Applied Horticultural Research Pty Ltd

Final Report for Horticulture Australia Ltd

Project VG10123 (completed November 30th 2011) Developing a strategy to control Anthracnose in lettuce

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Horticulture Australia Ltd Final Report

Project VG10123 Developing a strategy to control Anthracnose in lettuce

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November 2011

The purpose of this project was to provide growers, agronomists and other lettuce industry members with a concise summary of the latest techniques available for controlling Anthracnose in lettuce. A secondary purpose was to provide supporting data for fungicide permit applications, registration applications or further research into this disease. The outcomes were achieved by reviewing the latest research and other information available on controlling Anthracnose in lettuce, developing and distributing a best practice guide for the control of this disease incorporating comments from an industry workshop.

This best practice guide is an output of HAL project VG10123 and has been funded by HAL using the vegetable levy and matched funds from the Australian Government.

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Media Summary

There has been a recent sustained increase in the incidence of the foliage disease of lettuce known as Anthracnose (Microdochium panattonianum) resulting in significant crop losses of iceberg, cos and baby leaf lettuce.

In recent years, growing conditions have changed and now cold wet winter/spring periods are increasingly common. Under these conditions, anthracnose appears to have increased in severity and is leading to the major crop losses, and the fungicides prochloraz and chlorothalonil are no longer providing effective control especially under high disease pressure.

A review was conducted of the international literature and current best practices available for the control of anthracnose in lettuce, with a view to developing a control strategy and identifying fungicides which could have the potential to be made available to the Australian industry under APVMA permit system. Registration of new fungicides is also an option worth considering since it does not require regular renewal of permits. A workshop was held in Melbourne on the 4th May and was attended by 56 industry members to discuss a control strategy for this disease and a control strategy document has been prepared and distributed to growers.

Technical Summary

There has been a recent sustained increase in the incidence of the foliage disease of lettuce known as Anthracnose (*Microdochium panattonianum*) resulting in crop losses of iceberg, cos and baby leaf lettuce in the order of $M10 per annum.

In recent years, growing conditions have changed and now cold wet winter/spring periods are increasingly common. Under these conditions, anthracnose appears to have increased in severity and is leading to the major crop losses, and the fungicides prochloraz and chlorothalonil are no longer providing effective control especially under high disease pressure.

The project has brought together the latest research available on control measures for anthracnose in lettuce under cool wet conditions, including chemical and cultural controls, and develop these into an integrated, industry-endorsed control strategy for the short to medium term.

An industry workshop was held in May 2011 where the key findings were presented to 56 industry delegates and their input sought. A best practice guide has been produced and distributed to industry.

The key outcomes from the review and workshop were:

1. Greatly enhanced understanding of spore dispersal by wind and water splash and infection.

A minimum distance of about 10 m (a typical production "bay"), between all lettuce crops, is likely to be highly effective at preventing the spread of the disease.

Optimum conditions for infection are a temperature of 15°C and 8 hours of leaf wetness, but that infection could occur in the leaf axils without leaf wetness requirement provided disease inoculum was present. Spores remain viable in the soil for 18 weeks and can last 78 weeks on dry crop residues, making carryover of inoculum between crops a significant issue.

2. Cultural Controls

Soil health (microbial activity) and the presence of crop residues may have a major effect on disease carryover and infections of subsequent crops. There was agreement that cultural controls are the key area for disease management in medium term in the absence of genetic resistance or new fungicides.

3. Fungicides and other sprayed control agents.

Fungicides are not providing effective control, especially under high disease pressure, and this is related to the difficulty in getting the product into leaf axils where the disease tends to start. Prochloraz is the most effective conventional fungicide for controlling the disease but can only be used in "closed head" varieties and high residues are likely to seriously limit new permit uses of most fungicides in leafy lettuce.

The best prospects for newer fungicide chemistry lies with the new strobilurin fungicides, which have high and widely established global tolerances in lettuce (MRL's). Unfortunately, efficacy data on these chemicals is very limited.

Peter Dal Santo proposed a 3-point strategy for the short term:

1. Permit for Amistar adding anthracnose to existing sclerotinia permit.
2. Permit for prochloraz on leafy lettuce.
3. Permit for chlorothalonil for nursery use and field use with a long with holding period.

A data package has been produced and has been passed on to AgAware Consulting to support applications to extend permits if appropriate.

Introduction

There has been a sustained increase in the incidence of the foliage disease of lettuce known as Anthracnose or Shot Hole, which is caused by the fungus *Microdochium panattonianum*.

Crop losses from this disease have escalated in recent years, and especially since August 2010, to the point where it is causing significant economic losses in iceberg and cos lettuce. The current value of the Australian lettuce industry is about $M187 AUD (2008/9 AUSVEG) and the loss due to Anthracnose would be in the order of 5-10% of production (M. Titley, pers. comm.), resulting in a current loss to the industry of about $M10 per annum on the more conservative estimate.

Mike Titley, procurement strategist for our major lettuce processor (gsf Australia) has identified the following regions are being severely affected by Anthracnose:

- Central West NSW – Cowra & Mudgee (winter & spring supplies)
- Riverland – Robinvale /Wemen (winter and early spring supplies)
- East Gippsland – Lindenow /Bairnsdale (spring /early summer supplies)
- Sand belt – Rosebud /Somerville (spring / early summer supplies)
- Melbourne Metropolitan – Werribee (spring supplies)
- NW Tasmanian coast – Forth (late spring/summer supplies)
- East Tasmanian coast – Cambridge (spring supplies)
- Stanthorpe & Warwick (spring supplies)

The growers who are currently having major problems with Anthracnose are for the most part very experienced and yet all of them have been frustrated by their inability to control the severity of Anthracnose this year despite comprehensive spray programmes once the pathogen was observed.

Anthracnose control in Australian lettuce was undertaken by Vic Galea in the late 1980s (Galea and Price, 1988b). This work was carried out under predominantly dry mild winters, typical of those experienced over the last two decades in many of the lettuce growing areas in southern Australia. More recently, conditions have changed and now cold, wet winter/spring conditions are more common. And under these conditions Anthracnose appears to have become more of a problem and led to the major crop losses described earlier.

The disease was also studied extensively in the 1990s by Wicks et al (1994). In this work the fungicides prochloraz and chlorothalonil were recommended. However, under recent environmental conditions these chemicals are not providing effective control, especially under high disease pressure. This is largely related to the difficulty in getting effective coverage, particularly into leaf axils where the disease tends to start. In addition lettuce (particularly leafy lettuce) attract high residue levels on the edible parts compared with many other crops. This often makes shorter withholding periods extremely difficult to achieve. Prochloraz is the most effective conventional fungicide for controlling the disease but can only be used in “closed head” varieties and high residues are likely to seriously limit new permit uses of most fungicides in leafy lettuce (Wicks pers. comm.).

Modern varieties of iceberg, cos and baby leaf lettuce have multiple resistances to Downy Mildew, Nasanovia aphid and various viruses. However, these varieties are exhibiting very poor field tolerance to Anthracnose compared to the more robust iceberg varieties in use two decades ago, when the work by Wicks et al. (1994) was carried out.

Australia is currently experiencing a La Niña event, as the recent devastating floods in Queensland and NSW attest, and these wetter conditions are likely to continue for some time, almost certainly extending into the coming winter.

A further complication is the widespread growing of lettuce varieties grown for babyleaf production, in addition to the more conventional iceberg and cos lettuce types. These babyleaf types cover a wide range of mainly non-hearting types such as red/green oak, corals, butternut and the newer multileaf types – none of which have any genetic resistance to Anthracnose. These new types were not part of the industry in the 1980s when Galea carried out his work on Anthracnose.

This project has resulted in the development and implementation of an effective control strategy that will provide Australian lettuce growers with the latest information on how to control Anthracnose in the short term to medium term. In the longer term, given sufficient economic imperative, plant breeders expect to be able to introduce genetic resistance Anthracnose into current commercial varieties.

LETTUCE ANTHRACNOSE BIOLOGY & EPIDEMIOLOGY SUMMARY

This section incorporates two key sources of information:

1. An updated literature review on the biology & epidemiology &
2. Field reports and observations reported by growers, agronomists and seed breeders.

For this review, the most important reason for understanding pathogen biology, is to improve methods for controlling the disease. Rather than simply review the existing body of literature, this section attempts to identify those areas where field observations and research reinforce each other, and those where they do not. By looking at the level of linkage between both rigorously evaluated research and field observations, it should be possible to identify the most important aspects of biology that are still poorly understood and are therefore a future research priority.

Taxonomy

The disease is most commonly referred to as lettuce anthracnose, but also as lettuce shot hole or lettuce ring-spot elsewhere around the world. The causal agent is well understood and there is no obvious debate about the species responsible for the symptoms associated with lettuce anthracnose. It was first described in Italy by Berlese (1895) and designated *Marssonina panattoniana* (Berl.). However, in 1986, it was redesignated *Microdochium panattonianum* (Berl.) Sutton, Galea & Price after it was found to be morphologically closer to several other species in this genus (*Microdochium nivale*, *M. Oryzae* & *M. Stoveri*) by Galea, Price & Sutton (1986).

There is no evidence any other pathogen is responsible for the characteristic symptoms associated with lettuce anthracnose. However, it is sometimes confused with other lettuce diseases, such as Septoria, downy mildew, varnish spot, bacterial spot, lettuce ring necrosis virus & tomato spotted wilt virus (Tesoriero L & Wicks TJ, 2011, *pers. comm*). In general though, growers appear to be able to distinguish the characteristic symptoms, most of the time.

Importance of Anthracnose as a lettuce disease

While the disease is widely distributed around the temperate parts of the world (Australia, New Zealand, Europe, USA & temperate Asia), by far the largest body of research has been generated in Australia. The disease also appears to be of far greater economic importance in Australia & New Zealand than elsewhere based on a range of indicators, apart from the volume of research, such as the lack of global fungicide registrations for the disease. This suggests there is something different either in the way lettuce are produced in Australia & New Zealand or perhaps about the virulence of the local races compared with overseas. The potential difference in race virulence was highlighted during this review (Tesoriero L, & Price TV, *pers comm*., 2011) and is discussed in greater depth shortly.

The most likely difference in global production is exposure to wet growing conditions. Spain is the single largest lettuce producer in the EU, as well as the largest global exporter of lettuce, nearly twice the volume of the second largest exporter, the USA. Only 10% of

Spanish production is protected, yet anthracnose is not considered a major problem in most seasons. Lettuce anthracnose tends only to be a problem in the coastal production areas of California during wet springs (Titley M, 2011, *pers. comm*.).

Traditionally, most Australian production occurred under similar reliably dry conditions, either through winter production in regions such as the Riverina and Sunraysia or summer production only in southern regions with high winter rainfall. However, the last 5-10 years has seen a significant shift in patterns of lettuce production in southern Australia. As grower margins are squeezed, rising fuel and transport costs are likely to be responsible, at least in part, for a significant shift to more production in the inherently riskier production regions with higher winter rainfall, simply because they are closer to the main domestic markets.

Agribusiness in the EU and the USA does not see lettuce anthracnose as a major priority for lettuce. Discussions with seed companies based in Europe and the USA, revealed that while it is indeed possible to breed for resistance, it is a significantly lower priority than breeding for either *Nasanovia* (lettuce aphid) or downy mildew resistance. This is reflected in the seed lines currently coming to Australia. A major research and development fungicide manufacturer also mentioned that lettuce anthracnose is only included in global fungicide screens expressly as a local needs entry for Australia & New Zealand.

The high degree of variability in the frequency and severity of anthracnose epidemics on mainland Australia seems likely to be related to the high proportion of outdoor production. The industry has been able to rely on a generally dry climate in most production areas, in most years. In particular, the last 10-15 years of prolonged drought in SE Australia saw lettuce anthracnose rated as a low research priority by the lettuce industry. However, since 2009-2010, record breaking wet conditions moved the disease to one of the top ranked lettuce industry research priorities. This pattern of interest is likely to be repeated based on future weather patterns and the time it takes to bring resistant varieties to commercial release.

However, an interesting observation presented at the workshop (Minchinton L, 2011, *pers comm*.) indicated a number of southern Victorian growers, reported fewer problems with the disease in 2010, than in 2009, despite 2010 being much wetter than 2009. This was believed to have been related to reinvigorated vigilance and management of the disease in 2010, after disease in 2009 caught these growers much less well prepared, following many years of dry and relatively low disease pressure.

Nonetheless, from broad grower consultation in April-May 2011 there was a strong correlation between protracted wet, cool growing conditions and the severity of disease pressure these growers had recently experienced. The 2010 winter-spring period, through in some cases, to Summer 2010-2011 saw a level of disease pressure not experienced for decades. Production in the western Riverina was much less impacted, than it was further east in NSW, or in southern Victoria and Tasmania. The extreme wet impacted the Lindenow production region in East Gippsland later than the Melbourne production regions, with disease outbreaks starting later but also persisting even into December with the freak combination of wet and cool weather. Importantly, wet conditions not only provided a more conducive environment for disease but also hindered grower attempts to maintain sufficiently tight (short) spray intervals.

Strong growth in the fancy, leafy lettuce markets has also raised the level of the problem, with these varieties showing a greater tendency to retain moisture, difficulty in achieving good spray coverage and very restricted access to fungicides.
For the foreseeable future it is likely that Australia & New Zealand will continue to be the key centre for global research and expertise on lettuce anthracnose.

Sources of inoculum: relative importance and persistence

Conidia

Conidia appear to be the most thoroughly researched and best understood of the infectious propagules produced by the lettuce anthracnose fungus. Detailed taxonomic and growth descriptions of these are made in Galea, Price & Sutton (1986). To date, conidia have been found to be the main, if not the only source of field infection in Australia (Galea & Price, 1988b).

They can be a source of primary infection, either as free spores in the soil or on lettuce crop residues, as well as secondary infections from plant to plant (Galea & Price, 1988b). The most extensive and in-depth evaluation of conidia persistence in a variety of situations was conducted by Galea & Price (1998b) - Free, conidia inoculated into several different Victorian soils remained infective for up to 8-10 weeks, but results were highly variable and seemed to be linked to specific biotic conditions in individual soils. Conidia on lettuce debris, remained potentially infectious for longer than free conidia in soil. Soils containing infected leaf material remained infective for 10-20 weeks and varied according to whether debris was buried up to 10 cm deep or remained on the soil surface. While there was no clear trend to this (greater persistence of infectivity on the surface in the first year's trial and greater persistence of infectivity at depth in the second year's trial), conidia infected residues suspended above the ground, well out of contact with the soil could remain infective for 58-70 weeks. Pasteurising soil was also shown to effectively removed infective propagules but no further work has been undertaken to evaluate the effectiveness of soil solarisation.

Based on the persistence described above as well as discussions with both of the authors at the project workshop in May 2011, it was suggested that conidia infectivity in farm soils is very likely related to soil health and the rate that any remaining lettuce residues can be properly composted. Broadly the results suggest the shortest persistence of conidia infectivity were where high rates of soil microbial activity rapidly decompose lettuce crop residues. This may be a direct effect that deprives the fungus of its host or through direct predation of conidia by other microorganisms.

Microsclerotia

The second kind of infectious propagule associated with the *Microdochium panattonianum* are microsclerotia. These are essentially small, resilient vegetative pieces of fungus that can lie dormant, persist and remain infective for considerably longer periods than conidiospores but they remain poorly understood. In Australia, microsclerotia have never been isolated from plants, soil or plant debris, only from artificial growth media (Parman & Price, 1991a). However, research in the US has both found them in leaf cells and shown they can persist for up to 4 years (Patterson & Grogan, 1991) where they are considered the primary source of inoculum. This study also showed there was no significant difference in soil infectiveness for the first 3 years after microsclerotia were introduced.

Chlamydospores
These are thick walled, resilient variant of ordinary conidia. No information on this resting spore structure was described in any literature reviewed. Recent correspondence indicates this structure has still not been found in lettuce leaf tissue examined (Price TV, 2011, *pers. comm*.– Unpublished work by Parman).

Teleomorph (sexual form)

This has form still not been successfully cultured. Even after three years of incubating microsclerotia, no further structural development occurred, making it impossible to know whether microsclerotia are the precursors to the sexual form (Parman & Price, 1991).

Influence of environment (temperature and moisture) on infection

There are two key studies that comprehensively evaluated the combined effects of temperature and leaf wetness on disease incidence. The Australian model (Galea & Price, 1988a) was based on infections with conidia, while the US study (Patterson & Grogan, 1991) modelled infections caused by microsclerotia:

Conidia

Galea & Price (1988a) – Conidia germinated mostly by producing short appressoria which penetrated cells directly. Extensive colonisation of leaf tissue occurred within 72 hours and necrotic lesions appeared as sporulation in and on leaf tissue began after 96 hours. At 5-27°C, infection levels increased with increasing periods of leaf wetness following inoculation. Optimum infection conditions occurred at around 15°C, with a minimum leaf wetness period of 8 hours. The rate of infection at temperatures <5°C or >25-27°C, declined steeply. Disease incidence in the leaf axils was an order of magnitude greater than on the leaf lamina, although the trends were broadly the same. The sharpest increase in the rate of infection occurred between 8 & 12 hours of leaf wetness, while there was little difference in the rates of infection between 4 & 8 hours of leaf wetness.

However, there is evidence both in the study described and in discussions at the industry workshop, that the 8 hour minimum leaf wetness requirement may be much less where water is retained in leaf axils. After only a short period of rainfall, leaf axils can retain a great deal of moisture long after the leaf lamina have dried off, particularly in leafy lettuce types.

Microsclerotia

Patterson & Grogan (1991) – Microsclerotia infected plants from soil at 0-2cm depth. Microsclerotia germinated by production of hyphae that in turn gave rise to conidia. Germination as a function of temperature followed a classic bell shaped curve. Optimum conditions for germination were a minimum of 6 hours continuous leaf wetness at 20-22°C. However germination was possible at 10-30°C, but not at all when temperatures were raised or lowered by 2 degrees outside this range. Infection also occurred with as little as 4 hours of leaf wetness, but not at all with a leaf wetness period of 2 hours or less. While greater disease was generally established through longer leaf wetness periods at all temperatures, the influence of temperature was more important on disease establishment than additional time of leaf wetness beyond the critical 4 hour minimum. Levels of infection were not significantly (P=0.01) increased through increased leaf wetness, beyond a minimum period of 6 hours. Infection increased significantly (P=0.01) as the temperature increased from 10°C to 20°C and decreased as temperature increased from 24°C to 30°C.

Dispersal

The most significant new data that has come to light during this review is as yet to be published in mainstream science journals, and significantly defines the effects of water splash and wind dispersal of conidia. No pre-existing objective data was found on these conidia dispersal methods (splash and wind) during this review. The following information is based on data presented by Price TV, at the industry workshop, 04 May 2011 (full details are available in the workshop summary), and relates to Parman & Price (1991b):

Water Splash

In the absence of wind, water splash from overhead irrigation onto infected soil did not spread spores 100 cm or further from the source and only spread spores 60 cm from the source when less than 3 m from the sprinkler. The majority of spores only spread within 30 cm of the source.

In the field, irrigation methods which added to leaf wetness tended to increase the incidence of infection during relatively dry periods, but not during periods of higher natural rainfall. This strongly concurs with local epidemic histories which have been linked to periods of high rainfall more than irrigation method used, though minimising leaf wetness by using trickle irrigation, is still likely to reduce disease under lower pressure.

Wind

Conidia were liberated at wind speeds of 10 m/s (36 km/h) or greater, but not when wind speeds were 5 m/s (18 km/h) or less. A greater number of conidia were liberated as the wind speed increased. A greater number of spores were found at 20 cm than at 40 cm above the ground. The number of spores found, decreased with distance from the source. These direct measurements of spore numbers correlated well to levels of infection - Infection at wind speeds of 10 m/s (36 km/h) or greater, but not when wind speeds were 5 m/s (18 km/h) or less.

Combined effects of water splash and wind dispersal

Using simulated rainfall, most spores were found in the first 20cm (mainly the first 10 cm) above ground and within 50 cm from the source, at wind speeds of 0-2.5 m/s (0-9 km/h). One other simulated rainfall trial did show infection on trap plants up to 6 m from the source at wind speeds of 3.6 m/s (13.0 km/h).

In the field, isopathic contour maps of disease incidence, show a relatively short distance of dispersal (splash and wind) made over a 7 day period, that accords well with contributing wind and splash effects described above.

Another study (Galea & Price, 1987) did show how under field conditions, epidemics generally start from infection focal points. This observation is supported by growers who have successfully delayed or stemmed epidemic developments in field through vigilance for first symptoms and removing any plants showing symptoms very early in the development of an epidemic.

Seed

Growers seeing infected seedlings coming from nurseries have questioned whether this may be a result of seed borne infection. However, there is no evidence from research to date, that seed borne transmission of disease is likely.

While originally, disease was thought to be seed borne . Then Stevenson (1939) found low rates of seed transmission however the pathogen was not was not able to be isolated on seed from an infected crop and also found that artificially inoculated seed remained infective for not more than 7 days.

More recently, seed harvested from infected field plants failed to produce anthracnose lesions, whether germinated in seed trays of sand-peat compost or in artificial growth media (Galea & Price, 1988b). In the same study, even when seed was deliberately, artificially inoculated with conidia, these rapidly lost infectivity under normal storage conditions, with little difference whether stored at 5° or 20°C. While viability was greater when infected seed was grown on artificial growth media, no infected seedlings appeared from seed that had been stored for 24 days when planted into a sand-peat mix. Other unpublished work looking at seed infectivity was reported (Price TV, 2011, *pers. comm*. – Honours Thesis of McKenna K, La Trobe University, 1982.) also showing seed to be an unlikely source of disease transmission.

Quite apart from the apparent inability of seed to act as a disease vector, seed production in Australia is undertaken in dry production regions well apart from the major commercial production regions. Additionally, the pattern of severe epidemics over the last 20-30 years is far more clearly linked to weather conditions than changes in the way seed has been produced and supplied to the market.

Soil

The persistence of the fungus in the soil is discussed under the sources of inoculum section. However, no work was located that investigated the role of soil as a dispersal agent, such as how readily disease is spread by machinery or footwear or carried on wind-blown soil particles.

Host species range

Microdochium panattonianum can infect host species other than domestic lettuce (*Lactuca sativa*). A number of alternative host species have been evaluated, in particular weeds and related crops likely to occur nearby to where lettuce are commonly grown.

Of the prospective hosts from genera other than *Lactuca*, in the family Asteraceae, Galea & Price (1988c) evaluated endive (*Cichorium endiva* L.), chicory (*Cichorium intybus* L.), and common sowthistle (*Sonchus oleraceus*). None of these species were successfully infected with conidia that did successfully infect a wide range of varieties of *Lactuca sativa*.

Of the other *Lactuca* species evaluated, prickly lettuce (*Lactuca serriola*) and willow-leaf or wild lettuce (*Lactuca saligna*) were also evaluated by Galea & Price (1988c). Both these weeds are widely distributed throughout Australia. *L. saligna* was found to be immune, and *L. serriola* highly resistant to infections by conidia that did successfully infect a wide range of varieties of *Lactuca sativa*. Nonetheless, the study confirmed that prickly lettuce could be infected by races of the disease that could also cross infect domestic lettuce and this species was therefore a real alternative host. Prickly lettuce is an autumn-spring germinating annual or biennial weed commonly found in cropping as well as horticultural situations in southern Australia. In addition to this study, one of the authors noted how he had frequently found wild specimens of prickly lettuce showing disease symptoms in various rural locations around Victoria (Galea VJ, 2011, *pers. comm.*).

Work in the USA also screened some 449 lines of both domestic lettuce as well as lines of other *Lactuca* species (Ochoa, Delp & Michelmore, 1987) for resistance to *M. panattonianum*. Other species of *Lactuca* with lines showing resistance to at least one isolate of *M. panattonianum*, were *L. augustana*, *L. livida*, L. *perennis*, *L. saligna*, *L. serriola*

& *L. virosa*. Of these, only one line of *L. saligna* was found to be resistant to all isolates tested.

Varietal resistance and race virulence

To date there is no clear evidence that any commercially available cultivars are truly broadly resistant, although susceptibility is quite varied and seems dependent both on the pathogen race and the lettuce itself - variety and crop architecture, ie, the ability of a variety to retain moisture, particularly in leaf axils.

In the USA, the varieties “Salad Bowl” (and possibly “Alaska”) were found to be resistant to some races but not to others (Ochoa, Delp & Michelmore, 1987) and a backcross program to introgress resistance from Salad Bowl was initiated. Interestingly though, while 69 lines of domestic lettuce were found to have resistance to at least one isolate of the disease, no *L. sativa* line was identified as resistant to all isolates of the pathogen.

The most comprehensive local study evaluated the resistance of every commercial lettuce line available at the time it was conducted and included 35 varieties (Galea & Price, 1988c). The study showed some significant differences in varietal susceptibility, at the extremities of the range (P=0.05). However, all *L. sativa* varieties showed some level of infection, while endive, chicory & *L. saligna* were fully resistant and *L. serriola* was highly resistant. In addition there was no discernible pattern to relative levels of susceptibility. Varieties that were less susceptible in the glasshouse, in many cases, proved more susceptible under field conditions and the converse was also true (some varieties ranked more highly in the field, rated poorly in the glasshouse). In addition, there was no clear trend to susceptibility by lettuce type (cos, iceberg etc). A single isolate only was used to compare varietal susceptibility, unlike the US study. Based on the results of Ochoa, Delp & Michelmore (1987), it is quite likely varietal rankings would have differed, had multiple strains been evaluated. Nonetheless, the study proved quite clearly that no Australian lettuce cultivars are markedly resistant to *M. panattonianum*. Galea & Price (1988c) also noted the differences in symptom expression and latent period between varieties, as well as the ability of some cultivars to outgrow infections, indicating resistance was likely to be quantitatively inherited.

The work of Ochoa, Delp & Michelmore (1987) & of Galea & Price (1988c) indicate the most likely source of resistance genes for lettuce anthracnose would likely be from *Lactuca* species other than *L. sativa*, in particular *L. saligna* & *L. serriola*.

Recent industry observations on varietal performance accords with most of the findings in these studies. Varietal differences have been noted in the field, with certain varieties now not being grown during highest risk periods. This was the case both at a nursery and grower level. Nonetheless, many growers tend to grow one type of lettuce more than others. This meant comparisons of relative susceptibilities between varieties of different types was not always reliable, as familiarity with different types was variable. One type that was consistently rated as problematic was cos lettuce. Differences reported between leafy lettuce types was highly variable, sometimes rated worse than cos types grown alongside. Iceberg lettuce was an interesting case because even susceptible varieties tended to develop symptoms on the outer leaves much more than inner leaves, which were protected after the head had formed.

This leads to another key difference noted in lettuce susceptibility, more related to plant or crop architecture than genetic resistance as such. Cos was a particularly good example of a lettuce where for much of crop development, leaves are sufficiently separated to allow water to penetrate to the axils, but insufficiently separated for good fungicide coverage with standard spray methods. Many open headed, leafy lettuce, with highly dissected leaves and great capacity to retain moisture, were reported to be extremely susceptible. Direct seeded leafy lettuce for mechanically harvested baby-leaf production is often so densely planted it is impossible for any spray to reach the lower sides of the plants.

Many seed companies attended the anthracnose workshop. While there is a clear recognition that breeding for resistance is by far the most desirable and long term solution to the problem, it is likely to be about 5 years or more before any commercial lines are available in Australia. Nonetheless, it would appear back-crossing resistant lines and trials evaluating successive selections are already underway in Australia.

One of the most important outcomes from both of these varietal susceptibility studies and from workshop discussions was the need to identify key disease races in Australia. The severity of the problem and difficulty in controlling lettuce anthracnose in Australia and New Zealand may well be related to the virulence of local disease races. The other problem highlighted by Ochoa, Delp & Michelmore, (1987) was the was the greatly differing levels of resistance in varieties when challenged by different races of lettuce anthracnose. As new resistant varieties are introduced, it will be important to establish an accurate multi-race resistance profile.

Crop maturity, vigour and physical condition

Galea & Price (1988c) noted less disease on older leaves suggesting leaf resistance increases with age. Reports of better protection from disease when calcium sprays are used would also seem to lend weight to the idea of greater cell wall integrity improving resistance. In the same way, more vigorous varieties may be more susceptible but no studies evaluating this were reviewed. Periods of vigorous “soft” growth, induced by rapid mineralisation of nitrogen as soils warm in spring were also described as a likely reason for greater susceptibility to infection (Anthracnose Workshop, May 2011).

Some growers wondered whether physical crop damage influenced susceptibility to the disease. At the workshop, the question of why disease was generally more severe in the winter-spring period than the autumn-winter period was also raised. Some growers suggested it might be related to the greater stress on the plants from colder soil in the winter-spring period. While no research relating to the influence of crop stress or physical damage on infection was found, it seems unlikely to play a major role in crop susceptibility to the disease. Galea & Price (1988a), found the disease infects the host by direct penetration of the epidermal cells or via the stomata. Crop wounding is not then necessary for infection. At the workshop pathologists suggested there was no obvious reason why wounding would increase susceptibility based on the normal method of infection described. The explanation for the greater disease pressure in winter-spring rather than autumn-winter was also thought to be much more likely a function of the lag time required for sufficient inoculum to develop and initiate an epidemic, than a function of stress.

REVIEW OF EXISTING & CANDIDATE FUNGICIDES FOR CONTROLLING LETTUCE ANTHRACNOSE

DISCLAIMER

MANY OF THE FUNGICIDES DISCUSSED IN THIS SUMMARY ARE NOT REGISTERED FOR USE IN LETTUCE IN AUSTRALIA OR NEW ZEALAND.

This report is intended as a summary of the suitability of a number of fungicides based on efficacy and residue information. Always read the label before using any of the products mentioned.

Summary

In addition to the information in the following section, other more specific efficacy and residue data relating to specific use rates has been provided to AgAware Consulting to assist with permit applications.

Key Priorities for improving control of Lettuce Anthracnose using Fungicides

Lettuce anthracnose is an intermittent disease problem in Australia, driven mainly by extended periods of wet weather. When these conditions do occur, losses can be very severe. Unfortunately there are a number of reasons why fungicides don't and in fact are unlikely in the near future to be highly effective for controlling this disease under sustained disease pressure, which are outlined in the next section.

In addition to this, there are very few effective fungicides in use for controlling the disease, either in Australia or around the world. This is largely because of the difficulty in getting fungicides to disease on well protected parts of the plant, partly because the disease is readily avoided by growing in arid locations or protected situations and partly because of the difficulty with very high residues in lettuce that result from later applications. There are also few fungicides with high intrinsic efficacy against lettuce anthracnose.

Having reviewed in detail, existing and candidate fungicides for controlling lettuce anthracnose, the following list of priorities is recommended, should lettuce growers wish to pursue them:

1. Apply for a permit to allow the use of Octave® in all lettuce other than "closed head" varieties.

2. Conduct field screening trials to evaluate a range of newer (and some older) fungicides, against existing registered Australian standards. The details on specific products and why they should be included are discussed in the next section.

3. From (2.) above, look to applying for a permit to allow the use of the most effective products in all lettuce. If, however, it is simpler to arrange a permit for leafy lettuce which is a minor rather than a major crop (closed head lettuce), then this must be the priority. The most effective product identified to date (prochloraz), is already registered for use in closed-head lettuce. On the other hand, there are no fungicides with proven high level efficacy available

for use on leafy lettuce at the moment. If necessary a permit restricted to field grown (outdoor) leafy lettuce would be adequate.

4. Apply for a permit to allow the use of a lower resistance risk protectant fungicide, such as chlorothalonil, in nurseries and very early use in the field. This would help to ensure clean seedlings arrive for field planting, that have not been exposed to the same kinds of fungicides to be subsequently used in the field.

Introduction

Control of Lettuce Anthracnose using Fungicides

Having reviewed the studies available, it became clear that efficacy was highly dependent on spray interval, lettuce type and therefore coverage and disease pressure.

Apart from product efficacy, there appear to be three key issues relating to fungicide use for the control of lettuce anthracnose:

1. Crop Coverage
2. Spray Interval
3. Crop Residue Issues

Crop Coverage

Good coverage continues to be a major problem for controlling this disease. Cos lettuce is perhaps one of the most severely affected types of lettuce because the many erect leaves are sufficiently separated to catch water and allow disease to ingress, but close enough to take a very long time to then dry out after rainfall or irrigation. Additionally this also impedes effective spray penetration, although higher application volumes may improve coverage to some extent.

Spray Interval

Studies, such as that by Wicks, Hall & Pezzaniti (1994), showed extreme differences in product efficacy when 7 or 14 day spray intervals were compared. While excellent control was achieved in most of the trials at 7 day intervals. Control at 14 day spray intervals was universally poor and in some cases hardly different to the untreated. This kind of trend is very commonly seen with fungicide sprays in horticulture, but not often is the difference quite this stark.

Most growers interviewed admitted that a 7 day spray interval was not only rarely practised (10-14 days more commonly), but it is often impossible at the most critical times, when very wet, because the ground is too slippery/soft to allow machinery into crops to spray. There were also periods of nearly continuous leaf wetness for weeks that precluded spraying for similar reasons.

Crop Residues

Lettuce, particularly leafy lettuce present particular problems for the use of fungicides. Leafy lettuce presents a crop with an edible portion that has a very high surface area/mass ratio.

There is no peeling involved to prepare the crop for consumption (although head lettuce does present some advantages here) and the entire sprayed portion is edible. As a result, residues found immediately following a fungicide application in lettuce are often in the order of 10-100 times higher than in many other vegetable crops. This often means, that extremely long withholding periods are required for residues to decline to acceptable levels, comparable with those in other crops. As a result, many fungicides have simply never been registered for use in lettuce.

A thorough search of fungicides used in lettuce production across the world has shown a strong decline in product availability in lettuce. This is particularly the case in Europe and the USA. Specific data for some fungicides is described in the next section.

While lettuce anthracnose is found globally, it appears to be very much a local problem. Nearly all the publicly available efficacy studies conducted on lettuce anthracnose have been carried out in Australia and to a lesser extent New Zealand. More generally, the same is true for most other studies of the disease as well.

To date, there are no fungicides which have been identified that can give reliable and long lived protection (greater than about 7 days) during sustained periods of high disease pressure from this disease.

Prochloraz

The most effective lettuce anthracnose fungicide available anywhere in the world still appears to be prochloraz, although it only appears to be registered for this use in Australia (closed head varieties only). Prochloraz is the only FRAC Group 3 (SBI – Sterol Biosynthase Inhibiting) fungicide used for the disease. This mode of action group works by killing the pathogen after it has penetrated the leaf but before it sporulates. As such it is the only fungicide with any semi-systemic or curative ability in use for controlling lettuce anthracnose.

In one of two trials in the early 1990's (Wicks, Hall & Pezzaniti, 1994), applied preventatively every 7 days, under field conditions in head lettuce, prochloraz was found to be more effective than several other "candidate" SBI fungicides, including hexaconazole, flusilazole, and penconazole. Propiconazole and difenoconazole were both found to be numerically less effective but not significantly so. Propiconazole, however, showed crop stunting effects (phytotoxicity), and so discounted from any future work. It is worth noting that the rate of prochloraz used was around twice that of the currently registered rate in Australia. However, use rates for these other SBI fungicides were similar to typical use rates for diseases in other crops. Later trials in the same study showed efficacy with this higher rate was not significantly to the current label rate.

Use of FRAC Group 3 (SBI) fungicides on crops other than those appearing on product labels, have long been associated with these kinds of stunting effects and seems to be most often seen in those that are most readily taken up and mobilised inside the plant towards the apical meristems. The occurrence of this apical stunting is also very dependent on individual crop species. It is noteworthy that prochloraz belongs to the imidazole chemical group, whereas the other SBI fungicides evaluated belong to the triazole chemical group.

In another trial, in part of the same study, prochloraz was again shown to be more effective at inhibiting spore production and germination than propiconazole, tebuconazole, triadimenol and myclobutanil, although not always significantly so.

Worthy of re-examination in this original list is difenoconazole and tebuconazole.

Prochloraz has also proven to be the most effective fungicide for lettuce anthracnose in New Zealand trials (Broadhurst & Wood, 1996), although it is still not registered there for this use. The same observation is reflected in comments from interviews conducted with agronomists and growers in both Australia. Although still not registered for this use, it is also the only fungicide available for controlling lettuce anthracnose in the UK, under its SOLA (Special Off-Label Approval) permit system.

With a 7 day WHP, this makes prochloraz the most valuable fungicide currently available, for controlling anthracnose in closed head lettuce in Australia.

Taint

There is however, one very significant limitation with the current Octave® label. It is limited to use on "closed head" type lettuce. This takes the single most effective product away from any farmers growing "open head" type lettuce, which as described earlier, are an increasingly important proportion of the lettuce consumed.

Consultation with Bayer CropSciences Australia (Geoff Robertson & Anthony DeMonte *pers comm*, April 2011) indicates the history behind this is likely related to studies conducted in the 1980's & 1990's showing the potential for prochloraz to cause a flavour taint in some crops. The logic in registering in "closed-head" varieties only, presumably relates to residues from later sprays being mostly confined to the outside leaves, which are to a greater or lesser extent trimmed after harvesting, before being placed on display in markets.

Taint studies in lettuce have been conducted in the UK previously, but the intellectual property for these now resides with BASF and so not readily available to Bayer. BASF via Nufarm in Australia, have also been approached to see whether it may be possible to get access to this data, to help support a broadened use pattern in Australia for lettuce, but no information has yet been received. Elsewhere, prochloraz has been linked with taint issues in;
strawberries
http://books.google.com.au/books?id=W6ToDne5AsMC&pg=PA77&lpg=PA77&dq=prochloraz+taint+strawberries&source=bl&ots=83Jqqjf-7G&sig=62noG_wlxh8fT57p9JIt8C23hAY&hl=en&ei=C7OrTc2uOs_NrQfH7YynCA&sa=X&oi=book_result&ct=result&resnum=1&ved=0CBUQ6AEwAA#v=onepage&q&f=false
lychees
http://www.australianlychee.com.au/items/38/RIRDC%2001-163.pdf
coffee
http://www.researchkenya.org/?ID=6083&search=Fungicides
and most recently, in dragonfruit
http://www.chinapost.com.tw/taiwan/national/national-news/2008/10/12/178310/Tainted-dragon.htm

Taint is believed to be associated with a number of breakdown metabolites resulting from the parent prochloraz molecule, including 2, 4, 6-trichlorophenol (TCP). Trichlorophenols are readily transformed to the corresponding trichloroanisole (TCA) (Onani, 1995). The formation of TCA is believed to be associated with breakdown of prochloraz during wet weather or in a damp environment (Holscher et al, 1995).

There appear to be two key sources of the taint metabolite (Bayer AU, *pers. comm*.):

1. As an impurity in the active ingredient (only generally an issue with generic sources)
2. Microbial breakdown of prochloraz to the taint metabolite.

Keeping in mind the latter point, the agronomic practices, handling and processing of the sprayed produce can be pivotal in determining if a taint will be present, or if metabolic breakdown of the prochloraz residue follows a different route (Bayer AU, *pers. comm*.).

Were it not for the complications of this issue it would be worth applying for a permit that mirrors the UK SOLA use pattern without any new data (efficacy or residue). The worst case would be an application fee and rejection. However, as addressing the taint issue appears to be a matter of necessity, it is probably worth ensuring adequate efficacy and residue trials are simultaneously undertaken to ensure the approval if the taint is proven not to be a problem, following testing.

Chlorothalonil

Based on the most extensive fungicide study to date (Wicks, Hall & Pezzaniti, 1994), The second most effective fungicide, when applied on a tight schedule of protectant sprays (7 day intervals) was chlorothalonil. Despite this, chlorothalonil has never been registered or used under permit in lettuce in most countries in the world, including Australia. This is likely linked to the high use rates as well as the visibility and tenacity of residues.

Chlorothalonil is registered for use against lettuce anthracnose in New Zealand. However, reports from NZ agronomists and growers still indicate they are getting poor control with chlorothalonil under field conditions, during high disease pressure periods. The superiority of prochloraz was also shown in a New Zealand study conducted in the mid 1990's (Broadhurst & Wood, 1996).

Dithiocarbamate (FRAC Group M3) Protectants

Mancozeb

Maneb & the zinc complexed form of it (mancozeb), have been extensively registered and used in many crops including lettuce around the world, more likely due to the age (mancozeb was first registered in the USA in 1948) of these fungicides than because of other factors. However, it would appear some significant uses in the USA were voluntarily discontinued (many vegetable crops, including lettuce) in or around 2009.

Mancozeb does have intrinsic activity against lettuce anthracnose (Wicks, Hall & Pezzaniti, 1994 & Parman, Price & Lee, 1991 & Tan, 1983). Despite registration for use against lettuce anthracnose in Australia, mancozeb has generally given poorer disease control than prochloraz or chlorothalonil, in outdoor situations, under serious disease pressure (Wicks, Hall & Pezzaniti, 1994 & Broadhurst & Wood, 1996), and from discussions with growers and agronomists in Australia & New Zealand.

While it performed poorly when applied as a an unbroken sequence of (approximately) weekly sprays, it did prove to be effective as part of an alternating weekly spray program with prochloraz (Broadhurst & Wood, 1996). It has also been shown to be extremely effective in protected crop experiments (Broadhurst & Wood, 1996 & Parman, Price & Lee, 1991 & Tan, 1983). The markedly better performance in protected cropping suggests that perhaps the product is easily washed off with water (eg. rain or overhead irrigation). Mancozeb currently has a 14 day WHP in lettuce in Australia.

Propineb
Like mancozeb, propineb is an older fungicide belonging to the dithiocarbamate class of chemicals (FRAC Group M3). It has been registered widely throughout the world in vegetables including lettuce, but like mancozeb does not seem to be available any more for use in lettuce in the USA. While it is used for controlling lettuce anthracnose in other countries, it is not registered for this use in Australia. It is however registered in lettuce in Australia for controlling downy mildew. While there is not a great deal of rigorous data available, efficacy against lettuce anthracnose appears broadly similar to mancozeb (Wicks, Hall & Pezzaniti, 1994). Propineb has a 3 day WHP in lettuce in Australia.

Thiram
Like mancozeb, thiram is an older fungicide belonging to the dithiocarbamate class of chemicals (FRAC Group M3). It has been registered widely throughout the world in vegetables including lettuce, but like mancozeb does not seem to be available any more for use in lettuce in the USA. Thiram is registered for use in lettuce to control anthracnose but there is very little data on its activity and it does not appear to be widely used for this purpose. The only formal evaluation cited showed similar activity to mancozeb under field conditions (Parman, Price & Lee, 1991). Thiram has a 7 day WHP in lettuce in Australia.

Captan

Captan is not widely used in lettuce globally and is not registered for use in lettuce in Australia. However, it is registered for control of “leaf spot” in lettuce in New Zealand and has a 7 day WHP. The use rate is also very high, compared with typical use rates in other crops in Australia. The only results cited suggest it was significantly less effective than prochloraz, had similar activity to chlorothalonil and copper hydroxide and was better than mancozeb (Broadhurst & Wood, 1996). The registered uses of Captan appears to be generally diminishing globally. The highest MRLs set are generally in the Asia-Pacific region, with much lower tolerances in the Europe and the USA.

The related fungicide captafol was found to have similar activity to mancozeb in a single glasshouse trial (Parman, Price & Lee, 1991). However, this fungicide is no longer available in many parts of the world, including Australia and the USA, and elsewhere appears to have had its uses restricted.

Efficacy data is not sufficiently compelling to justify the use of resources for pursuing a permit for Captan in lettuce in Australia.

Non Triazole DMI Fungicides

As prochloraz is an imidazole class chemical, not a triazole like most of the other DMI fungicides evaluated, it is worth seeing what other non-triazole class DMI’s may be worth evaluating for activity on lettuce anthracnose:

Fenarimol
Fenarimol is a pyrimidine class chemical. However, poor efficacy in Wicks, Hall & Pezzaniti (1994), suggests there is little value in re-evaluating this fungicide.

Triforine
Triforine is a piperazine class chemical. However, it has already been shown to be clearly phytotoxic in lettuce (Maxon Smith JW, 1979).

Triazole DMI Fungicides

Flutriafol

Flutriafol is mainly used throughout the world in wheat, barley & canola. Flutriafol is not known to be registered for use in lettuce anywhere in the world. However, it was shown to have high systemic activity on lettuce anthracnose compared with other DMI fungicides (including prochloraz) when applied as a soil drench, 48 hours prior to an infection event (Parman, Price & Lee, 1991).

This of interest because flutriafol, which has historically been used mainly as a seed treatment product for cereals and canola, recently extended this use pattern in Australia into cereal crops as an in-furrow fertilizer treatment. Applied to granular fertilizer at planting, and gradually accessed by developing roots, flutriafol has been shown to give very long lasting protection (often up to the beginning of jointing) in cereals against diseases such as stripe rust. By gradually absorbing the fungicide through the roots rather than the leaves, plants seem to achieve continuous protection of both existing and new growth alike.

While this would appear to offer the prospect of a very elegant means to controlling disease there are significant other considerations very likely to preclude such a use:

1. Firstly, the efficacy of such a treatment has not been clearly demonstrated in field trials. An effective dose and best means of delivery (on fertilizer or as a soil drench, etc) needs to be properly explored.

2. It is quite likely that flutriafol could prove phytotoxic, and cause severe stunting in lettuce. This stunting potential of DMI fungicides has already been described and tends to be associated with more mobile members of the group.

3. As this represents a very novel application method in lettuce to controlling the disease and not merely the extension of a common foliar use pattern, there may be significant regulatory obstacles. Even if effective, such a use may be required to be registered rather than legalised through a permit.

4. There are no established MRL's for flutriafol in lettuce, other than a very low leafy vegetable MRL in the European Union, presumably to cover incidental use.

5. There are likely to be significant resistance management concerns raised. Such a use pattern puts the fungicide under continuous resistance selection pressure for as long as an effective dose is taken up by the plant. The continuous dosing of the active ingredient, with no break may well preclude the foliar use of any other DMI fungicide, including prochloraz over the life of the crop.

Difenoconazole

Difenoconazole is not registered for use in lettuce in Australia but does appear to be registered for use in lettuce in Spain (for *Alternaria spp.* control) and Brazil (for *Septoria lactucae* control). This triazole is of interest, mainly because it is likely to be well tolerated in lettuce and was shown to have activity against lettuce anthracnose (Wicks, Hall & Pezzaniti, 1994). In this study it showed less activity than prochloraz but still compared well with other DMI fungicides evaluated.

Tebuconazole

Tebuconazole does not appear to be used in lettuce globally, although there is a very low leafy vegetable MRL in the European Union, presumably to cover incidental use. Tebuconazole is already registered for use in lettuce in Australia, for the control of sclerotinia, but only in field grown crops (not in protected or hydroponic lettuce crops) and has a 5 week WHP. It is of interest principally from its inclusion in two of the trials conducted by Wicks, Hall & Pezzaniti (1994). In the first trial, after a series of weekly applications, it was the next most effective fungicide for reducing disease damage after prochloraz (not significantly different), although it was a less effective anti-sporulant. In the second trial tebuconazole showed similar curative activity to prochloraz.

Prothioconazole

Prothioconazole does not appear to be used in lettuce globally. There is a very low leafy vegetable MRL in the European Union, presumably to cover incidental use. The interest in including it in future screens is based solely on it being a relatively new addition to the triazole group, not on any evidence from biological activity screening.

Epoxiconazole

Epoxiconazole does not appear to be used in lettuce globally. There is a very low leafy vegetable MRL in the European Union, presumably to cover incidental use. The interest in including it in future screens is based solely on it being a relatively new addition to the triazole group, not on any evidence from biological activity screening.

QoI (FRAC Group 11) Fungicides

The Quinone outside Inhibitors (also generally referred to as the “strobilurins”, after the fungal genus *Strobilurus*, where these chemicals were first discovered), are the most significant new family of fungicides to have appeared since the DMI fungicides, and in conjunction with their generally broad spectrum efficacy considered one of the most likely candidates to evaluate for activity against lettuce anthracnose.

Unfortunately there are very few detailed or reliable studies to date, particularly in any published work that have looked at lettuce anthracnose activity. Some of these already have global registrations for control of other anthracnose diseases in other crops, particularly for controlling *Colletotrichum* species, but almost no specific registrations for the control of lettuce anthracnose.

Azoxystrobin

Azoxystrobin, in fact seems to be the only QoI fungicide currently registered specifically for the control of lettuce anthracnose (*Michrodochium panattonianum*), anywhere in the world (Current Quadris® & Amistar® labels in the USA). As a result, there has been a good deal of expectation, that this might be the most effective new control option. However, the only data cited, provided by Syngenta USA, showed that after a sequence of 4 weekly sprays, azoxystrobin performed similarly to mancozeb and captan under moderate disease challenge in both head and cos lettuce (one trial in each). In fairness, there were no other treatments that showed other fungicides (like prochloraz for example) giving high levels of control. It is therefore hard to know whether, the lack-lustre level of control could be directly attributed to products or whether there were complicating factors such as poor coverage.

Growers who have been using Amistar® under a current permit for sclerotinia suppression, have not reported any associated benefits in better control of anthracnose.

Pyraclostrobin

There is even less efficacy information available for pyraclostrobin. While it is widely used in lettuce throughout the world, anthracnose (*Michrodochium panattonianum*), is not on any of the labels cited, although *Colletotrichum spp*. are listed on some labels, such as the Cabrio® label in the USA, under non-Brassica leafy vegetable diseases.

The only solid evidence of activity to date comes from some very recent controlled environment work conducted in South Australia, as part of HAL VG07127, which has been submitted but not yet published. This work indicates pyraclostrobin is one of the most effective new products, but caution is advised until these results are demonstrated in field trials (Mancozeb often looks very good in glasshouse studies but then shows poor performance in the field).

This situation is a major set-back to improving lettuce anthracnose control with fungicides, as there are relatively high MRLs set in lettuce globally, including Codex MRLs, for both azoxystrobin & pyraclostrobin.

Further Efficacy Trials

There would be great value in repeating the kind of outdoor efficacy work conducted by Wicks, Hall & Pezzaniti (1994), but including a range of prospective new options. Fungicides recommended for inclusion and prospective use rates have been summarised along with a protocol for field trials and sent to AgAware Consulting for future reference.

Summary and key outcomes of the industry workshop – Werribee 2011

The workshop organised by Applied Horticultural Research (AHR) was attended by 56 delegates including growers, seed companies, nurseries and other industry representatives.

The meeting was strongly supported by the research community, with all the major researchers involved with this disease present, including: Prof. Terry Price, Prof. Vic Galea, Dr Trevor Wicks, Dr Len Tesoriero, Dr Liz Minchinton, Dr Hoong Pung and leafy vegetable expert, Mike Titley.

Key outcomes of the workshop

1. Greatly enhanced understanding of spore dispersal by wind and water splash and infection.

Professor Terry Price presented his work on the distance spores can travel assisted by wind or water splashing (rainfall or overhead irrigation), and the effective crop separation distances required to minimise the spread of disease from infected to newly planted (clean) lettuce crops. A minimum distance of about 10 m (a typical production "bay"), between all lettuce crops, is likely to be highly effective at preventing the spread of the disease.

Prof Vic Galea discussed infection and pointed out that the optimum conditions for infection are a temperature of 15°C and 8 hours of leaf wetness, but that infection could occur in the leaf axils without leaf wetness requirement provided disease inoculum was present. He also said spores remain viable in the soil for 18 weeks and can last 78 weeks on dry crop residues, making carryover of inoculum between crops a significant issue.

2. Cultural Controls

Soil health (microbial activity) and the presence of crop residues may have a major effect on disease carryover and infections of subsequent crops.

Data was presented on the use of other control agents, particularly calcium products indicated this might be a much more productive path to pursue as they do not face the same regulatory requirements of conventional fungicides.

There was agreement that cultural controls are going to be a key area for disease management in medium term in the absence of genetic resistance or new fungicides.

3. Fungicides and other sprayed control agents.

Fungicides are not providing effective control, especially under high disease pressure, and this is related to the difficulty in getting the product into leaf axils where the disease tends to start.

Prochloraz is the most effective conventional fungicide for controlling the disease but can only be used in “closed head” varieties and high residues are likely to seriously limit new permit uses of most fungicides in leafy lettuce.

The best prospects for newer fungicide chemistry lies with the new strobilurin fungicides, which have high and widely established global tolerances in lettuce (MRL’s). Unfortunately, efficacy data on these chemicals is very limited.

AHR has prepared a data package to support permit applications.

Peter Dal Santo proposed a 3-point strategy for the short term:

1. Permit for Amistar adding anthracnose to existing sclerotinia permit.
2. Permit for prochloraz on leafy lettuce.
3. Permit for chlorothalonil for nursery use and field use with a long with holding period.

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Acknowledgements:

AHR would like to thank the many lettuce industry people consulted over the course of this project, in particular the following:

Plant Pathologists: Prof. Terry Price, Prof. Victor Galea, Dr Trevor Wicks, Dr Len Tesoriero, Dr Hoong Pung, Dr Elizabeth Minchinton.

Growers: Ian Willert, Wayne Stockton & Jeff Beavis (Boomaroo); Ed & James Fagan, Paul Gazzola, Andrew Bulmer, Lawrence Cowley, John Mirrabelli, Garry Yee (NZ).

Agronomists & Consultants: Mike Titley, Peter Aird (ServeAg), Karl Riedel, Gary Graham (Pukekohe Growers Supplies, NZ), Marc Hinderager (Elders), Paul Wyneham (EE Muir & Sons).

Chemical Industry: Peter Dal Santo (AgAware); Geoff Richardson, Anthony DeMonte (Bayer Crop Science); Nicholas Bennett, Brian Cassar & Rob Battaglia (Syngenta); Doug Wilson (Nufarm); Peter Nott (Dow AgroSciences); Tim Hammond (Du Pont).

Seed Industry: Dan Trimboli (ex. Enza Zaden & Yates); Steve Mitchell (Enza Zaden); Steve McCraith, David McDonald (SPS); Alan Mackinley (Lefroy); Harry Turna (Rijk Zwaan).

Technology Transfer

The following technology transfer activities have been carried out:

1. An industry wide national workshop was held on the 4th May 2011, at Werribee, Victoria and was attended by 56 delegates including growers, seed companies, nurseries and other industry representatives.

2. A best practice management guide has been produced for growers and industry professionals on the latest techniques for the control of Anthracnose in lettuce. This has been emailed to lettuce growers and made available on the AUSVEG and AHR websites.

3. A review of the literature and research on biology, epidemiology and fungicide control options available for Anthracnose has been conducted and is included in this publicly available report. A summary of the key findings of this review has been presented in the best practice management guide.

4. An extended summary of candidate fungicides and issues, residue and efficacy data and trial protocols which would assist in the preparation permit applications has been produced and passed on to AgAware Consulting which handles the minor use crop applications to APVMA for the horticulture industry.

Recommendations

The following areas were identified as having potential for further research into developing methods for the effective control of Anthracnose in lettuce.

Cultural Controls:

- Confirm separation distances and crop rotations suggested by the current research on infection and spread using common lettuce types.
- Investigate the impact of soil health and organic matter on the disease. Current information suggests a good link between persistence and the level of microbial activity (High activity gives shorter persistence).
- Investigate the importance of properly burying crop residues on disease carry carryover.
- Evaluate Ca products e.g. Folical, calcium nitrate, Serenade, Bion to determine efficacy under field conditions and use rates.
- Investigate the impact of crop nutrition (esp. nitrogen) on susceptibility to infection and spread.

Varieties:

- Breeding new resistant lines in the longer term (5 years to first commercial lines).
- Evaluate relative levels of susceptibility in current main lines.

Plant Pathology:

- Identify the range of *M. panattonianum* pathotypes occurring in Australia and their relative virulence on a range of varieties.

- Develop a rapid and reliable laboratory method (eg. molecular probe) to track the fungus in the environment.
- Determine whether microsclerotia are an important source of disease inoculum across a broad range of growing regions in AU (Previous research in Victoria suggests not but found to be important in California).
- An epidemic forecasting tool based on temperatures and periods of leaf wetness.

Fungicides

- Submit permit applications suggested by Peter Dal Santo
- A field efficacy study on selected new fungicides against the existing benchmark, prochloraz. A comprehensive review of candidate fungicides has been sent to Peter Dal Santo. The longer list can be trimmed to about 5-6 key entrants. This should be undertaken before committing to residue studies to extend permits for fungicides in leafy lettuce.
- A residue study (multiple trials in leafy lettuce, including babyleaf) to evaluate the persistence of prochloraz (local and half local use rate).
- A residue study (multiple trials in leafy lettuce, including babyleaf) to allow a permit for the nursery use of chlorothalonil (very long withholding period), to assist with control and disease resistance management in nurseries.

Appendix 1. Notes collected from the workshop

1. Disease Reaction and Cultural Controls

INFECTION AND THE DISEASE TRIANGLE
Host --- Pathogen --- Conditions

- Anthracnose is an aggressive pathogen
- The optimum conditions for infection are a temperature of 15°C and 8 hours of leaf wetness
- Infections in the leaf axil do not need wet conditions on the outer leaves.
- The spores are spread by water splash
- Spores remain viable for 18 weeks in the soil and last **78 weeks** on dry crop residues
- The key time for infection is springtime coming out of winter. This coincides with the changing weather patterns, often longer periods of leaf wetness occur at this time of the year which favours infection. In addition there can be a build-up of inoculum during winter which would assist infection in spring.
- The spring period also coincides with the mineralisation of nitrogen and subsequent release of nitrate encouraging soft growth more prone to infection.
- Soil health and soil organic matter may have an impact on the disease
- Properly burying crop residues may have a beneficial impact on reducing disease carry carryover
- The disease can occur in nurseries and in field crops
- Have not found micro scerotia in Australia
- Spread is usually down the rows

Wind is a significant factor in the spread of spores. Wind blown rain is effective at spreading the disease. Higher winds → greater spread.

SUGGESTED CULTURAL MANAGEMENT PLAN (Vic Galea)

1. Nursery hygiene
2. Trash management in the field
3. Resistant cultivars
4. Crop rotation and weed management
5. Fungicides

CROP ROTATIONS

- The question is should there be a gap of 20 weeks or four years between an infected crop and the planting of the subsequent crop. This depends on the type of anthracnose storage structure (micro sclerotia) that we have in Australia.
- The question of separation distances between plants need to be determined and this is based on how far the disease organism can spread. The suggestion is to separate crops by the width of one bay or 10 m.
- The other strategy is to plant new crops up wind of infected crops.

PREDICTION MODEL

- Because leaf wetness is not a critical factor for infection in the leaf axil, it is doubtful whether a prediction model can effectively predict infection.
- The effectiveness of a prediction model is further challenged by the lack of appropriate fungicides to control this disease.
- Vic Galea and Liz Minchinton have worked on a anthracnose prediction model, or are able to do this work if required.

SEED

- The disease is not seed transmitted
- What seed treatments are currently being used ?

NURSERY

- Q. From a grower - Does physical damage plants to make them more susceptible to infection by anthracnose (No – Vic Galea's explanation)
- Overhead fans seem to be effective at drying leaves and reducing the incidence of the disease (in nurseries)
- Spore kill (Didecyldimethylammonium Chloride (DDAC) and chloride not prove effective at controlling infection (Dr Hoong)
- Boomaroo nursery currently trialling alternative control methods

IRRIGATION

- Trial plastic mulch with trickle
- Expense of plastic mulch to high

WEEDS

- Prickly lettuce resistant to enter it is but can be infected
- Wild lettuce is not susceptible?

SOURCE POINT INFECTIONS

- Economics
- Scouting need more information on threshold levels
- What are the criteria for disease scouting
- There is a soil test possible for detecting anthracnose which can infect at very low soil inoculum levels of 6.5 colony forming units (cfu) (?) per gram of soil.

NUTRITION

- Nitrate release in Spring is a problem. The issue relates to rapid release of nitrate in the soil following a temperature increase in spring. Leads to soft tissue and lettuce plants being more readily infected.
- Calcium: there has been promising results from the use of calcium products to toughen the leaf surface. This work could fit in with general studies on cropping nutrition at the impact on anthracnose infection in lettuce.

PROTECTED CROPPING

- The group did not consider there was great potential for protecting cropping for the production of lettuce.
- This aspect relates only to the nursery production of seedlings. It could potentially relate to production of baby leaf lettuce under structures.

2. Fungicides

ISSUES

- Residues required for efficacy but a problem for safe use close to harvest, especially if no MRL is established – withholding periods.
- Spray coverage
- Products specific for nursery use
- Develop a list of candidate fungicides (TM and PDS) to provide
- Thiram – there is a label for anthracnose on lettuce – effective
- Don't exclude chlorothalonil

SHORT-TERM STRATEGY suggested by Peter Dal Santo

1. Obtain a permit for Amistar - add anthracnose to existing sclerotinia permit
2. Obtain a permit for prochloraz on leafy lettuce
3. Obtain a permit for chlorothalonil four nursery use and field use with a long (eg. 35 day) with holding period.

- Octave is expensive and has major residue issues in leafy lettuce
- Trevor Wicks investigated a whole range of alternative new products as well as conventional fungicides. Work to be published with Liz Minchinin.
- More work is needed on evaluating new fungicides for effective control.

3. VARIETIES

- Conditions which favour anthracnose infection also favour other foliage diseases such as downy mildew. Need combined disease resistances.
- There are many competing demands for breeding lettuce. Anthracnose is on the list but competes with other important diseases such as Downy mildew (downy & nas resistance are global priority 1; anthracnose = 3)
- Under high disease pressure all varieties appear to be susceptible
- In the United States five selections have been identified with resistance to the disease (Steve Mitchell – Enza-Zarden)
- In general terms we are probably looking at field resistance rather than full genetic resistance (Enza-Zarden

From Rijk Zwaan

- There is some resistance available and at this stage in the F4 stage
- There are no marker genes available for the resistance which makes selection more difficult
- There will be a review of the variety ***sniper*** to check resistance to anthracnose
- Anthracnose is the number three issue after downy mildew and lettuce aphid

- We need formalised field trial to properly assess resistance to anthracnose not simply observation

Comment from Mike Titley:

- When cos and iceberg lettuce are planted together, there are more infections on the iceberg which originate on the cos lettuce. This means it is important to separate the two types of lettuce in field plantings. Cos lettuce are more susceptible to the disease than iceberg.
- At this stage cultural controls appear to be more important than chemical controls or varietal.

3. Research Issues

- Large fungicide screening trial, 18 months of screening work which is a longer-term control. We can't apply for an emergency APVMA permit application.
- There appears to be a project plan already developed and submitted between PDS and HAL.
- Varieties: test against isolates from different states to get an idea of the distribution of strains within Australia and varietal sensitivities.
- Evaluate non-fungicide controls such as Folical, Serenade, Bion
- Develop a prediction model based on leaf wetness, humidity and temperature
- Investigate the impact of soil health and organic matter on the disease
- Investigate the importance of properly burying crop residues on disease carry carryover
- Potential to develop a molecular probe to determine the presence of anthracnose (Len Tesoriero)
- Investigate alternative controls - e.g. Folical looks promising
- Leaf wetness monitoring
- Develop a risk management strategy to contain outbreaks
- Understand how anthracnose control fits into the bigger picture of foliage disease management