust at ten sites assessed on three dates in winter-spring 2013. Figures are the number of stems with lesions per plot of 1,000 bulbs.

Associations between daffodil rust, SWC and other factors

In the first year of the project preliminary data analysis was used to assess which, if any, of the soil, water and other factors recorded might have an association with the levels of daffodil rust. Various ways of expressing daffodil rust 'levels' (incidence, severity, number of leaves affected per plot of 1,000 bulbs) were tried; the number of leaves affected by late-March was found to be most useful. The basis of these assessments was box-and-whisker plots and simple scatter plots. In the second year multiple regression analysis will be used to explore the associations and interactions further.

<u>Associations with SWC</u>

In 2013 flower-picking was over at all sites by the end of March and, by implication, the maximum expression of daffodil rust on picked stems would have been reached by that time. SWC and weather data had been logged from 1 November 2012, once all plots had been planted, so there were five months' data to explore.

Figure 26 is a box-and-whisker plot of average SWC (i.e. the average of the 100, 200 and 300mm-deep readings) over the five-month period November 2012 to March 2013, in relation to rust incidence (expressed as the number of stems with lesions per plot at the end of March) at the ten sites. The legend of Figure 26 serves as a general example of how the box-and-whisker plots are set out. To help visualise any relationships the sites were colour-coded according to their rust level - red for high, yellow for middle, green for low or white for zero. The most noticeable feature is that three of the four sites with a high incidence of daffodil rust were associated with high values of SWC – St Buryan, Tregiffian and Rosevidney, the exception being Penventon. Figure 27 summarises data from the separate sensors at each soil depth, and the same trend, though somewhat weaker, can be seen.

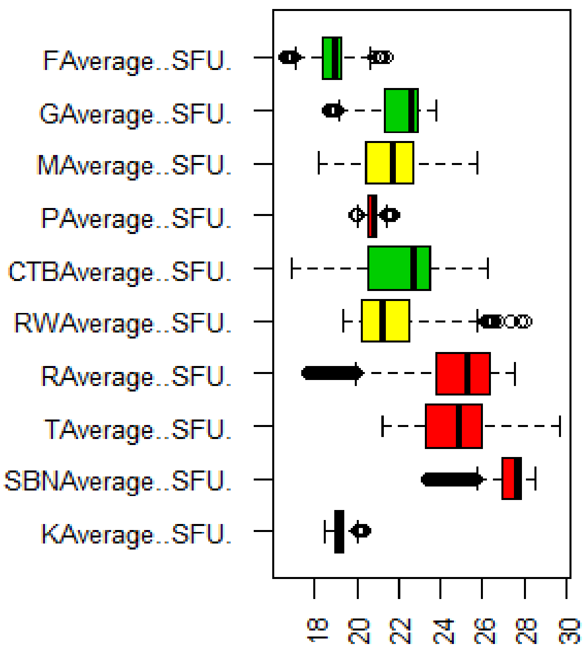


Figure 26. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded in the preceding five-month period (1 November 2012 to 31 March 2013).

Daffodil rust levels were expressed as the number of stems per plot with symptoms in late-March 2013, shown by the boxes coloured red, yellow, green or white for high, middle, low and zero levels, respectively. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. The sites are coded (from top to bottom on the vertical axis) in roughly east-to-west order: **F**ourburrow, **G**oonhavern, **M**awla, **P**enventon, **[CT]B**odilly, **R**ose**W**orthy, **R**osevidney, **T**regiffian, **S**t **B**uryan and **K**elynack. Note the high SWC values associated with the rust-prone sites at St Buryan, Tregiffian and Rosevidney but not Penventon.

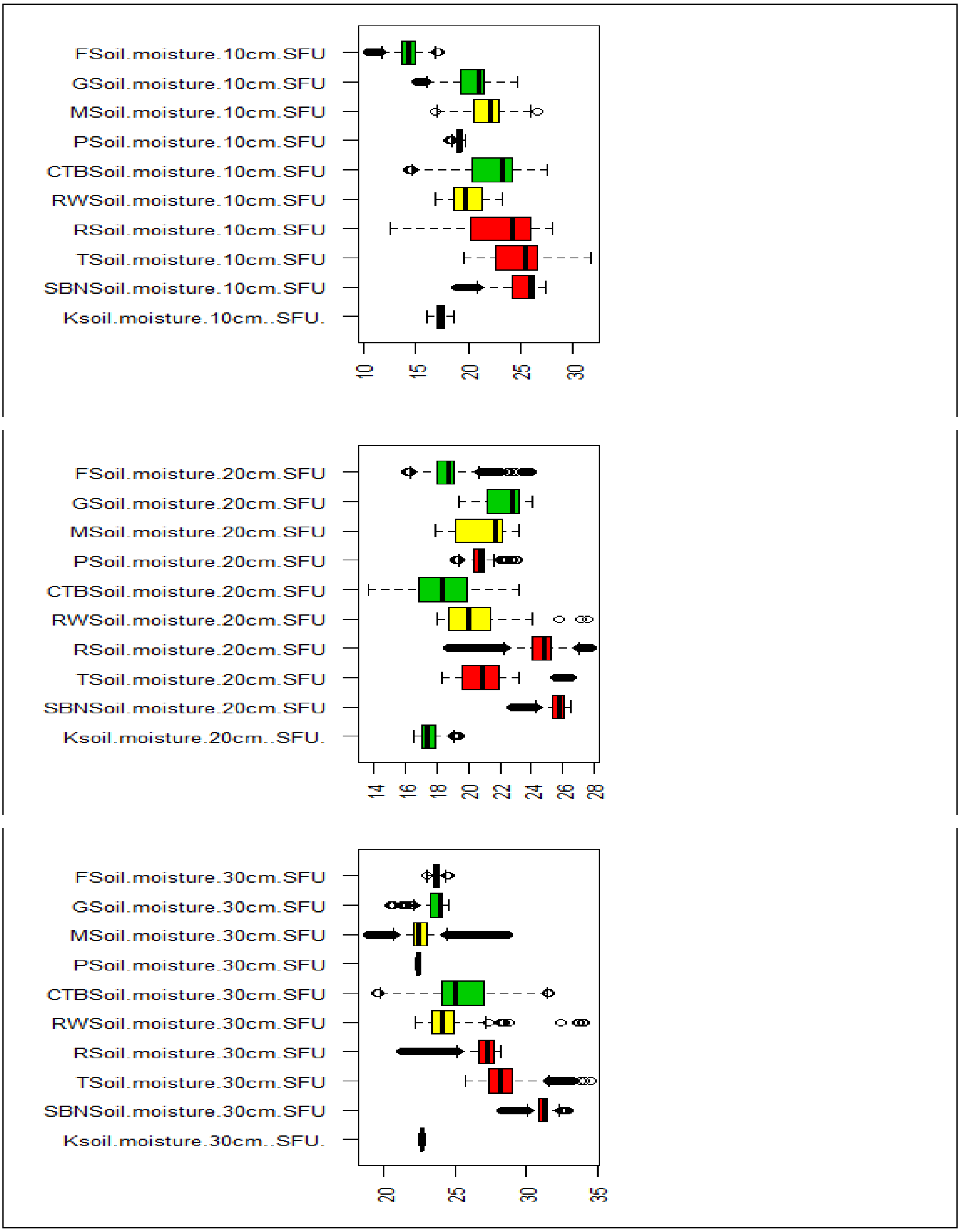


Figure 27. Box-and-whisker plots showing the level of daffodil rust at the ten sites in relation to SWC at 100mm (top), 200mm (middle) and 300mm soil depth (bottom) recorded in the preceding five-month period (1 November 2012 to 31 March 2013). For other details, see legend to Figure 26.

Figure 28 is a plot of average SWC similar to that of Figure 26, but with rust incidence expressed as incidence scores rather than number of leaves affected. It shows that both ways of expressing rust incidence gave similar results, and that the results using SWC separately at 100, 200 and 300mm-soil depth were also the same as before (data not shown).

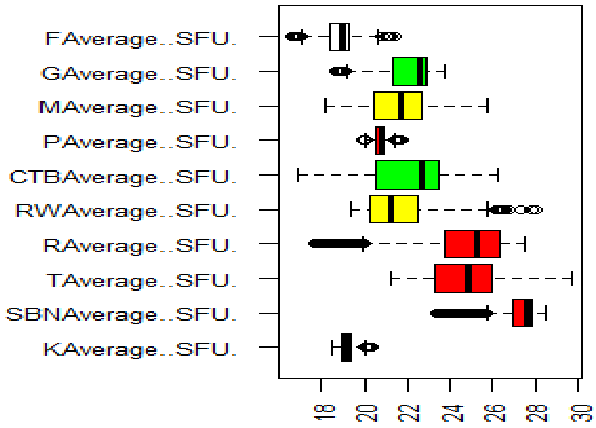


Figure 28. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded in the preceding five-month period (1 November 2012 to 31 March 2013). Daffodil rust levels are expressed as the incidence scores in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. Note the high SWC values associated with the rust-prone sites at St Buryan, Tregiffian and Rosevidney but not Penventon. For other details, see legend to Figure 26.

These data suggest the possibility of an association between SWC over the five-month period prior to flower picking (November to March), but this might be a short-term effect (say, SWC affects stem growth at the time of rapid shoot growth that immediately precedes flowering) or a longer-term one (perhaps an accumulating effect of SWC over the previous months, or at some earlier key-stage in development). To investigate these possibilities, the equivalent data for shorter periods were also examined, i.e. December to March (Figure 29), January to March (Figure 30), February to March (Figure 31) and March only (Figure 32). Comparing this series of box-and-whisker plots (Figure 26 and Figure 29 to Figure 32), it can be seen that the apparently rust-enhancing effect of high SWC has lessened by the January to March data-set and was not evident thereafter, implying that the key-period of the putative SWC effect was November to December rather than January to March. One rust-prone site, St Buryan, exhibited very high levels of SWC throughout. As found with the first data-set (November to March), these later sets showed that the effect of SWC was less evident using readings from the individual SWC sensors at different depths (data not shown), suggesting that the overall SWC is the important factor. If the rust level was expressed instead as the incidence score, the same conclusions were drawn (data not shown). All the above assessments of rust levels were based on the number of stems affected by rust or the incidence score in late-March 2013, when the disorder was considered fully expressed. It was not practical to use the severity score as this was relatively low in all cases. Neither was it practical to use the rust assessments made in early- or late-February 2013, since the levels of rust at this time were very low.

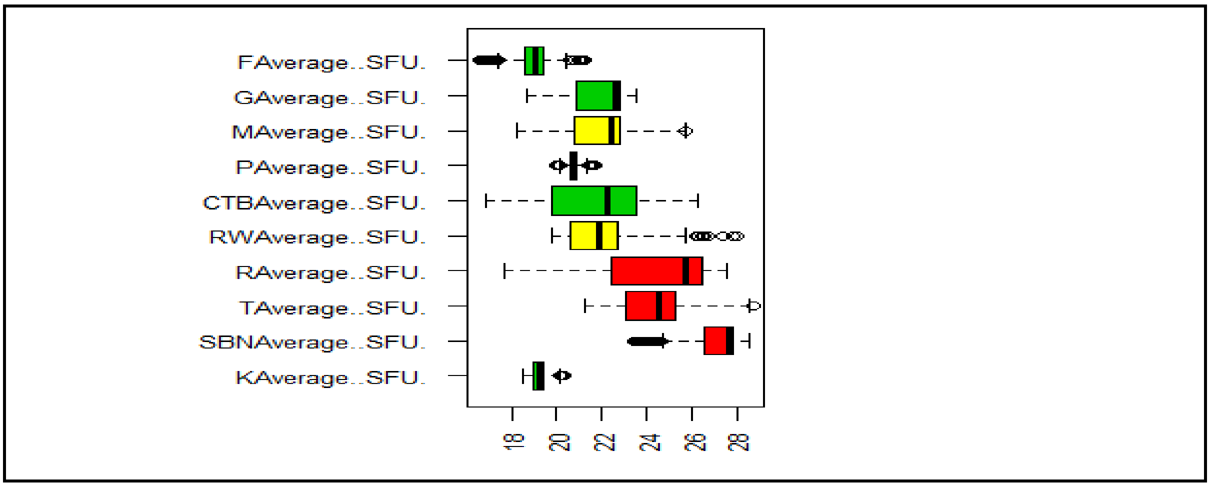


Figure 29. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded in the preceding four-month period (1 December 2012 to 31 March 2013). Rust levels are expressed as the number of stems per plot with symptoms in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. Note the high SWC values associated with the rust-prone sites at St Buryan, Tregiffian and Rosevidney (but not Penventon). For other details, see legend to Figure 26.

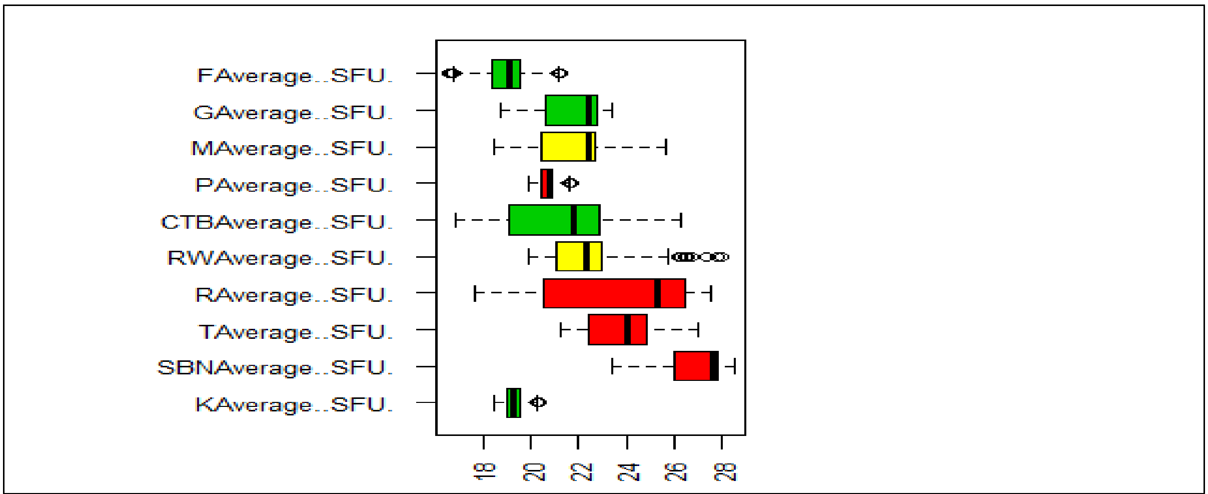


Figure 30. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded in the preceding three-month period (1 January 2012 to 31 March 2013). Rust levels are expressed as the number of stems per plot with symptoms in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. For other details, see legend to Figure 26.

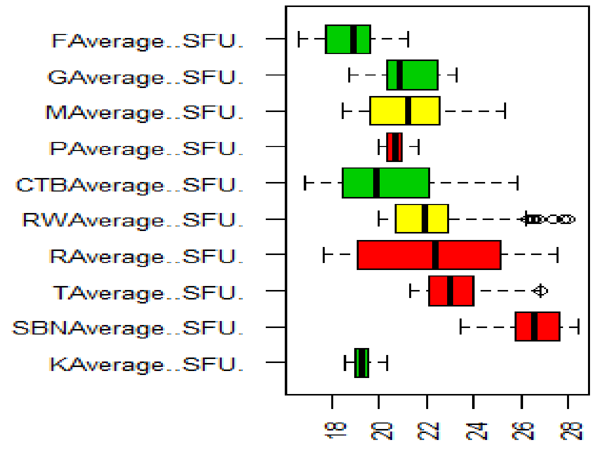


Figure 31. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded in the preceding two-month period (1 February 2012 to 31 March 2013). Rust levels are expressed as the number of stems per plot with symptoms in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. For other details, see legend to Figure 26.

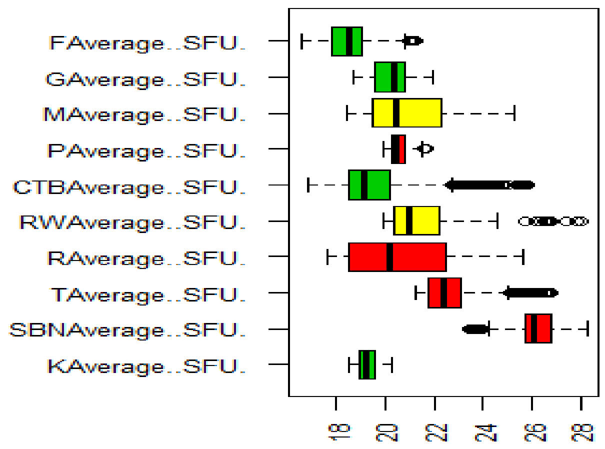


Figure 32. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded in the preceding month (March 2013). Rust levels are expressed as the number of stems per plot with symptoms in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. For other details, see legend to Figure 26.

In order to narrow-down further the key period in which SWC appears to affect subsequent rust levels, the SWC data were next considered month-by-month and week-by-week. SWC data for the four months February to November are given in Figure 33 to Figure 36, respectively (the data for March were presented in Figure 32). These results showed that SWC during November and December appeared to be associated with higher levels of rust, while in January, February and March the effect was diminished. As previously noted, the same relationship held true for rust incidence scores as for the number of stems affected, and a weaker effect was again seen when SWC data were expressed as the readings of the sensors at different soil depths (data not shown).

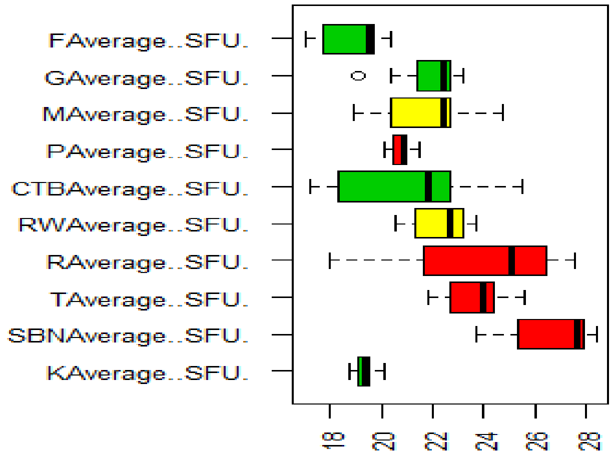


Figure 33. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded in February 2013. Rust levels are expressed as the number of stems per plot with symptoms in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. For other details, see legend to Figure 26.

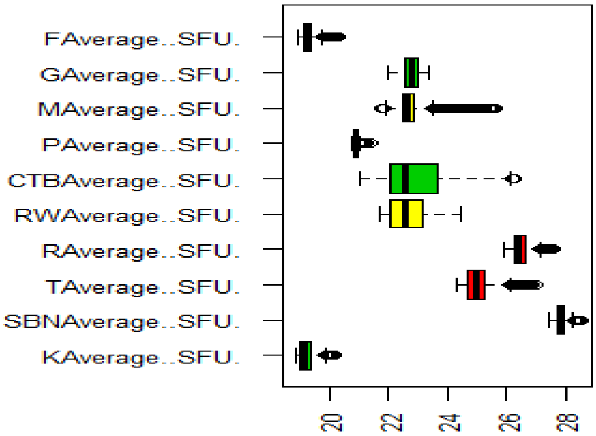


Figure 34. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded in January 2013. Rust levels are expressed as the number of stems per plot with symptoms in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. For other details, see legend to Figure 26.

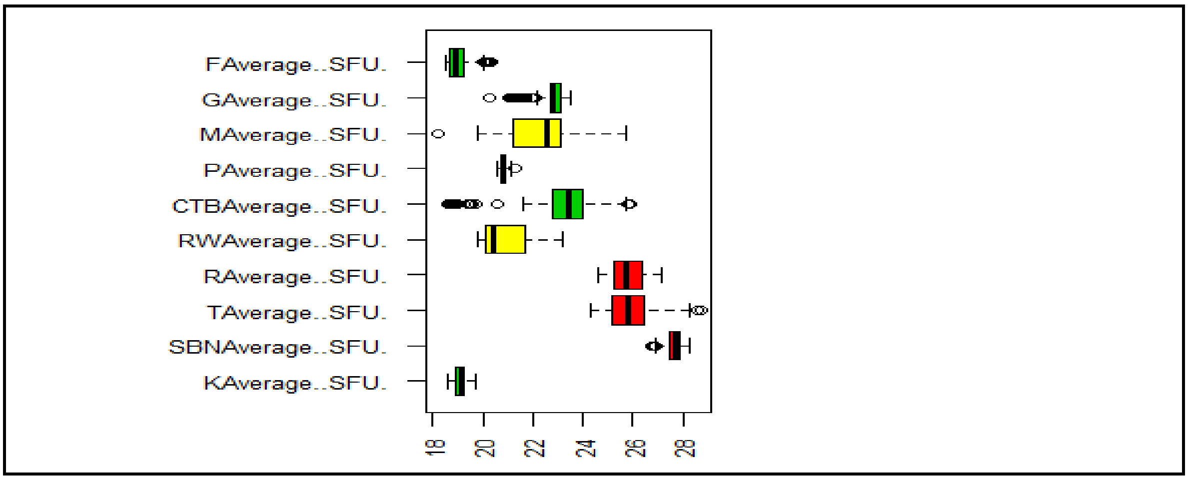


Figure 35. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded in December 2012. Rust levels are expressed as the number of stems per plot with symptoms in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. For other details, see legend to Figure 26.

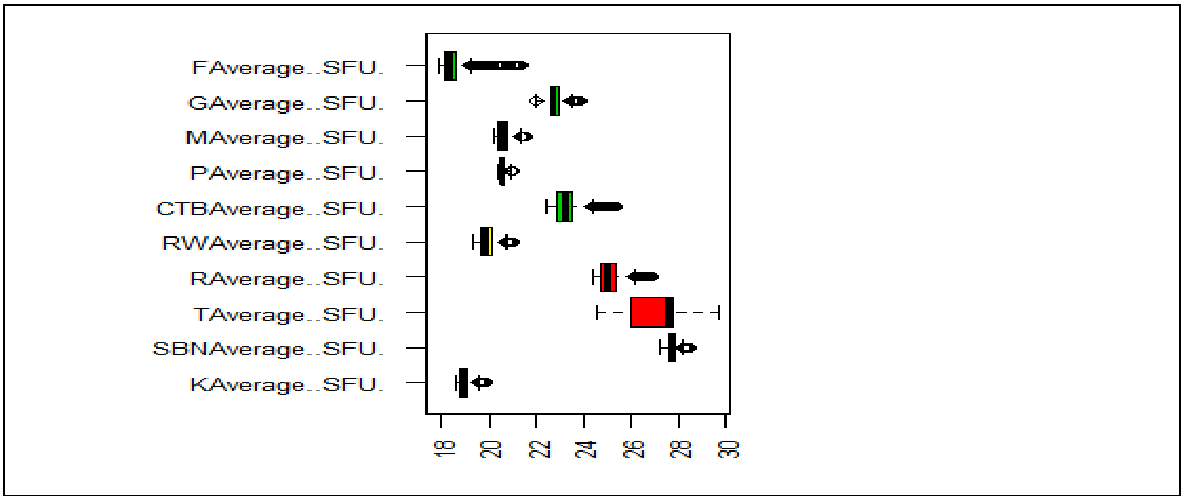


Figure 36. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded in November 2012. Rust levels are expressed as the number of stems per plot with symptoms in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. For other details, see legend to Figure 26.

SWC data were also examined for the separate weeks of November 2012 and March 2013. In the November data there was a clear association of high SWC and high rust levels at the three sites previously noted, while in the March data the effect was weak. Examples are shown in Figure 37 and Figure 38. The data for these periods were re-plotted as scatter plots with a best-fit straight line and the value and significance of the correlation coefficient, r (Figure 39). In these cases the pattern of points is not particularly tight and the highest value of r obtained was 0.601, significant at the 0.1 level of probability only. This suggests an association, particularly using November's data, but one that should be treated with caution until confirmed by further results.

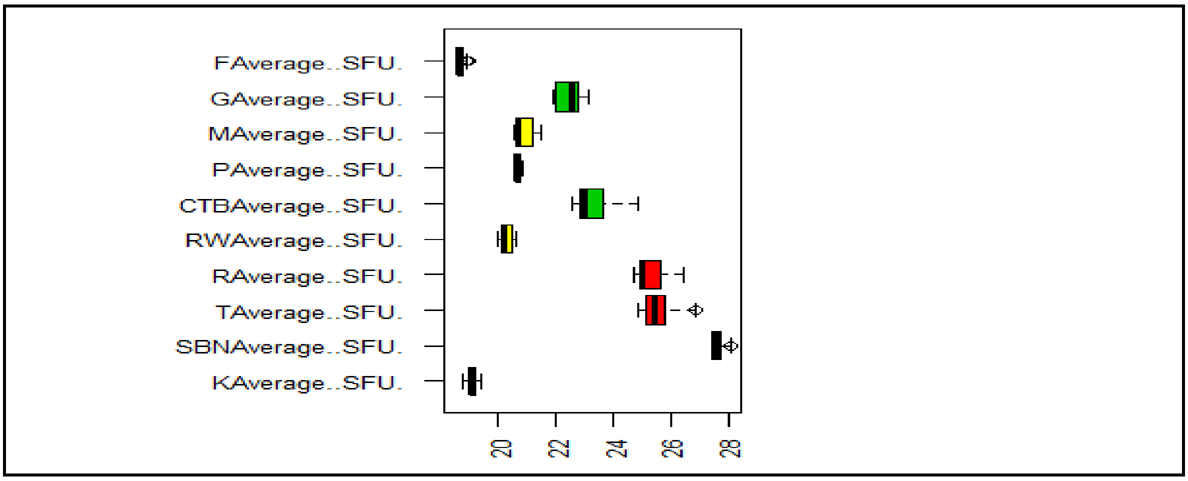


Figure 37. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded during the week 29 November to 5 December 2012. Rust levels are expressed as the number of stems per plot with symptoms in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. For other details, see legend to Figure 26.

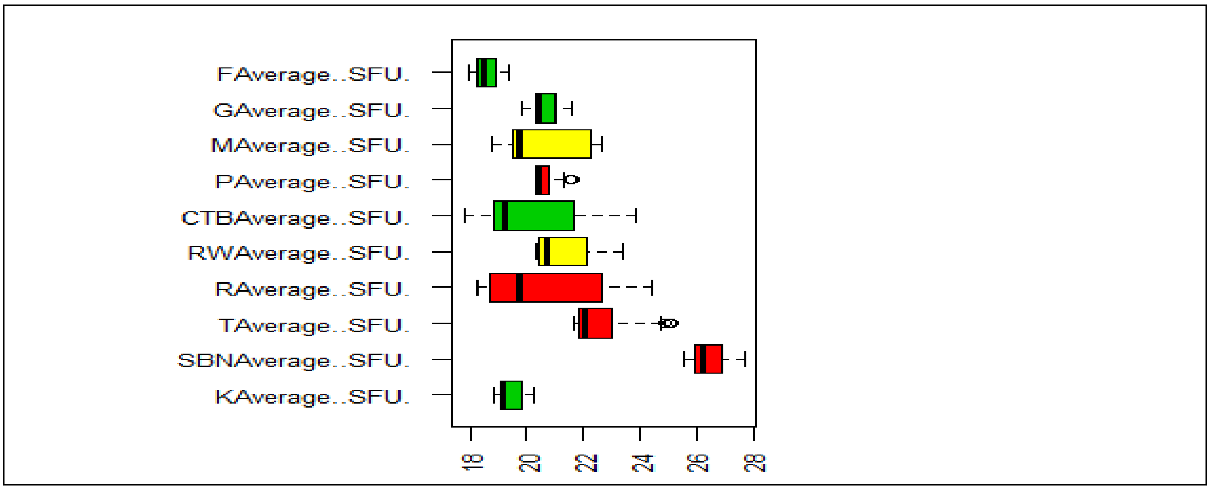
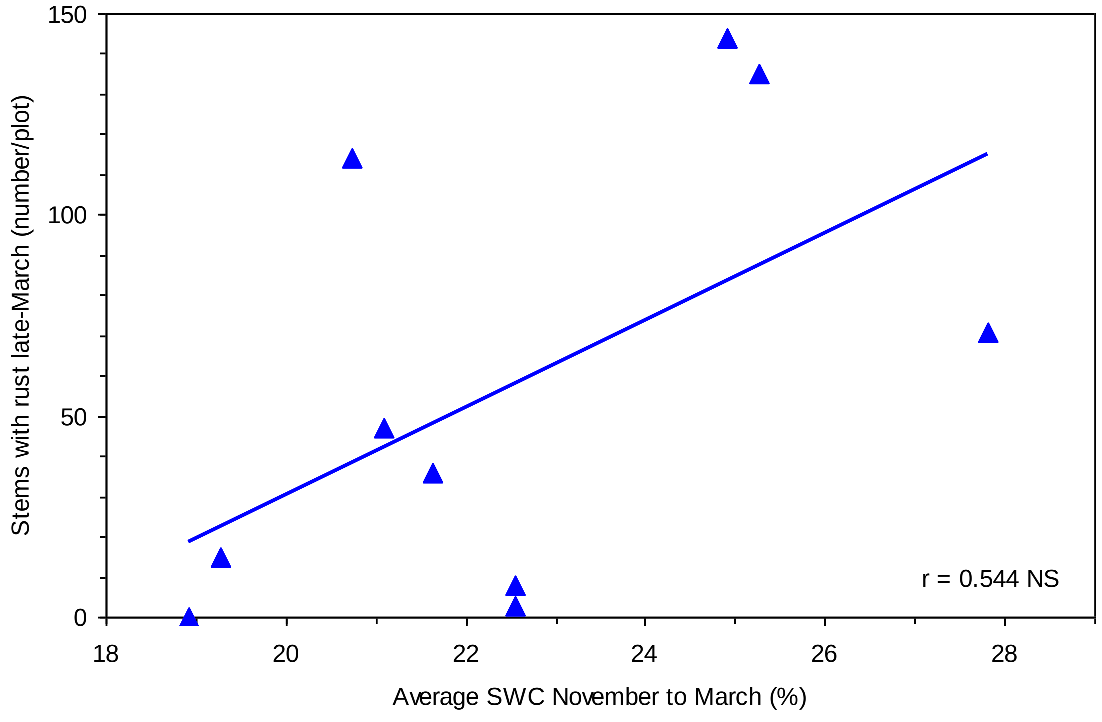


Figure 38. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to average SWC recorded during the week 11 to 17 March 2013. Rust levels are expressed as the number of stems per plot with symptoms in late-March 2013. SWC, shown on the horizontal axis, is the average across sensors 100, 200 and 300mm deep. For other details, see legend to Figure 26.



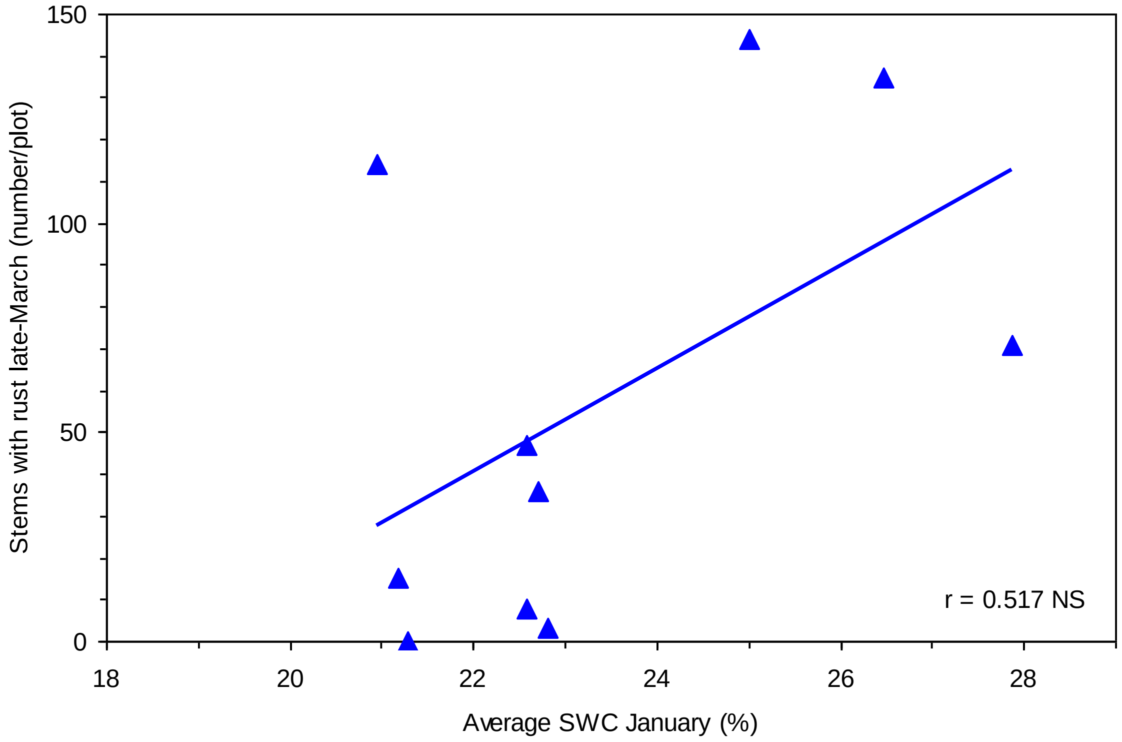
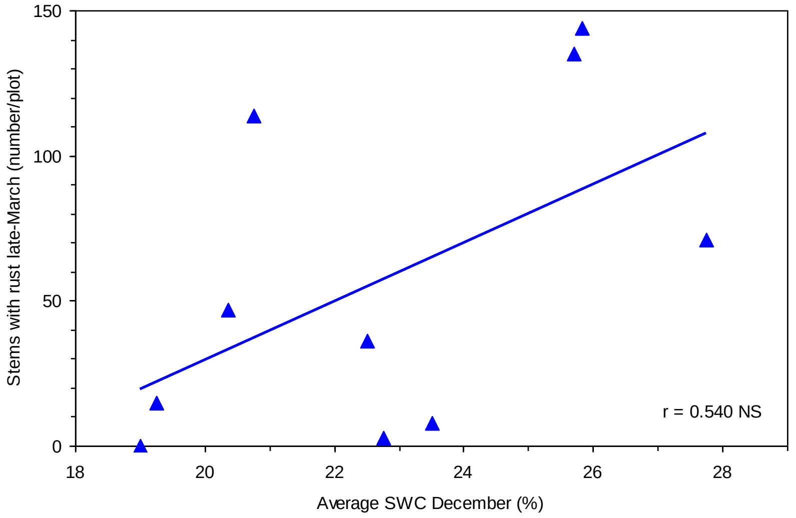
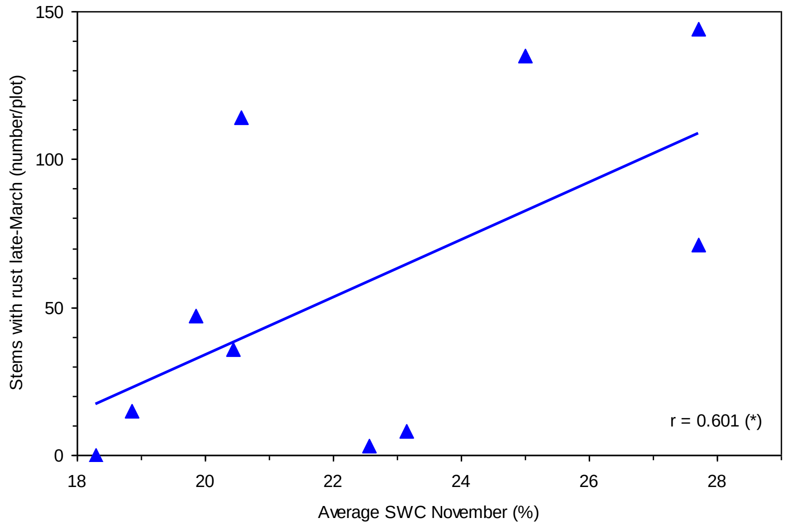


Figure 39. Scatter plots showing the level of daffodil rust and the average SWC at the ten sites for (above, this page) November to March and January and (below, next page) December, November and the week beginning 29 November. The best-fit straight line and the value and significance of the correlation coefficient (r) are shown (see text).





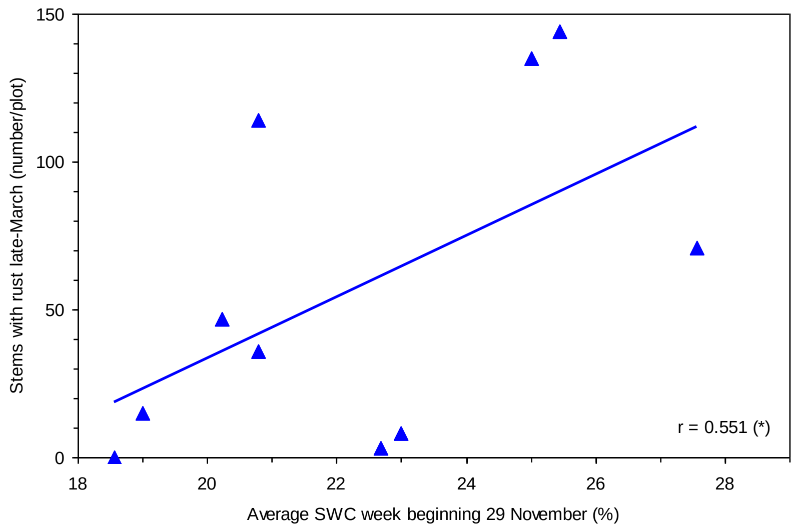


Figure 39 (continued from previous page).

Associations with other weather factors

The levels of daffodil rust at the ten sites were also examined in relation to air temperature and RH. However, using the same approach as described above for SWC, there was no suggestion of much variation in temperature or RH across the ten sites. In all cases the range of temperature or RH were similar. Two examples are shown for temperature (Figure 40 and Figure 41) and two for RH (Figure 42 and Figure 43).

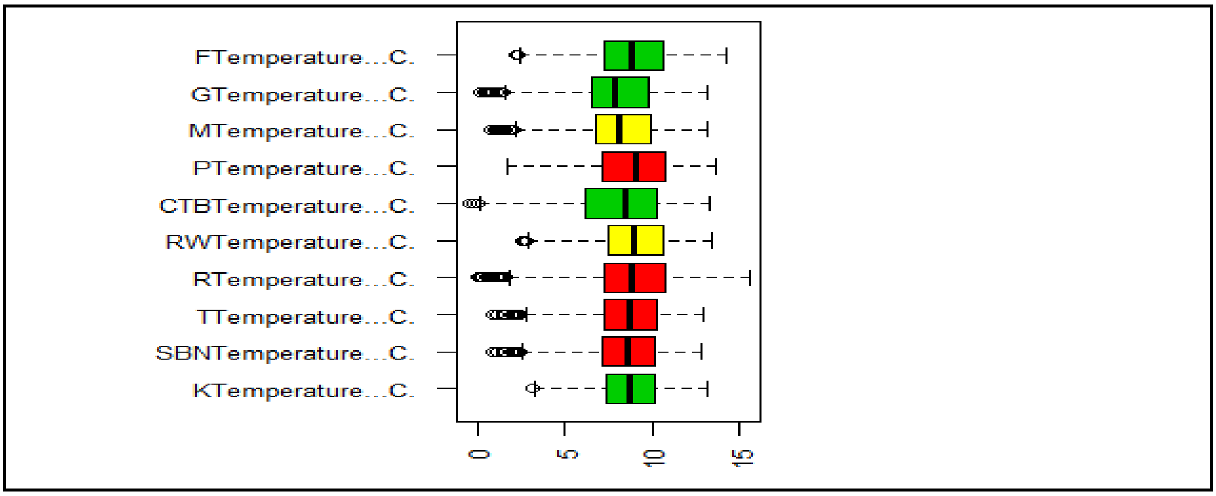


Figure 40. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to air temperature recorded in November 2012. Daffodil rust levels are expressed as the number of stems per plot with symptoms in late-March 2013, shown by the boxes coloured red, yellow and green for high, middle and low levels, respectively. Temperature (°C) is shown on the horizontal axis. For other details, see legend to Figure 26.

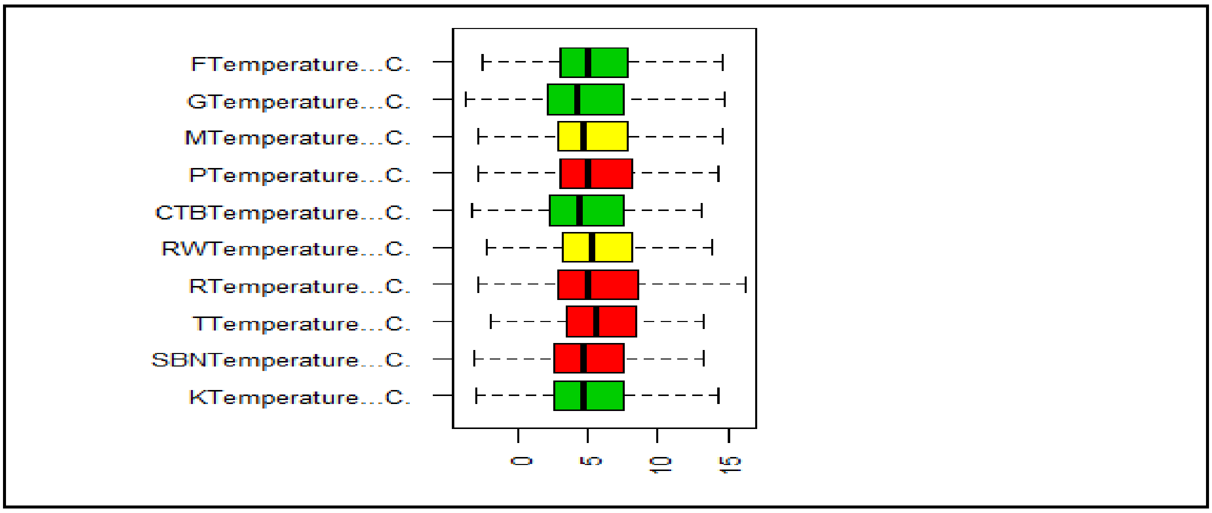


Figure 41. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to air temperature recorded in March 2013. Daffodil rust levels are expressed as the number of stems per plot with symptoms in late-March 2013, shown by the boxes coloured red, yellow and green for high, middle and low levels, respectively. Temperature (°C) is shown on the horizontal axis. For other details, see legend to Figure 26.

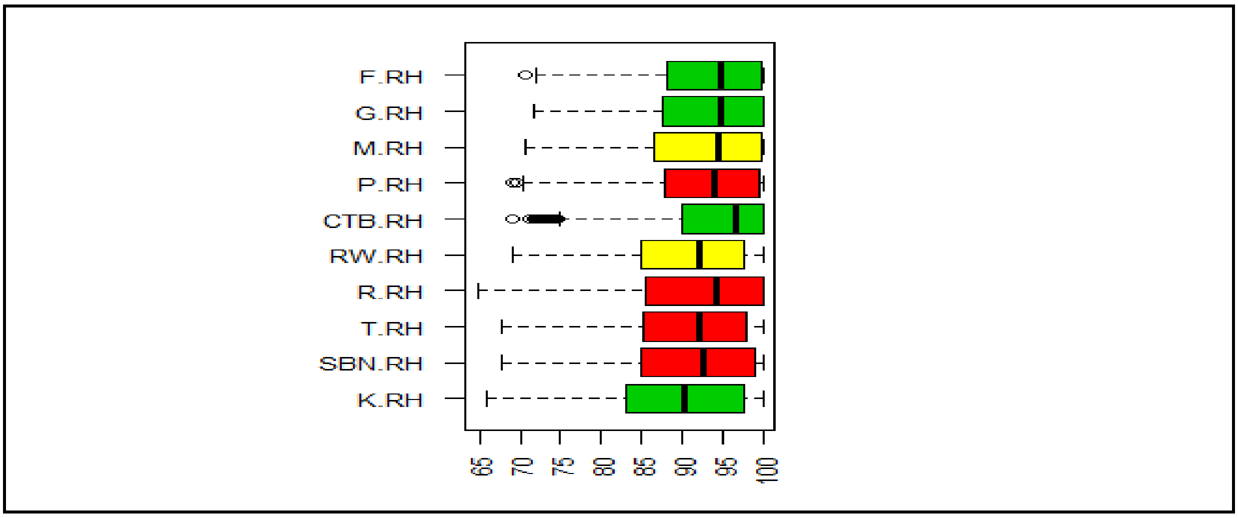


Figure 42. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to RH recorded in November 2012. Daffodil rust levels are expressed as the number of stems per plot with symptoms in late-March 2013, shown by the boxes coloured red, yellow and green for high, middle and low levels, respectively. RH (%) is shown on the horizontal axis. For other details, see legend to Figure 26.

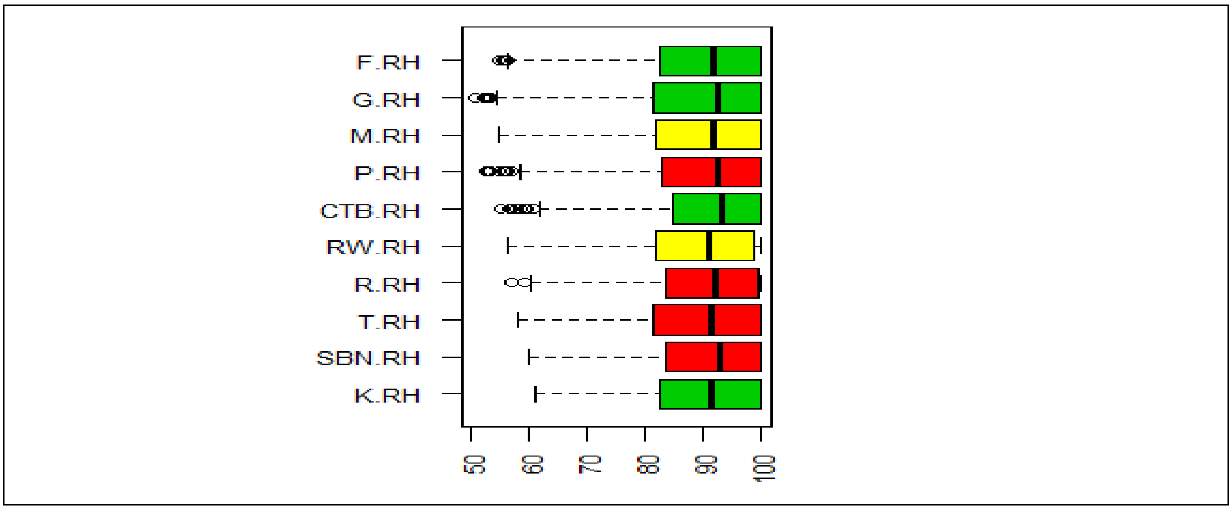


Figure 43. Box-and-whisker plot showing the level of daffodil rust at the ten sites in relation to RH recorded in March 2013. Daffodil rust levels are expressed as the number of stems per plot with symptoms in late-March 2013, shown by the boxes coloured red, yellow and green for high, middle and low levels, respectively. RH (%) is shown on the horizontal axis. For other details, see legend to Figure 26.

<u>Associations with geographical, soil, nutritional and crop factors</u>

In addition to the analysis of SWC and the other logged weather data using the box-and-whisker approach, simple scatter plots with best-fit lines and correlation coefficients (r) were used to search for potential associations between the levels of daffodil rust at each site (usually taken as the number of stems per plot with rust lesions in late-March) and other factors.

Testing rust levels against geographical data – longitude, latitude, altitude and distance from the sea in the prevailing wind direction (south-westerly) (see Figure 44) – showed no significant correlations, with r ≤ 0.516 (NS).

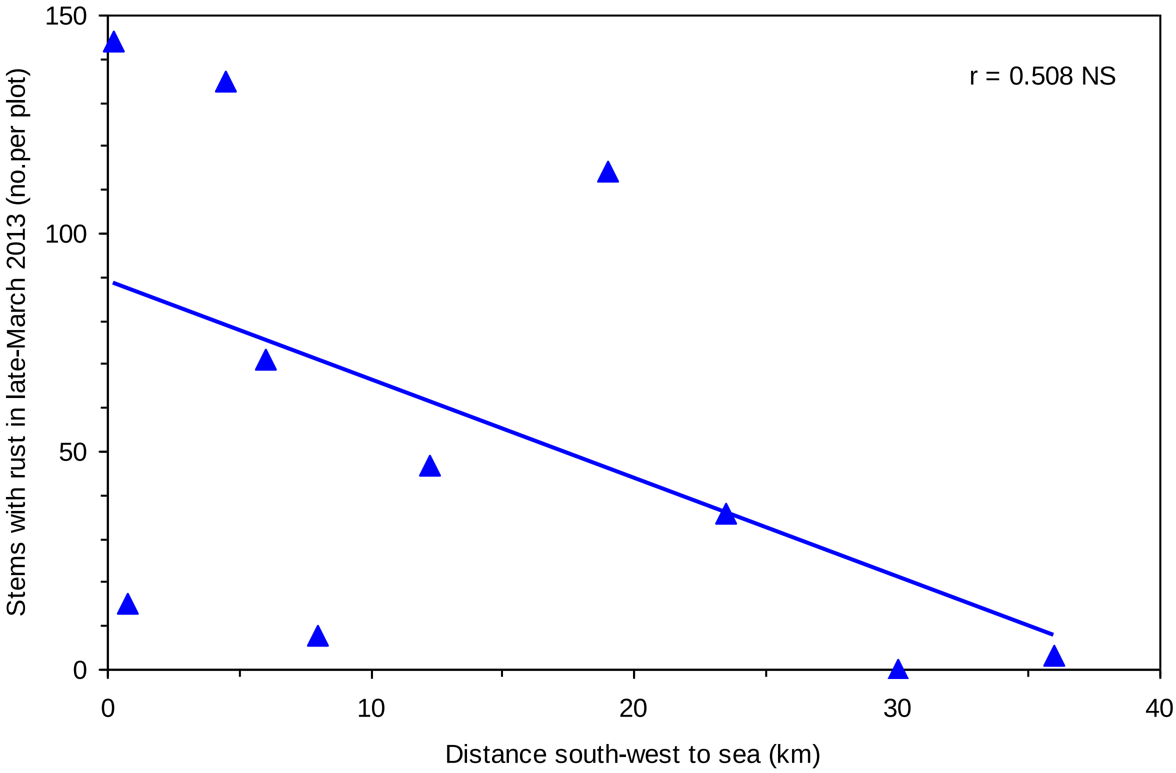


Figure 44. Scatter plot showing the relationship of the level of daffodil rust (y-axis) in relation to the distance to the sea in a south-westerly direction (x-axis) at the ten locations. The best-fit straight line and the value and significance of the correlation coefficient (r) are shown.

Soil structure factors also failed to show any association with rust levels, with r ≤ 0.486 (NS) in all cases and usually much lower. The factors tested were VSSQA (see Table 9), depth of top-soil and of cultivable soil (Figure 17), and the fractions from the soil particle size analysis (see Figure 15 and Figure 16). Although not amenable to formal analysis, there were also clearly no clear associations between rust levels and ADAS soil texture (

Table **13**) or soil series or group (Table 14): high or low rust levels were not confined to one particular soil type.

Table 13. The ten sites and their rust level (number of affected stems per plot by late-March) grouped by ADAS soil texture (see Table 9).

Site		Rust level	Soil texture
D	Rosevidney	135	SCL
E	Roseworthy	47	SL
G	Mawla	36	SL
I	Fourburrow	0	SL
A	Kelynack	15	SSL
B	St Buryan	71	SSL
C	Tregiffian	144	SSL
F	Bodilly	8	SSL
H	Penventon	114	SSL
J	Goonhavern	3	SSL

Table 14. The ten sites and their rust level (number of affected stems per plot by late-March) grouped by Soil Survey of England & Wales soil series and sub-group (see Table 9).

Site		Rust level	Soil sub-group	Soil series
D	Rosevidney	135	541k	Denbigh 2
E	Roseworthy	47	541k	Trusham
G	Mawla	36	541k	Denbigh 2
H	Penventon	114	541k	Denbigh 2
I	Fourburrow	0	541k	Denbigh 2
J	Goonhavern	3	541k	Denbigh 2
B	St Buryan	71	611b	Mortonhampsted
C	Tregiffian	144	611b	Mortonhampsted
F	Bodilly	8	611b	Mortonhampsted
A	Kelynack	15	612b	Moor Gate

Figure 45 summarises the type of fertiliser applied at each site prior to planting the bulbs, generally just P and K, but with N applied at Roseworthy and organic fertiliser at Penventon, and shows that at seven sites brassicas (which leave high residues of N) preceded bulbs (see Table 6). There is no evidence that high or low levels of rust were associated with the type of fertiliser applied or with the previous cropping of brassicas. Correlation analysis confirmed there was no evidence for associations between levels of rust and the concentrations of soil macro-nutrients or soil pH in the analyses of summer 2012, autumn 2012 or spring 2013, with $r \leq 0.375$ (NS) in all cases and generally much lower (seeSoil and plant sampling and disease diagnostics

The project calls for further sampling across all plots when commercial levels of daffodil rust are apparent on some plots. Since only low levels of daffodil rust were present on the plots in 2013, these samples will be taken in 2014.

- Soil and leaf samples will be taken for full nutrient analysis to be carried out at WCC.

- Stem samples will be taken for disease diagnostics at a commercial plant clinic.
- Small stem tissue samples will be taken, frozen in liquid nitrogen, and stored at -70°C at WCC for the determination of viral RNA.

Table 5, Table 10 and Table 11). For micronutrient concentrations (measured in spring 2013 only) the same was true (Table 11) except for a suggestion of a correlation of rust levels to soil aluminium concentration, with r = 0.558, significant at the 0.1 level of probability only (Figure 46).

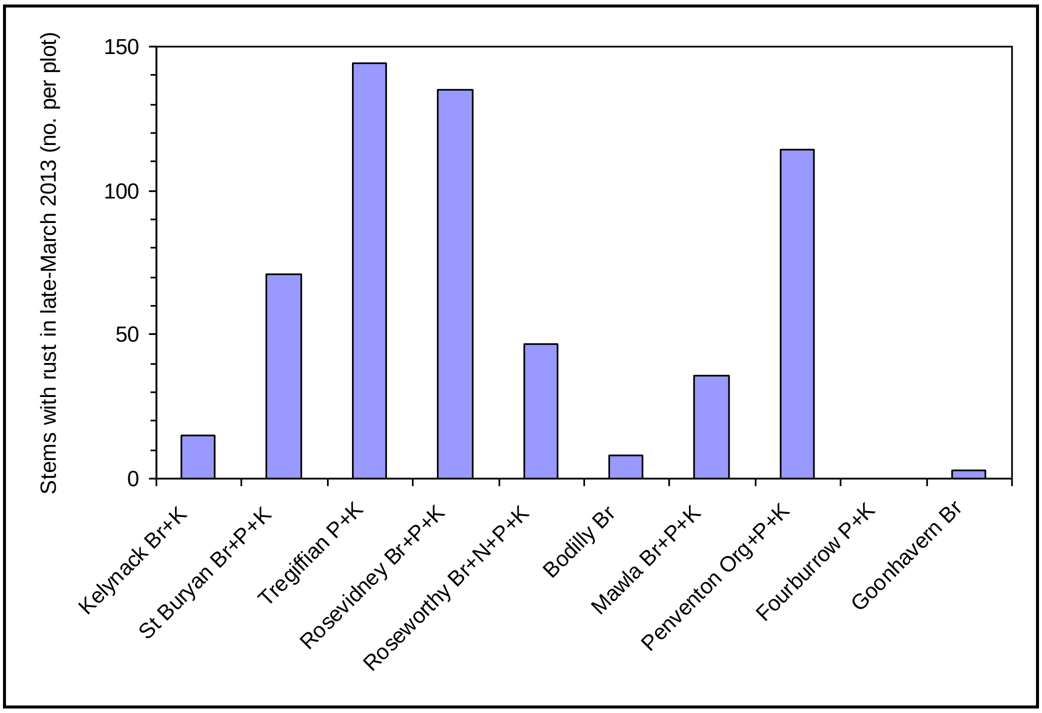


Figure 45. Rust incidence (number of stems per plot affected in late-March 2013) for the ten sites. The type of fertiliser used at each site (N, P K and organic fertiliser (Org)) and the sites where bulbs were preceded by brassicas (Br) are

indicated.

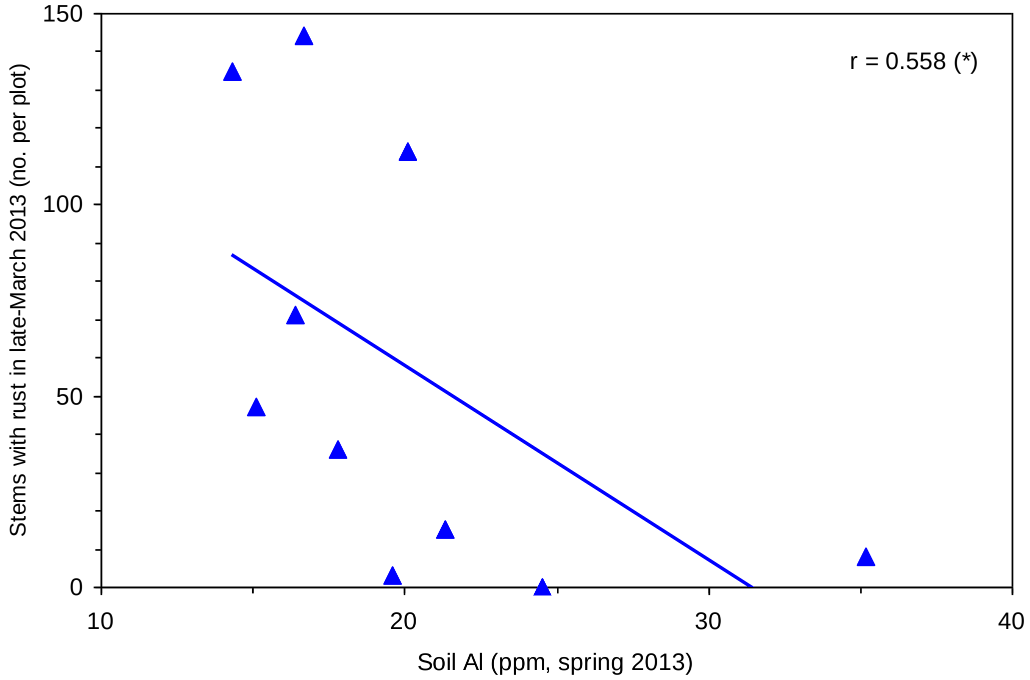


Figure 46. Scatter plot showing level of daffodil rust (number of stems per plot affected in late-March 2013) and the concentration of soil aluminium in spring 2013. The best-fit straight line and the value and significance of the correlation coefficient (r) are shown.

Because of continuing wet weather in autumn 2012 bulb planting was delayed at some sites (see Table 6). However, there was no association of planting date with rust levels in 2013, with r = 0.199 (NS) (Figure 47).

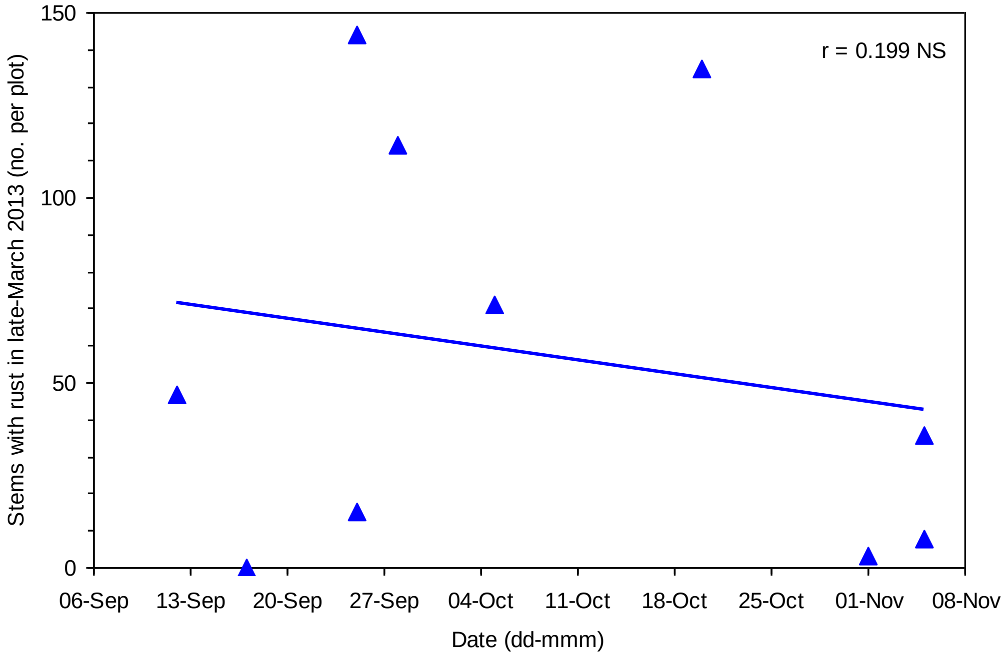


Figure 47. Scatter plot showing level of daffodil rust (number of stems per plot affected in late-March 2013) and the date the bulb plots were planted at ten locations. The best-fit straight line and the value and significance of the correlation coefficient (r) are shown.

To investigate the possibility that rust levels might be associated with the rate of plant development and growth in the run-up to flowering, rust levels were analysed in relation to shoot/leaf and stem length in early-February and their increase in length during February (see Figure 18 and Figure 19). In only one case – that of shoot/leaf growth through February - was there a suggestion of a correlation with rust levels (r = 0.604), though only at the 0.1 level of probability (Figure 48). In the other cases r ≤ 0.512 (NS).

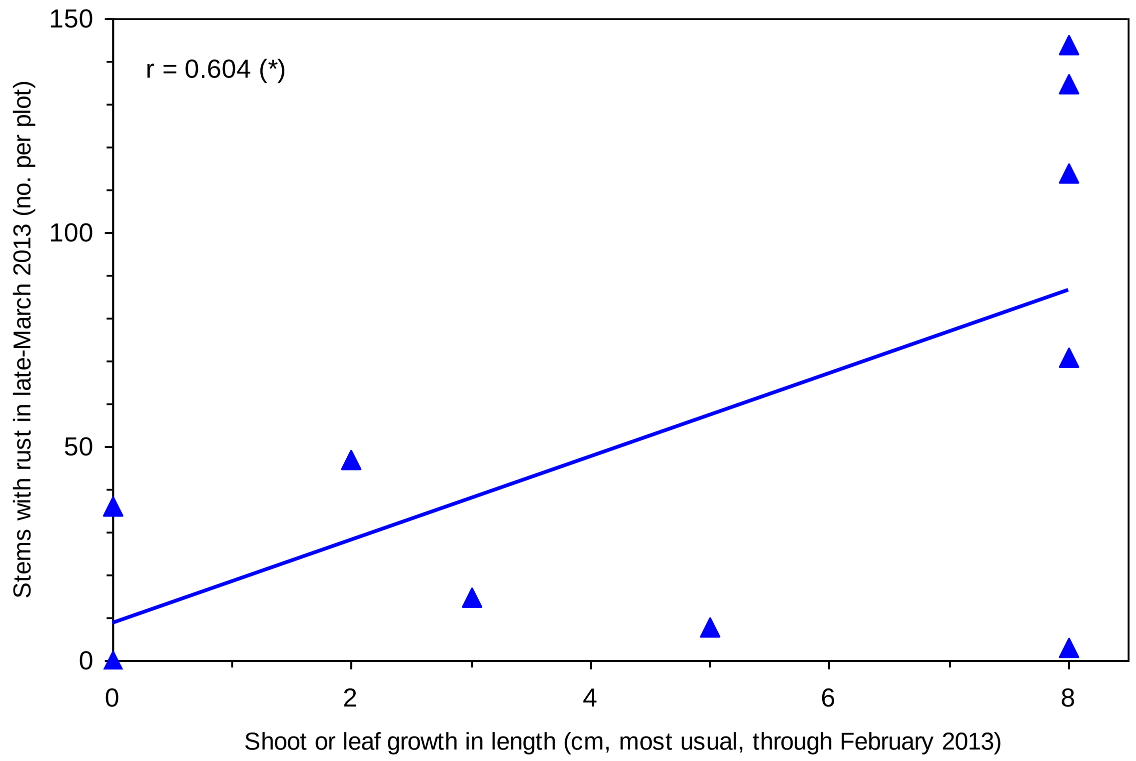


Figure 48. Scatter plot showing the level of daffodil rust (number of stems per plot affected in late-March 2013) and shoot/leaf growth through February. The best-fit straight line and the value and significance of the correlation coefficient (r) are shown.

Discussion

Daffodil rust in context

Awareness of the physiological disorder known as daffodil rust, and its recognition as a significant problem for UK daffodil growers, has come about since the early-1990s, a relatively short period of time. The sporadic nature of daffodil rust – between years, locations and cultivars – has greatly added to the difficulties of developing a rational R&D strategy aimed at understanding and managing the disorder. Thus the present HDC-funded project was initiated after one of most severe rust episodes, spring 2011; since then, rust levels have been much lower (Figure 1), and by general consensus particularly so in the present year (2014). Naturally there may be reluctance to fund the resolution of a problem that appears to have "gone away". However, the HDC's surveys of growers, conducted in 2002, 2003, 2011, 2012 and 2013, showed that daffodil rust is indeed an enduring problem, with real cost implications for daffodil growing businesses. In the survey for 2011, the estimated losses in flower revenue due to daffodil rust were said to vary from 1 to 15% in a serious rust year, while in the good years no losses were incurred through the disorder. Growers responding to the earlier surveys quoted an average reduction in turnover over several years up to 2003 of between 2 and 3%. In the absence of more comprehensive figures, and taking account of the growing awareness of daffodil rust, maybe it would not be unreasonable to work on a 3% average annual loss (across all years), or losses of 10% one year in three with negligible losses in the between years. Because of the way 'official' horticultural statistics are collected in the UK, it is only possible to quote estimated values of the UK daffodil industry.[3] Using these estimates a loss of 3% in turnover of UK daffodil cut-flowers due to daffodil rust would amount to about £0.7m annually, or around £2.3m every three years. While these amounts are modest, in reality there would be additional cost implications associated with unpredictable yields and quality. More importantly, a continuing problem with daffodil rust could result in lowered customer perception of the product and loss of sales on which UK bulb growers depend.

Do adverse soil/water/environmental conditions cause daffodil rust?

With the understanding that daffodil rust probably does not result from a pathogen or a nutritional issue, the suggestion being examined is that some adverse soil/water or other environmental factor is responsible, with some parallels with physiological disorders that cause spotting in other horticultural and agricultural crops (Swain, 1985). At present there is

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[3] Data from the University of Reading (Crane *et al*., 2014) give an annual value of £14m for UK cut-flower exports. Assuming that daffodils make up the bulk of these exports, and that 60% of UK daffodil cut-flowers are exported, UK production would be somewhat in excess of £23m annually. These figures are in line with other estimates used recently (Hanks, 2012).

no possibility of simulating daffodil rust so that its control or management could be investigated experimentally. Therefore in project BOF 76 uniform plots of bulbs were grown at a series of locations, relying on natural conditions and variations to elicit daffodil rust at at least some of the sites. The sporadic occurrence of daffodil rust means that the chance of daffodil rust occurring needed to be maximised by planting the rust-prone cultivar 'Golden Ducat' at a sufficient number of sites in a rust-prone region (west Cornwall). SWC and soil temperature were monitored continuously, along with air temperature, RH and precipitation, at all sites. In this first year of the project the incidence and severity of daffodil rust were low - well below what would be needed to constitute a commercially significant amount of rust – but the incidence of the disorder varied across the ten sites, allowing associations between rust levels and soil/water and other factors to be tested. In preliminary data assessments covering the period from planting to the end of the flowering the following spring, of the environmental variables logged only SWC varied sufficiently between sites to allow possible relationships with daffodil rust levels to be investigated. Relatively high levels of daffodil rust developed in four of the ten sites, and three of these sites also had high SWC, suggesting that high SWC over winter may predispose the onset or development of daffodil rust. Sites with lower SWC generally had lower levels of daffodil rust, but one site had a high rust level but lower SWC. Regressions of rust levels on SWC during November suggested a directly proportional relationship between the two, but, with correlation coefficients (r) of about 0.6 this was indicative only of a weak relationship ($p \leq 0.1$) that should be further investigated before a definite association can be confirmed. Since one of the four high rust sites had a relatively low SWC, there may be some factor interacting with SWC. The 'atypical' site, Penventon, has some features that distinguished it from the other sites, such as a strong, free-draining, south-facing aspect and a previous history as a three- to four-year ley with cattle grazing and no inorganic fertiliser applied before bulb planting. The other sites had varying aspects, slopes, drainage and prior crops (but often brassicas). This tentative relationship between high SWC and high rust levels, and any interactions, will be further tested in 2014. Waterlogged conditions may lead to restricted water uptake and the formation of lesions or oedema in patches of cells of the rapidly growing stems. The early indications are that daffodil rust symptoms in February-March 2014 are both more frequent and more severe than in 2013.

Little is known, overall, of the water relations of daffodils. In studies by the Skierniewice, Poland, group of flower-bulb researchers, SWC was shown to have marked effects on bulb yields in daffodils (Strojny, 1975; Goniewicz et al., 1976) as well as more subtle effects on root nutrient content (Dabrowska, 1975) and root anatomy and stomatal numbers (Goniewicz et al., 1976). This group found that increasing SWC from 40 to 95% of available water capacity increased root (but not bulb) P and K content, and bulb yields were highest in soils near field capacity. In the UK, irrigation was shown to have distinct benefits on yields (Anon., 1985), and while it is rare for daffodil crops to be irrigated in the UK, it is generally accepted that a high SWC after flowering, when the new bulb units are bulking up, is beneficial to bulb yields. In developing a daffodil crop model, Wurr *et al.* (2002) showed that (a) yield of bulbs in the first crop-year was highly correlated with rainfall accumulated between planting and leaf senescence, (b) the yield of bulbs in year 2 was determined by rainfall from planting in year 1, rainfall from emergence in year 2, and by day-degrees >0°C accumulated between flowering and leaf senescence in year 2, and (c) the yield of flowers in year 2 was correlated with bulb yield in year 1 and therefore to the rainfall from planting to senescence in year 1. This raises the possibility that a high SWC might change from adverse to beneficial over a fairly small range.

As well as the flowering disorders found in daffodils, also possibly related to adverse soil/water conditions, daffodils can exhibit severe but temporary wilting when grown under protection if bright morning sunshine follows a cool night (AR Rees, personal communication). This may indicate that the stomata of daffodils do not respond particularly efficiently to stress, and indeed Barton-Wright & Pratt (1931), taking porometer readings of daffodil leaves, found that the stomata took about 2 hours to open or close fully. They were relatively insensitive to stress, for example remaining open under a heavy rain shower. It is unfortunate that no more recent work on daffodil stomatal physiology has been reported.

Are other factors involved in daffodil rust?

Pathological or nutritional causes were suspected as responsible for daffodil rust when the disorder first became significant (see 'Introduction'). Although no pathogen or nutritional disorder has been linked with daffodil rust, this cannot be entirely ruled out and further diagnostics and plant and soil analysis will be carried out in 2014 utilising material from all ten sites. The weak association ($p \leq 0.1$) found between soil aluminium concentration and the level of rust in 2013 will need to be tested further.

In the current project it was shown that soil temperature, air temperature, RH and precipitation were unlikely to be associated directly with the levels of daffodil rust. No associations were found between rust levels and a number of physical soil properties, such as texture, depth, particle size analysis, soil series or soil group. There were no associations between rust levels and longitude, latitude, altitude and distance from the sea in the prevailing wind direction (which might affect the amount of salt-spray). There was no evidence that rust levels were correlated with the type of fertiliser applied prior to planting the bulbs or with the previous cropping of brassicas. There was no evidence for associations between levels of rust and the concentrations of soil macro- or micro-nutrients or pH (other than for aluminium, see above). Finally, there was no correlation with the date bulbs were planted or with crop earliness, though there was a weak relationship ($p \leq 0.1$) to rust levels with the extent of shoot/leaf growth before flowering.

Since daffodil rust is a relatively recent concern, its appearance (or manifestation) might be a consequence of the substantial changes in bulb husbandry that have occurred since the 1970s, when daffodil growing may be said to have changed from ‘traditional’ to ‘contemporary’ practices. Principal changes include the introduction and adoption of hot-air drying, bulk handling, more efficient HWT, early planting, substantially increased planting densities, longer growing cycles (two-years-down at least) and more frequent flower picking and at an earlier growth stage. No specific connections have been proposed, however.

A simple genetic cause of daffodil rust appears unlikely because of the observed wide range of cultivars affected by the disorder and the sporadic nature of symptoms. However, adjacent rusty and healthy daffodil varieties can be observed growing in the same field, suggesting there may be varietal differences in susceptibility. In variety trials at Rosewarne EHS (see ‘Introduction’), not all cultivars showed symptoms in both years of a study, so no inference of a genetic or transmissible basis could be made.

A final suggestion is that physiological disorders such as daffodil rust could be caused by some form of pollution, although no coherent case has been presented. Published reports about the adverse effects of likely pollutants on bulb species are sparse or dated. De Hertogh & le Nard (1993) mention ethylene and fluoride, but give no examples for daffodils. The effects of ‘illuminating gas’ (containing *ca* 3% ethylene) on daffodil cultivars was reported by Hitchcock *et al.* (1932); the effects included the retardation of leaf, stem and bud growth, slight to extensive distortion or curling of the leaf-tips, and bud necrosis. Fluorine, as an air pollutant or a contaminant in superphosphate, can cause a leaf-tip scorch (Spierings, 1969; Gould & Byther, 1979). In maritime situations crops may be exposed to salt spray driven by strong winds, which has been suggested as a possible cause of damage to daffodils.

What do we know about daffodil rust?

The first good description of rust on stems was provided by Andrew Tompsett and used in the HDC survey of growers in 2002 (see Table 1). Tompsett perceptively observed that the mildest or earliest stage of rust was a “[1] slight blistering of epidermis only - ‘not really rust[-like]’”. This was followed by [2] slight cracking and rusting, [3] moderate cracking and rusting that was becoming disfiguring, [4] severe rust that was very disfiguring, and [5] very severe rust with cracking and stem bending. ‘Rusting’ is taken to mean spots or lesions having a distinct rust colour. Stems of category [3] would be unacceptable to discerning customers, while those of [4] and [5] would be unmarketable. In the first year of the current project, samples of commercial stems were seen with this full range of rusting, stem cracking and stem bending, however in the trial plots only the milder stages of rust were observed, leading to a distinction between non-commercial and commercially significant levels of rust. In assessing rust levels in the trial plots, since it was of interest to understand the earlier stages of rust development, more emphasis was placed on the earlier stages, with distinctions made between stems with one or two small rust spots, groups of small rust spots, and larger aggregations of rust lesions that were increasingly accompanied by cracks across the stem. Rust lesions appear to develop anywhere along the green stem. Sometimes stems also have elongated rust-coloured lesions forming streaks down the stem, or more irregular blotches, which at this time are assumed to be a variation of rust symptoms. Rust-coloured lesions and irregularities also appear on leaves, and as the dark-chocolate-coloured lesions of daffodil chocolate spot may also be seen on leaves (and possibly stems), there may be the possibility of confusion. What is the relationship between stem and leaf rust lesions, and is there some relationship of rust with chocolate spot?

The presumed early-stage rust lesions in the form of ‘slight blistering’ of the epidermis, were very widely seen on stems in the trial plots, perhaps most clearly when found in association with one or two small rust lesions. Small, slightly sunken tracts are also seen along the stems, which may also be connected with daffodil rust. Such observations raise the possibility that rust is always present in some form in daffodil crops, or at least in ‘Golden Ducat’. In the trial plots rust lesions appeared to be present from soon after the top of the stem had become visible between the shoots, and to increase slowly up to and beyond the flowering (or flower-picking) stage. It is possible that the early-stage lesions are already present in the lower part of the stem (that growing through the ground or in the bulb itself), a factor that could change thoughts about the origin of daffodil rust and which will be checked in 2014. In commercially significant cases of daffodil rust, presumably the increase in lesion numbers and severity is much more explosive. The length of time the lesions will be visible in a commercial crop will depend on the rate of growth and development of the stem from soon after shoot emergence to flower picking, but this is a window of only about three weeks. The question to ask may be “what factors cause daffodil rust to develop quickly between shoot emergence and flower picking, producing unmarketable stems?”

Although the interpretation of daffodil rust was hampered in 2013 by the low incidence and low severity of rust in the trial plots, at least one factor – a high SWC in winter – has been tentatively identified as a predisposing factor for rust. This factor cannot operate in isolation however, as only three of the four sites with high rust levels were associated with a high SWC, so there may be some interaction of factors. While 2013 was a year for preliminary data analysis, with further information available from the 2014 season multiple regression analysis will be used to study this and other more complex interactions.

Progress and aims of the project

Of the aims of the project, the surveys of growers have been summarised (aim 1) and the field trial studying the effect of the soil/water environment and other factors on the incidence and development of daffodil rust is well into its second crop-year (aim 2). Since only low, non-commercially significant levels of daffodil rust were observed in the plots in 2013, the nutrient analysis of normal plants and plants with rust (part of aim 2), the diagnostic testing of affected stems for fungal pathogens (aim 3) and the low-temperature storage of stem tissue with lesions for possible virus testing (aim 4), have been deferred until 2014. The industry has been kept informed of progress so far, and further technology transfer will take place after the second year's results have been analysed, in late-2014 or early-2015 (aim 5).

Despite the low levels of daffodil rust in the trial plots in the first year, not unexpected from the earlier consensus that rust is not a problem in the first crop-year, the different levels of rust found at the ten sites, and the varied physical features of the sites themselves, enabled associations between rust levels and soil/water and other factors to be tested, at least in a preliminary way, tentatively identifying SWC as one key factor in rust development. This will be built-on in the second crop-year, though the severe storms of early-2014 may affect the results in an atypical way. This is another reason why the project would greatly benefit from a third year of work, as proposed originally.

The exact work in 2014-2015 will be influenced by the results of the nutrient analysis and diagnostic testing of affected plants in 2014, for example the identification of a primary pathogen would require this area of research to be expanded (such as the relationship of daffodil rust with early-stage lesions, leaf lesions and chocolate spot). However, it is expected that most success will come from the further study of soil/water and other environmental factors on rust levels. As an adjunct to this, future work should include studying the subterranean part of the daffodil stem to determine when rust lesions first appear and how this is related to environmental factors.

Assuming progress continues to be made on the soil/water/environmental front, what are the prospects for managing daffodil rust? While risk-avoidance, such as avoiding unsuitable sites and replacing rust-prone cultivars, is an obvious answer, pressures on suitable farming land might make this impractical on any significant scale. Knowing the environmental cause(s) of daffodil rust would at least lead to better risk-assessment regarding suitable sites for rust-prone cultivars. On the other hand, some practical steps might be worthwhile, such as attention to improving drainage in potentially waterlogged fields or low-lying parts of fields by digging temporary ditches or running tines along the furrows before winter rains. It has also been suggested that daffodils may have relatively poor water-use efficiency, and if this were to be confirmed the water use of daffodils might be manipulated at key growth stages through the use of anti-transpirants and practical measures to change stomatal frequency.

Technology transfer

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Appendix 1

<u>2013 Daffodil rust survey form: date of first occurrence</u>

HDC survey of daffodil 'stem rust' 2013: dates of first occurrence of symptoms

The HDC conducted surveys about daffodil stem rust in 2001 and again in 2011 and 2012. Little was known about the disorder, so the results proved very useful. In connection with the HDC rust project, BOF 76, we would like to conduct the survey again.

One important piece of information is the date when the symptoms of daffodil stem rust first appear, because that may be related to weather or soil conditions. This form is designed for you to record this information quickly and easily as the season progresses – perhaps you can keep it on the top of your desk or in your usual vehicle?

Company name and postcode*	Area of daffodils grown
………………………………………………………	………………………ha OR…………………...acres
*These details will not appear in any subsequent compilation	

Variety name	Year-down	Date rust first seen	Severity and commercial significance of stem rust marking				
			Score 1 (very slight rust) Not an issue	Score 2 (slight, not disfiguring) No effect on sales	Score 3 (moderate rust) Downgrade or find other buyers	Score 4 (severe rust) Unmarket-able	Score 5 (very severe) Unmarket-able
Please fill in when rust first seen			*Please assess near flowering or picking time (see diagram)* *Tick one column – or two if intermediate or varying across the stock*				

'Negative' information is also very important. Please could you enter here other varieties grown which remained free of stem rust:

Thank you for completing this form.

2013 Daffodil rust survey form: summary

HDC survey of daffodil 'stem rust': summary for 2013

The HDC conducted surveys about daffodil stem rust in 2001 and again in 2011 and 2012. In connection with the HDC rust project, BOF 76, we are conducting the survey again.

We recently distributed a survey form to enable you to keep a record of the dates of first occurrence of rust symptoms for your daffodil crops in 2013.

At the end of the 2013 flower season, could we please ask you to complete this summary form, which is concerned with the overall impact daffodil stem rust has had on your business over the past few years? *There is no need to repeat information you have already provided on the 'date of first occurrence of symptoms' form.*

1. Company name and post-code (these details will not appear in any subsequent compilation).

2. Area grown *(please insert area of daffodils grown in hectares).*

………

3. Have you seen any occurrence of daffodil stem rust over the past 5 years? *Please tick box.* PLEASE COMPLETE THE FORM UP TO THIS POINT AND RETURN TO US, EVEN IF YOUR ANSWER IS 'NO' – 'negative' information is important to us.

Yes	No

4. Have you ever had rust samples sent for diagnosis of pests/disease, or had plant or soil from affected plants analysed for nutrient content, and if so what were the results and would you mind sharing them with us? IF YOU COMPLETED THIS PART OF THE FORM LAST YEAR, THERE IS NO NEED TO COMPLETE IT AGAIN UNLESS YOU HAVE HAD NEW SAMPLES TESTED IN 2012 – 2013.

5. Economic importance of daffodil stem rust over the last 5 years.
What were the implications of daffodil stem rust on your business?
(please tick all that apply)

	Yes	No	Comments
1. Overall flower sales reduced			
2. Down-grading of flower stems			
3. Unable to supply preferred customer			
4. Need to seek alternative market outlets			
5. Flowers completely unsaleable			

Please provide an estimate of the potential losses in revenue due to daffodil stem rust.

	% Reduction in turnover	Or Actual losses in £ (if you wish to indicate)
Year of highest incidence of rust		
Year of lowest incidence of rust		

6. **Occurrence and severity of daffodil stem rust for the 2013 season.**

Using the pictures attached, please provide an indication of the severity of daffodil stem rust and the varieties most affected during the 2013 season. **IF YOU HAVE COMPLETED THE 'DATE OF FIRST OCCURRENCE OF SYMPTOMS' FORM THERE IS NO NEED TO REPEAT THE INFORMATION HERE.**

Variety	First year [Yes/No]	Second year [Yes/No]	Third year [yes/no]	Fourth year or older [yes/no]	Severity of rust Please tick the score in reference to the pictures attached					
					0	1	2	3	4	5

7. **How serious do you think daffodil stem rust is, compared to 2001 when the first survey was conducted? *(please tick one option)***

Much more serious		More serious	
About the same		Don't have a problem now	
Never had a problem			

8. **Any other comments?**

Thank you for completing this form.