A Report On Mundulla Yellows In Australia

Mundulla Yellows Task Group - May 2004

EXECUTIVE SUMMARY

Mundulla Yellows (MY) is a fatal disease that affects eucalypts and other native plants. It is characterised by progressive yellowing and dieback of foliage. It was first observed in the vicinity of Mundulla, South Australia in the 1970's. MY has been widely documented in South Australia, where it seems to be spreading and has been observed in varying degrees in other States. To date, its cause(s) is not known. The Mundulla Yellows Task Group (MYTG) comprising nominated plant experts from all States was set up in September 2002 by the Land, Water and Biodiversity Committee (under the Natural Resource Management Ministerial Council) to determine (1) the current status of research, (2) a risk management approach for the problem, (3) a flexible research strategy and (4) a funding strategy for research and management activities.

(1) Status of current research into the cause and spread of Mundulla Yellows: It is not clear whether Mundulla Yellows is caused by biotic (living) agent(s), or abiotic (environmental) factor(s), or a combination of both. In terms of biotic causes, unusual nucleic acids (MY-RNAs) were found to be associated with MY-affected Eucalypts. Their size and biochemical characteristics indicate that they may be associated with a virus or viroid.

Research into possible abiotic causes of MY indicates that environmental factors (such as applied chemicals, soil type, moisture levels, physical disturbance) may either be the cause, or predispose vegetation to infection by a biotic causal agent(s), or may interact with the causal agent(s) to exacerbate, or sometimes suppress, disease symptoms and damage. Surveys are also being carried out to investigate the distribution and spread of MY in relation to site parameters.

(2) Risk Management: In May 2003 Broadleaf Capital International Pty Ltd (commissioned by the MYTG) conducted a Risk Management Workshop to review research on the causation and epidemiology of Mundulla Yellows, review research directions, carry out a risk analysis, and consider possible management measures. The current distribution of MY in Australia was examined and four risk scenarios for further spread were evaluated. These scenarios addressed MY spread to and within Australia's: (a) local government areas; (b) plant nursery industry; (c) world heritage areas/national parks and state forests, and (d) forestry industry. Analysis of the four scenarios by Broadleaf Capital International Pty Ltd showed that without a clearer understanding of disease causation and epidemiology, and a reliable diagnostic indicator for MY, the unmitigated risk associated with this disease is considered unacceptable and intolerable. However, the MYTG considered only two of these 4 scenarios (a and b) to be unacceptable based on current information.

(3) Research strategy: The following 8 areas have been identified by the Task Group as future MY research priorities: (i) identification of causal agent(s) and development of a diagnostic test, (ii) modes of transmission and spread, (iii) environmental factors and predisposition, (iv) systematic characterisation of symptoms, (v) effect of disease on host – biochemical, physiological and anatomical, (vi) systematic determination of local and national distributions and host range, (vii) genetic resistance or tolerance, and (viii) risk assessment.

(4) Funding strategy for research and management activities: The Task Group is unanimous in advising that, based on the available information, MY is a serious new plant disease threat that must be addressed. Basic work is urgently required to define the problem and identify its cause(s), mode(s) of spread, and develop a diagnostic test.

There are two primary areas of research underway by The University of Adelaide (Waite Campus) (SA) and the Department of Primary Industries, Knoxfield (Vic) that are yielding promising results and which the MYTG believes should continue. The MYTG recommends every effort be made to promote synergies between the two research programs and minimise unnecessary duplication to maximise the utility of the research. This will be best facilitated through the continuation of the MYTG with revised terms of reference and possibly expanded membership. This facilitation should be supported by the establishment of a dedicated website which promotes links between MY stakeholders.

Once more is known about the disease it will be appropriate for regions where Mundulla Yellows is identified as a problem to seek funding for further research and for their management of the problem, through existing channels. At that time it would be useful to revisit the composition of a multi-jurisdictional review group to coordinate research and facilitate communication and the development of management strategies.

Realistic estimates of the threat posed to different stakeholders, and therefore their willingness to fund research, are not possible until a causal agent(s) is identified. The MYTG believes under these circumstances it is difficult to identify further non-government funding sources. In addition, the MYTG is of the view that no definitive recommendations concerning disease management can be made until the causative agent(s) has been determined.

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1. BACKGROUND AND STATUS OF RESEARCH

The Land, Water and Biodiversity Committee (under the Natural Resource Management Ministerial Council) established the Mundulla Yellows Task Group (MYTG) in September 2002 comprising nominated plant experts from all States and the Australian Government. The terms of reference of the MYTG are given in Appendix 2 and this report details their findings.

1.1 Background to Mundulla Yellows[1]

Mundulla Yellows (MY) is a disease of eucalypts and other native plants that is characterised by progressive yellowing and dieback of foliage, resulting in the death of the affected plants over several years.

MY was first observed in the vicinity of Mundulla, South Australia, in the 1970's. The earliest visible symptom on plants affected by MY is interveinal chlorosis (yellowing of the leaves, while their veins and adjacent tissues remain green). This begins at the periphery of the plant, usually on a single branch, which then dies back. Epicormic growth develops lower on the affected branches, the leaves of which also yellow and die. These symptoms gradually spread through the plant, causing the whole canopy to die back over several years or decades. The disease is always fatal. To date there has been no success in re-establishing healthy plants on sites with Mundulla Yellows dieback.

The cause of MY has not yet been determined. For the purpose of this report MY will be referred to as a disease, on the understanding that the term "disease" refers to the abnormal functioning of a plant. A plant disease may be caused by pathogenic micro-organisms, insect or other pests, unfavourable environmental, genetic, or nutritional factors, or a combination of these.

This disease has several unusual attributes that are worth noting. The combination of slow, progressive expression, early stage symptoms of branches with interveinal chlorosis interspersed with healthy limbs, the presence of healthy trees and shrubs amongst dead and dying trees, and the lack of recovery of affected plants are unique amongst currently understood abiotic and biotic diseases of eucalypts and other native plants (see Appendix 3). The disease is unusual in the broad range of taxa apparently affected. In addition, the presence of the disease at a site appears to be long-term. When trees are removed after they die from Mundulla Yellows, and the site is replanted with healthy trees, the new plantings also develop the disease and die.

Since the disease was described, the symptoms have been reported in trees of all ages. Ongoing surveys have documented the spread of the disease over the last ten years and it has been reported in all Southern Australian states.

There has been extensive scientific and community concern about the potential environmental and economic impacts of this disease. In response to these concerns, a workshop was organised in April 2002 by the South Australian Department for Environment and Heritage and the Australian Government to review research and survey programs that had been undertaken. The workshop recommended the development of a comprehensive and strategic research program and risk

[1] The MYTG acknowledges the information contained in this background was drawn from the reports and papers listed in the bibliography (Appendix 1).

management framework. There was agreement at this forum that the issue of highest priority was identification of the causal agent(s) of MY.

As a follow-up to this meeting, a Risk Management workshop was convened by the MYTG in May 2003 with an invited team of plant experts from all States. This workshop reconfirmed and refined the research priorities outlined in the previous April workshop. These priorities are discussed in part three of this report.

1.2 Distribution

Geoff Cotton, an apiarist based in Keith, South Australia, documented the first MY symptoms in eucalypts in approximately 1975. In South Australia since 1992, MY symptoms have been observed from Mt. Gambier north to Gammon Ranges, on Kangaroo Island, and west to Ceduna. The disease appears to occur across a range of climatic zones and soil types. To date it has not been found in the high rainfall and high topographic regions of the Mount Lofty Ranges. There appears to be a strong association between the presence of MY symptoms and vegetation on roadsides, and/or in areas that had been disturbed in some way by humans, such as urban parklands, farm paddocks, quarries, cemeteries and car parks.

To date at least 87 species of 29 genera have been documented as expressing MY-like symptoms in South Australia. Many of these species are amenity plantings outside of their natural range of occurrence in Australia and some are non-Australian species (see Appendix 3 for a complete list of suspected host species). However, doubts have been raised about the consistency of symptom expression, even within the genus *Eucalyptus*. Therefore, it is important to note that there is no robust evidence to confirm beyond reasonable doubt that other species are affected by MY. This cannot be done until a routine diagnostic test is available, or the cause of MY elucidated.

Symptoms of MY have been observed in over 30 species of eucalypts at a number of locations in all states in southern Australia. Outside Australia, MY-like symptoms have been observed in eucalypts grown in southern Spain, and on the Spanish island of Ibiza. Surveys that have been undertaken within areas where MY-affected trees are present, indicate that the disease appears to spread to adjacent asymptomatic trees at varying rates.

Symptom expression may vary in intensity and timing between affected species and between sites. Various other plant disorders can also cause degrees of yellowing and dieback. Additionally, if the cause is biotic, it is probable that non-symptomatic, but potentially infected plants, occur in natural and revegetated landscapes. In these circumstances, single observations of symptoms alone are not a reliable basis for diagnosing MY. Due to inconsistencies in the expression of symptoms in eucalypts at the national scale, further validation of the occurrence of the disease in specific eucalypt species, as well as in other plant genera, is required. It is therefore necessary, if the cause is biotic, to develop an efficient routine diagnostic test before further large-scale survey efforts are undertaken to ascertain both the distribution and host range of the disease.

1.3 Status of Research into Causes

It is not clear whether Mundulla Yellows is caused by biotic (living) agent(s), or abiotic (environmental) factor(s), or a combination of both. Information for this report has been drawn from work listed in the bibliography and from unpublished reports from research underway.

1.3.1 Possible Biotic Causes

1.3.1.1 Early research

In 1992 soil samples from around the base of symptomatic trees were tested for the presence of known plant pathogens, such as *Phytophthora* species. *Phytophthora megasperma* was isolated from a few samples. *Eucalyptus camaldulensis* seedlings inoculated with fungal and bacterial treatments did not express the yellowing symptoms. However, the time and spatial scale of the study limited opportunities for findings that were statistically significant.

In 1993 soil samples at five diseased sites around Mundulla were tested, none of which yielded any species of *Phytophthora.* Importantly there was no evidence of root rot in even the smallest secondarily-thickened roots of five excavated saplings of diseased *E. camaldulensis*.

Between 1994 and 1999, field trials were conducted to determine whether or not yellowing in mature trees could be controlled with treatments of fungicide, antibiotics, insecticide and a nutrient solution. This study included a control treatment. The magnitude of yellowing and the location of the symptoms on each tree were measured and photographic records were taken. This revealed no obvious effects of treatment on symptom development. A gradual decline in health of affected individuals was recorded. However, the trials were not stratified sufficiently, or with requisite sample size, time and spatial scale, to be conclusive. Furthermore the ability to record responses was limited because some of the test trees were pruned inadvertently by a third party.

As the disease was different from previously described forms of eucalypt dieback, a description of the development of symptoms was published in a field guide in 1999. Early, medium and late stages of MY were described. Results of a preliminary, bark-patch grafting experiment appeared to show that symptoms could be transmitted to healthy recipient plants in the glasshouse. This suggested that an infectious, biotic agent may be involved. Investigations using molecular methods were then focussed on pathogenic micro-organisms as the possible cause. Actual contagion has still not been determined.

1.3.1.2 Phytoplasmas

In 2000 testing of the phytoplasma hypothesis was undertaken. Phytoplasmas were found in some eucalypts both with and without symptoms. This suggested that there was no evidence for a causal association with MY. However, minor cross contamination within the lab cannot be excluded as an explanation for detection of TBB phytoplasma in eucalypt species. More samples and improved laboratory procedures through isolation of operations amongst facilities were required for a greater level of confidence in these preliminary results. A new species of phytoplasma was identified in an *Allocasuarina muelleriana* displaying yellowing symptoms. This provided the first convincing evidence for the occurrence of natural infection of native species with a new phytoplasma.

In 2001 further work on phytoplasmas was undertaken and a greater number of samples were analysed. This supported the earlier conclusion that no strong association could be demonstrated

between the presence of phytoplasmas and trees with MY symptoms. This study concluded that phytoplasmas do not seem to be responsible.

Electron microscopy was then undertaken to look for morphological features typical of phytoplasmas or viruses but did not lead to the identification of a causal agent. There was no improvement in the condition of a small number of tissue cultures derived from trees with MY-like symptoms following tetracycline treatment which is known to inhibit phytoplasmas. This further supports the hypothesis that phytoplasmas are not a primary cause of MY. However an extended study was not done. A trial with tetracycline injection of established trees is needed to complete the tests to discount a role for phytoplasmas in MY.

1.3.1.2 Fungi and bacteria

In the initial studies undertaken, only saprophytes were found in cultures of bacteria and fungi from MY-affected field trees collected by researchers. This suggests that there is no evidence for known pathogenic bacteria or fungi as causal factors. However, as this work was not comprehensive, further research is required to discount bacteria and fungi as potential causal factors.

1.3.1.3 Virus-like organisms

In 2000 testing of the hypothesis that MY is caused by a virus-like organism commenced using molecular methods. Because no viruses or viroids of eucalypts have been characterised in Australia or elsewhere, it was first necessary to develop a molecular assay for virus-like agents in eucalypt tissue before trees with MY symptoms could be tested for the presence of virus-like genomic components. Researchers undertook nucleic acid analyses to compare extracts from healthy and MY-affected *E. camaldulensis.*

Unusual nucleic acids (MY-RNAs) were found to be associated with MY-affected *E. camaldulensis* in the South-East of South Australia. Their size and biochemical characteristics indicate that they may be associated with a virus or viroid.

Preliminary results from a southern Australia survey revealed that in South Australia, 100% of trees with typical MY symptoms had MY-RNAs. If this is a biotic disease, there is potential for non-symptomatic trees to be infected but not yet expressing the symptoms.

If symptom expression and development are influenced by environmental and host factors, the visible symptoms alone become an unreliable tool for disease diagnosis. In addition, other tree disorders can cause some symptoms similar to those of MY. The potential for MY-RNAs to be used as routine diagnostic markers of MY is therefore being investigated.

In 2002, the characterisation of MY-RNA commenced. A system was developed for finger-print analysis of small RNAs. The results so far show a number of single-stranded RNAs, of a size range between 300-600nt. Progress has been made with isolating, cloning and analysing (sequencing) some of the MY-RNAs.

1.3.2 Possible Abiotic Causes

Observations of yellowing symptoms on eucalypts in suburban South Australia were made between 1991 and 2000, and it has been suggested that observations of MY-like dieback are associated with herbicide use. However, analysis of foliage has not shown the presence of herbicide in MY affected

trees. MY must first be distinguished from other yellowing and dieback symptoms and trials with chemical treatments undertaken to test this hypothesis.

The current research program is investigating environmental correlates with symptom expression, including the distribution of herbicide use and data on chemical soil contaminants.

In 1999 analysis of nutrients in symptomatic leaves from MY trees and green leaves from normal trees was done. However, the results were difficult to interpret due to small sample sizes, and the large variation in absolute values of nutrient levels between sites. Moreover, changes in foliar nutrient levels may be an effect of physiological responses to tissue yellowing, rather than a cause.

One of the most striking features of the distribution and spread of MY is the apparent strong correlation with sealed or unsealed roadways, vehicular tracks, and sometimes with man-made and natural watercourses. Researchers and observers of the disease have reported this correlation throughout Australia. The correlation was investigated in 1994, and it was then noted that the disease was rarely found in trees or shrubs not adjacent to roadsides or vehicular tracks.

The April 2002 workshop stressed the possibility of environmental factors mediating the expression of the disease's symptoms. It was suggested that environmental factors (such as applied chemicals, soil type, moisture levels, physical disturbance, etc) may either be the cause, or predispose vegetation to infection by a biotic causal agent(s), or may interact with the causal agent(s) to exacerbate, or sometimes to suppress, disease symptoms and damage. When the causal factors are known, it will be necessary to systematically assess interactions and dependencies between biotic and abiotic factors (see Part 3 below).

1.4 Theories on how Mundulla Yellows Spreads

1.4.1 Biotic Causal Agents

In 2002 studies to assess whether or not Mundulla Yellows could be spread through revegetation activities commenced. MY-RNA was found in leaf tissue before yellowing symptoms were expressed. MY-RNA was also detected in revegetation sites and broad-acre plantations less than one year after planting in areas previously without MY, as well as in nursery-raised seedlings maintained in the glasshouse. While the time between infection and visible expression of symptoms of the disease has not yet been established, the results suggest there may be a risk of spreading MY through seedlings in revegetation programs. This would have serious implications for these programs in all states, and also for the plant nursery industry, and warrants further investigation.

Attention has also been given to possible insect vectors of MY. While there is no conclusive evidence yet to support this hypothesis, some interesting preliminary findings have been made suggesting possible transmission of a biotic agent by insects.

The roadside distribution of MY might suggest that it is less likely that a flying insect vector is involved in the transmission of MY. If a flying insect was involved, the disease would be more likely to spread outwards in all directions and not just along roads, unless there is some other environmental factor that exacerbates symptom expression in disturbed sites. However, the relatively dense "canopy corridor" that is an artefact of roadside vegetation may enhance opportunities for insect transport between plants.

The wide array of plant species apparently expressing symptoms of the disease suggests that it is unlikely that a single species of plant-feeding insect is responsible for transmission. Few phytophagous insects feed upon as many different plant families as have been identified with MY symptoms.

As a consequence of these difficulties with the single-insect-species vector hypothesis, soil-borne (eg. nematodes) and water-borne vectors will also need to be considered, and more complex interactions should be investigated.

1.4.2 Abiotic Causal Agents

Survey work in South Australia implicated soil and water as possible transmission media. In 2001-2002 preliminary tests of the soil-borne hypothesis were conducted. No clear relationship between MY symptoms and soil treatment was observed. However, soil transmission cannot be discounted as a factor because the experiment was unable to implement requisite hygiene controls and was not carried out long enough for symptoms to be expressed and interpreted.

1.5 Impacts (social, economic, environmental)

Concern has been expressed about the implications of MY for revegetation programs. With the steady increase in financial investment into revegetation for salinity and biodiversity rehabilitation programs (through natural resource management via the National Action Plan for Salinity and Water Quality, and the Natural Heritage Trust), there are growing concerns that outcomes may be compromised by the disease. The possibility that MY can spread to new planting areas via nursery seedlings adds a further dimension to the concern.

Local government areas in several states are already encountering significant management and economic problems due to MY. For example, the City of Geelong estimates that it is currently removing more that 10,000 m^3 of plant material each year related to Mundulla Yellows dieback. The financial and public safety risks associated with the spread of Mundulla Yellows in a municipality such as Geelong have yet to be documented.

Reduced levels of flowering and seed set in living trees expressing MY symptoms have also been documented. This presents broader implications for the regenerative capacity of affected plants and for fauna species requiring nectar, pollen and seed resources.

Although MY appears to be restricted mostly to roadside vegetation, the range of overstorey and understorey species apparently affected may have major environmental implications in fragmented landscapes, such as South Australia's South East, where much of the remnant native vegetation exists along roadsides and supports significant populations of threatened plant species. While the death of a single tree in highly fragmented landscapes results in environmental losses, the importance of scattered trees has yet to be documented precisely. The destruction of scarce remnant vegetation in areas where broad-scale land clearing has been carried out is seen as a significant environmental loss. Biodiversity conservation will be affected where rare and threatened species are involved.

The death of ancient eucalypts can also impact on both European and Aboriginal cultural heritage. However, to date, no deaths of specific trees of cultural significance have been documented.

2. RISK MANAGEMENT

2.1 Risk Management Workshop

Broadleaf Capital International Pty Ltd were commissioned by the Mundulla Yellows Task Group in May 2003 to conduct a Risk Management Workshop. The purpose of this meeting was to review and discuss technical information on the causation and epidemiology of Mundulla Yellows, to review research directions, to carry out a risk analysis exercise on MY, and to consider possible interim and subsequent management measures.

The workshop was attended by invited participants from Government, industry and research institutions in all states. Participants in the workshop were chosen because of their affiliation with particular stakeholder groups or organisations, or because of their expertise in the field of one or more of the identified potential causal mechanisms.

The workshop was divided into five components:

1. A review of the Australian and New Zealand Standard for Risk Management, AS/NZS 4360;
2. A review and discussion of present knowledge of MY causation and epidemiology;
3. A review and discussion of research opportunities;
4. A review and discussion of interim management measures, and measures that might follow from improved diagnostics or improved understanding of the epidemiology of MY; and
5. Discussion of the policy framework for ongoing monitoring and review of research and management options.

Within these discussions, the current distribution of MY in Australia was examined and four risk scenarios for further spread were identified, analysed and evaluated by the consultant. It should be noted that these scenarios were not worked through at the forum, but rather developed by the consultant as a response to information gleaned from the workshop. This exercise is based on the framework and guidelines for risk management described in the Australian and New Zealand Standard AS/NZS 4360.

The consultants chose the following four risk scenarios:

- Scenario 1: Spread of MY to and within Local Government Areas throughout Australia;
- Scenario 2: Spread of MY to and within Australia's plant nursery industry;
- Scenario 3: Spread of MY to and within Australia's World Heritage Areas, national parks and state forests, and
- Scenario 4: Spread of MY to and within Australia's forestry industry.

The workshop participants generally agreed that in the absence of conclusive data on the cause(s) of MY, the mode(s) of transmission and possible contributing factors, a detailed risk analysis is premature. However, while it must be stressed that the likelihood of the disease spreading to and within any of the areas listed in the scenarios is purely speculative, the consultant believed that the impact, if this happened, can be evaluated.

The risk assessment included analysis of the **likelihood** and **consequences** of MY spread in each of the four identified scenarios, and the subsequent evaluation of **risk** as a "worst case" outcome.

The **likelihood** of MY spread in each of the four risk scenarios was evaluated qualitatively by the consultant after the workshop. The qualitative descriptors adopted for this purpose are those commonly used for many risk assessment exercises (Table 1).

Table 1. The qualitative descriptors used to estimate the likelihood of MY spread in each risk scenario.

Likelihood rating	*Likelihood description*
1	*Rare*: the scenario would only occur in exceptional circumstances;
2	*Unlikely*: the scenario could occur at some time;
3	*Possible*: the scenario might occur at some time;
4	*Likely*: the scenario would probably occur; and
5	*Almost certain*: the scenario is expected to occur.

The **consequences** of MY spread in each of the four risk scenarios were analysed qualitatively by the consultant after the workshop. Each qualitative analysis was based on the systematic examination of the direct or indirect impact of MY on the environment (e.g. ecosystems, biodiversity and endangered species), relevant industries (e.g. forestry, nurseries, agriculture, apiculture), Government (Local, State and Australian Government) and the amenity values of affected communities.

The qualitative descriptors used to describe the impact of MY on the environment, industry, Government or the community are listed in Table 2.

Table 2. The qualitative descriptors used to estimate the consequential impact of MY spread in each risk scenario.

Consequence rating	*Impact description*
1 Insignificant	Consequences associated with the risk scenario are unlikely to be recognised;
2 Minor	Consequences associated with the risk scenario are likely to be recognised; although unlikely to exceed the impact of most existing plant disease problems;
3 Moderate	Consequences associated with the risk scenario are likely to exceed the impact of most existing plant disease problems;
4 Major	Consequences associated with the risk scenario are likely to exceed the impact of most existing plant disease problems, and include a significant but reversible impact; and
5 Catastrophic	Consequences associated with the risk scenario are likely to exceed the impact of most existing plant disease problems, and include a significant and irreversible impact.

The **likelihood** and **consequence** estimates for each scenario were combined for evaluation using the **qualitative risk** curve in Figure 1. The terms 'tolerability' and 'acceptability' have been defined as follows:[2]

1. '**Tolerability**' refers to the willingness to live with a risk to secure benefits, on the understanding that it is being properly controlled. Tolerability does not mean 'acceptability'. Tolerating a risk does not mean that it is regarded as negligible, or something that can be ignored, but rather as something that should be reviewed and reduced still further, if and when this is possible.
2. '**Acceptability**' relates to risks that are at an acceptable level and do not need further consideration. The expression 'acceptable level of risk' refers to the level at which it is decided that further restricting or otherwise altering the activity is not worthwhile; e.g. it will not result in significant reduction in risk, or additional expenditure will not result in significant advantages of increased safety.

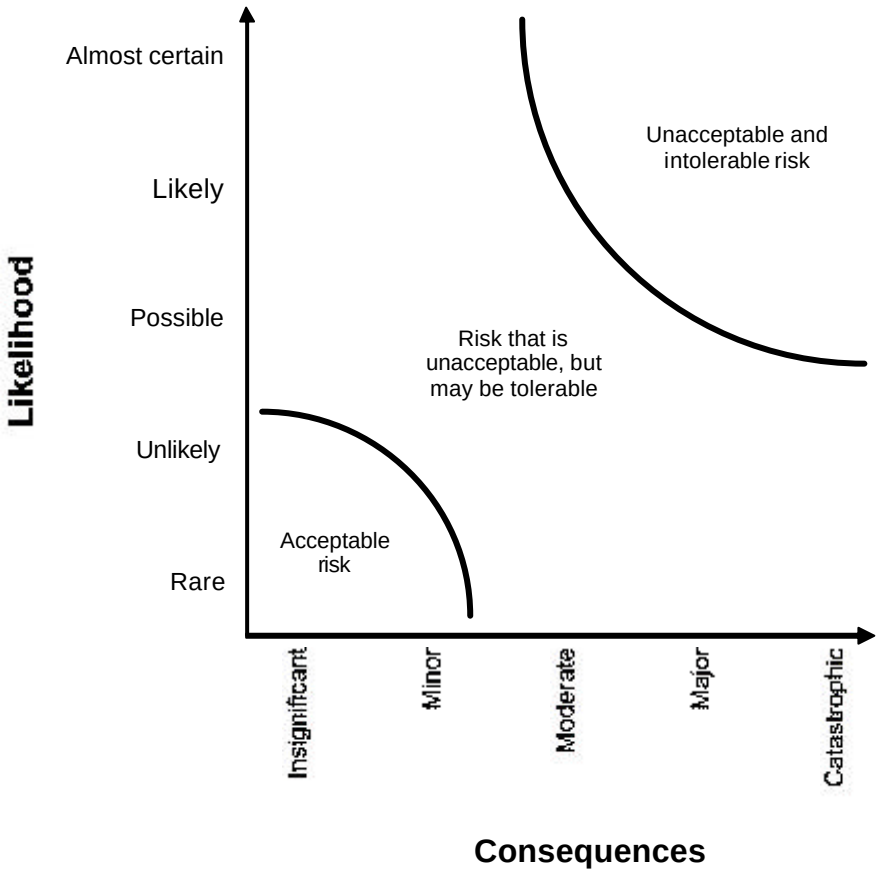

Figure 1: Generic Qualitative Risk Curve as presented by Broadleaf Capital International Pty Ltd at the Mundulla Yellows Risk Management Workshop.

[2] Adapted from HB 203:2000, Environmental Risk Management: Principles and Processes, Standards Australia / Standards New Zealand.

2.2 Risk Management Scenarios

Analysis by Broadleaf Capital International Pty Ltd suggested that the "worst case" risk associated with Scenarios 1, 2 and 3 was considered unacceptable and intolerable. This reflects the seriousness of the threat associated with a fatal and potentially transmissible disease affecting many genera of Australian native flora. The result also reflects the current uncertainty associated with the causation and epidemiology of the disease, and the current inability to identify infected plants reliably. The reasoning behind Broadleaf Capital International Pty Ltd's assessment is as follows.

2.2.1 Scenario 1: MY Spread and Local Government Areas

Likelihood: Evidence provided at the workshop demonstrated that vegetation in some local government areas (LGAs) in Australia is seriously affected by MY. Affected plants may be located either in rural roadside or riparian remnant vegetation or plantings (and in adjacent privately owned farmland), or in urban parklands or amenity plantings (including private gardens). The plants may be either remnants of natural vegetation or cultivated native or exotic species. Some LGAs in Australia are more seriously affected by MY than others.

Risk factors for the severity of disease at the council level are not known, and it might appear that the difference between areas with a low prevalence of affected trees, and those with a serious amenity and environmental problem, may simply be the time that has elapsed since the first incursion of the disease. However, some site types may be more conducive to the development of the disease than others.

It was believed that the present occurrence of disease was strongly correlated with the distribution of sealed or unsealed roadways, vehicular tracks, man-made and natural watercourses, and with areas where people congregate. There were reports that seedlings of many genera and species planted into affected areas developed the disease, and that the disease was invariably fatal. These observations were coupled with a suspicion that disease may be transmitted to new areas with contaminated wood chips produced from prunings of diseased limbs or from felled trees, or with contaminated pruning or felling equipment. It was also feared that the planting of infected nursery stock within LGAs may lead to the distribution of contagion to previously unaffected areas.

Based on reports of the apparent spread of MY within particular LGAs, it was concluded that without mitigation, spread of MY to and within other LGAs was likely (Table 1) to occur. It was recognised that clarification of the cause and means of transmission of disease would lead to refinement of the estimate as well as to the development of effective containment or eradication protocols.

Consequences: The impact of MY on LGAs throughout Australia would depend very much on its rate and extent of spread and whether some site types are more conducive to MY development than others. Potentially, if an infectious agent(s) is involved, the impact would be major (Table 2). It was noted that until a cause is identified, it is not possible to legally restrict the movement of possibly infectious material.

Risk assessment: Spread of Mundulla Yellows was considered likely (Table 1) to have a major impact (Table 2) if widespread amongst and within LGAs through Australia. The worst-case risk level for this scenario was therefore assessed as unacceptable and intolerable (Fig. 1). In making this assessment it was again recognised that clarification of the causation and epidemiology of MY, and

the provision of reliable diagnostic indicators for infection, would remove many uncertainties and enable a more precise estimate of risk.

2.2.2 Scenario 2: MY Spread and the Plant Nursery Industry

Likelihood: Evidence provided at the workshop indicated that there is a possibility that spread of MY to parts of Australia's nursery industry may already have occurred. As the causative agent(s) and the method(s) of transmission remain unknown, direct comparisons with other more closely studied diseases is difficult. A concern raised by workshop participants and by the MY Task Group, based on other disease scenarios, was the role the nursery industry might unknowingly play in spreading the disease through in-house contamination, and through the subsequent distribution of infected plants or propagation materials by national park authorities, forestry, local councils or individual consumers for use in revegetation programs. For example, it was noted that many tens of millions of seedlings are supplied annually from nurseries for planting under salinity action plans and other land-care programs. The workshop and the MY Task Group recognised the importance of the nursery industry adopting best practice nursery hygiene standards.

Thus spread to and within the nursery industry was considered likely (Table 1), if an infectious biotic agent(s) is involved. Clarification of the uncertainties associated with causation and epidemiology would lead to a clarification of the likely pathways for exposure, and a refinement of the likelihood estimate.

Consequences: Without an approximation for the rate of spread of MY within the nursery industry, or the ramifications of containment or eradication policies, it is difficult to estimate the impact of this risk scenario with any precision. However, it is apparent that the range of susceptible hosts is very large and may incorporate many Australian native plant genera. A disease of this breadth would be likely to have at least a major impact (Table 2) on affected parts of the nursery industry. It is also likely that spread to the nursery industry would have an indirect impact on reforestation of farmed, harvested or otherwise damaged ecosystems, or on the provision of amenity plants to local councils or individual consumers.

Risk assessment: Available evidence suggests that spread of MY to and within Australia's nursery industry is likely (Table 1) if an infectious biotic agent is involved, and would result in a major level of impact (Table 2). The worst-case risk level for this scenario was therefore assessed as unacceptable and intolerable (Fig. 1). Although further spread from the nursery industry by national park authorities, forestry, local councils or individual consumers would inflate this substantially, these impacts are considered within separate risk scenarios.

2.2.3 Scenario 3: MY spread and World Heritage Areas, National Parks and State Forests

Likelihood: Although MY has not yet been identified in Australia's World Heritage Areas, national parks and state forests, or in any undisturbed stands of native vegetation, the widespread distribution of susceptible native species, and the construction, maintenance and relatively high use of vehicular and pedestrian access routes in these areas, could favour its entry and establishment. In the worst-case outcome, if an infectious agent(s) is involved, its spread and expression within forest and heathland could then later be favoured by conditions becoming more conducive to the disease. Also relevant is the movement into parks of timber and other tree-based materials for construction, as well as road-building and other materials for the preparation and upkeep of amenity areas. These activities would provide localised conditions in many ways analogous to parks and gardens in many urban and

suburban areas where the disease has established and appears to be spreading. Although less well understood, watercourses, and in particular man-made watercourses (including roadside drains), appear to be associated with the spread of the disease, and therefore could be implicated in the realisation of this scenario.

The Task Group concluded that any rating of likelihood (Table 1) for spread of MY in this scenario, based on current knowledge, will clearly be highly speculative. If an infectious agent(s) is involved, its arrival and spread is at least possible.

Consequences: If it were to spread to Australia's World Heritage Areas, national parks or state forests, the impact of MY on the natural environment may be catastrophic (Table 2), if an infectious agent(s) is involved. It is important to differentiate clearly here between spread to the areas, (ie arrival, which on present indications will not lead to much off-road damage); and spread within the areas away from roads, which would be catastrophic (Table 2), if it were to happen.

Broadleaf Capital International Pty Ltd assigned the catastrophic rating (Table 2), on the basis of a reduction in seed set and flowering, and the subsequent death of native trees and shrubs of many genera, which could endanger habitats, ecological communities and ecosystems, and lead to a significant reduction in biodiversity. Moreover, the inability to reconstruct ecological communities or even to replant into affected areas would mean that the effect of the disease might, in some areas, be irreversible, and that the survival of endangered species or ecological communities could be threatened. Under this scenario, the economic impact of MY is less easily predicted, although tourism and ecotourism are likely to be at least moderately affected (Table 2). The impact on Government is also likely to be moderate to high, although it is recognised that control and eradication costs (including intensive research into the causation and epidemiology of the disease) could be substantial if MY established in larger national parks or World Heritage Areas. The impact of MY on the amenity value that the Australian community attributes to state forests, national parks and World Heritage Areas is the most difficult to estimate. However, intense public interest in and commitment to mitigating natural disasters or industry activities affecting these areas, would suggest that the incursion of an apparently fatal disease of many native species is likely to be considered at least of major importance.

Evidence provided during the workshop indicated that with spread to and within Australia's World Heritage Areas, national parks or state forests, the impact of MY would be considered catastrophic (Table 2). In making this assessment it was again recognised that clarification of the causation and epidemiology of MY, and the provision of a reliable diagnostic indicator, would remove many uncertainties and enable a more precise estimate of likely impact.

Risk Assessment: Available evidence suggests that spread of MY to and within Australia's World Heritage Areas, National Parks and State Forests is at least possible (Table 1) if an infectious biotic agent is involved, and could result in a catastrophic level of impact (Table 2). The worst-case risk level for this scenario was therefore assessed as unacceptable and intolerable (Fig. 1). However, it is important to note that this is probably the most speculative of the four scenario risk assessments, as MY has not been reported in undisturbed stands of vegetation anywhere. While Broadleaf Capital International Pty Ltd consider the risk scenario as being unacceptable and intolerable (Table 3), the MYTG judged that the possible spread of MY to roadsides in a significant stand of native vegetation could be tolerable and potentially manageable.

2.2.4 Scenario 4: MY Spread and the Forestry Industry

Likelihood: If its cause is biotic, MY spread to areas within the forestry industry would readily be facilitated by the abundance of susceptible tree species, current forest management practices and as for Scenario 3, by the construction, maintenance and relatively high use of vehicular and pedestrian access routes. Although considered separately (see Scenario 2 above), it is relevant that spread of MY to nurseries and to propagated seedlings would enhance its rate of spread through the forestry industry. If introduced by this means, widespread dissemination of the disease would be likely. Moreover, because trees replanted into MY-affected sites are likely to develop the disease, forest plantations could be adversely affected and the sustainability of forestry sites threatened. While not all replanted trees are likely to develop MY, those planted in MY sites, or wherever infected planting stock is used, could be susceptible.

Broadleaf Capital International Pty Ltd considered it likely (Table 1) that, if unmitigated, MY would spread to Australia's forestry industry. However, because the causation and epidemiology of MY are largely unknown, it was difficult to estimate the likely rate and extent of spread through Australia's forestry industries. Clarification of these uncertainties through research efforts directed at the causation and epidemiology of MY is necessary.

Consequences: If MY was to spread to Australia's forestry industries, its economic impact is likely to be at least moderate (Table 2). Difficulties associated with the replanting of trees, the sustainability of forestry in affected areas, and the effect on industry confidence may further inflate this estimate.

Different sectors of "forestry industry" (eg native forest vs plantation, undisturbed vs highly disturbed) are all managed very differently, and hence would have different risks. Each component of the Australian forestry industry would need to be assessed as a separate risk scenario for this to be accurate.

Risk Assessment: Available evidence suggests that spread of MY to and within Australia's forestry industry is at least possible (Table 1) if an infectious biotic agent is involved, and could result in a moderate level of impact (Table 2). The risk for this scenario was therefore assessed as unacceptable but may be tolerable (Fig. 1).

The results of Broadleaf Capital International Pty Ltd's evaluation of the four risk scenarios are summarised in Table 3.

Table 3. Evaluation of the risk of spread of Mundulla Yellows in four Risk Scenarios as determined by Broadleaf Capital International Pty Ltd following the Mundulla Yellows Risk Management Workshop in May 2003

Scenario	Likelihood	Consequences	Risk
1. Spread of MY to and within LGAs throughout Australia	Spread to these 4 areas is possible, if not likely. This will be better able to be determined following results of research into the cause and a diagnostic indicator.[1]	Major	Unacceptable and intolerable
2. Spread of MY to and within Australia's plant nursery industry		Major	Unacceptable and intolerable
3. Spread of MY to and within Australia's World Heritage Areas, national parks or state forests		Catastrophic	Unacceptable and intolerable
4. Spread of MY to and within Australia's forestry industry		Moderate	Unacceptable but may be tolerable

[1] The likelihood ratings by the consultant have been omitted as they were thought to be unnecessarily alarmist, considering the lack of information available.

It can be seen from Table 3 that under the general qualitative criteria adopted for this analysis, the risk associated by Broadleaf Capital International Pty Ltd with three of the four scenarios for the further spread of MY in Australia were considered both unacceptable and intolerable. The MYTG considered only two of these four scenarios (Scenarios 1 and 2) to have unacceptable levels of risk based on current information.

These results reflect the seriousness of the threat associated with a fatal and possibly transmissible disease that has the potential to affect many genera of Australian native flora. However, these results also reflect the extent of uncertainty associated with the causation and epidemiology of the disease, and the current lack of a sensitive diagnostic tool to identify reliably both symptomatic and pre-symptomatic infected plants.

2.3 Recommendations: Risk Assessment

The MYTG found the analysis of the four scenarios for further spread of MY using the Broadleaf Capital International Pty Ltd assessment based on the Standards Australia/Standards New Zealand *Environmental Risk Management: Principles and Processes* to be valuable. However, it was of the view that without an understanding of disease causation and epidemiology, and a reliable diagnostic indicator for MY, the risk associated with this disease is unable to be realistically assessed.

2.4 Recommendations: Risk Treatment

2.4.1 Recommendations on Disease Management

The workshop participants made the following recommendations regarding the implementation of interim management measures for the containment of MY:

1. Endorse best practice guidelines for nursery hygiene, replanting or reforestation, and for hygiene when handling known MY-infected material (it was noted that a set of guidelines has already been developed by Dr's Randles and Hanold but that these need to incorporate soil transmission protocols similar to those adopted for *Phytophthora*);
2. Develop/endorse existing best practice guidelines for handling and disposing of limbs or felled trees suspected to be infected with MY;
3. Develop a fact sheet for councils and other government bodies responding to frequently asked questions on MY. This should include:
   - 3.1 Symptoms and things to look out for,
   - 3.2 Who to contact,
   - 3.3 Reinforce existing best practice guidelines for hygiene and plant management.

However, the MYTG is of the view that no definitive recommendations concerning disease management can be made until such a time that the causative agent(s) has been determined. The MYTG noted that a definitive description of MY is available at:
http://www.agwine.adelaide.edu.au/research/plant/path/pv/MundYellow.pdf

3. RESEARCH DIRECTIONS

In order to manage the disease, a minimum of four necessary tools have been identified. These management tools are as follows:

1. A reliable routine diagnostic test for identifying MY-infected trees with or without typical disease symptoms;
2. Identified host and environmental risk factors that could be mitigated where possible to minimise the spread of MY and the infection of individual trees;
3. Identified key transmission pathways that could be mitigated to minimise the spread of MY and infection of individual trees, and the introduction of MY to new areas; and
4. Identified disease-resistant or non-susceptible species, strains or cultivars that could be used in the rehabilitation of MY-affected areas.

In order to develop these tools the research priorities listed in 3.1 have been agreed.

3.1 Agreed Research Priorities

3.1.1 Actions contributing to identification of the cause

1. Identification and characterisation of causal agent(s)
2. Development of a routine diagnostic test for the causal agent(s)
3. Determination of transmission/spread mechanisms, and possible vectors
4. Investigation of the role of environmental factors, and predisposition (if the cause is biotic)

3.1.2. Actions which will flow from understanding the cause

1. Systematic characterisation of the symptoms on a variety of hosts and different site types
2. Studies on the effects of the disease on the host: biochemical, physiological and anatomical
3. Systematic determination of local and national distributions and host range
4. Genetic resistance/susceptibility studies
5. Systematic assessment of environmental, economic and social costs associated with the current distribution of MY in Australia
6. A comprehensive risk analysis and development of management/intervention strategies and procedures

3.2 Discussion of Priority Research Areas

3.2.1 Identification of Causal Agent(s) and Development of a Diagnostic Test

All possible pathogen groups, and other potential causes, must be investigated and excluded systematically. Some, such as xylem and phloem bacteria, have yet to be considered. Manion (1991) provides a comprehensive overview of approaches to biotic, abiotic and tree decline diseases and an entry to the literature.

A consistent and inexpensive routine diagnostic test for the disease is required to enable the reliable identification of MY-affected plants, so that they can readily be distinguished from those affected by other disorders that may cause some similar symptoms to MY. Only then can appropriate management of diseased vegetation be carried out. This routine test is also needed before the breadth of species impacted, and sites affected, can be systematically and accurately documented.

Given the attention it has received, it is necessary to establish whether or not MY is caused by an organism associated with MY-RNA or whether or not the RNA is a metabolic response by the tree to the disease. The putative pathogenic organism(s) (virus or viroid) must be characterised. Trials could then be conducted to determine, according to Koch's postulates, whether it is the cause of the disease. The possible existence of different strains of such an organism will then need to be investigated, their distribution documented, and their pathogenicity levels and host-specificities tested. If research proves that MY-RNA can be used as a diagnostic marker for MY, a rapid method of screening for presence of the RNA, such as a molecular probe, will be a priority.

The possible role of bacterial or fungal pathogens as causal agents of the disease could be further explored by repeating some of David Paton's experiments (Appendix 4), and applying treatments of fungicides and antibiotics on a more intensive and extensive scale in an experimental framework both under laboratory and field conditions. A trial with tetracycline injection of established trees is needed to complete the tests to discount a role for phytoplasmas in MY.

The effects of soil chemistry and nutrition on eucalypts and other affected plants should be studied as well as the role of herbicides and other environmental chemicals.

3.2.2 Modes of Transmission and Spread

The modes of transmission of the disease must be investigated. This research will include the necessary verification of preliminary evidence regarding the transmissibility of the causal agent(s), including further graft trials, and of the involvement of flying insects as vectors. The roles of other potential vectors such as soil arthropods, fungi, and nematodes should be examined. Typically, these types of trial also require a means of assaying the vector itself for the agent transmitted. This work will require a sensitive and specific molecular test for the putative causal agent.

A further physiological consideration is whether or not putative causal agents can be detected in plant organs other than leaves. Such information may suggest transmission routes within a plant.

Soil and water movement, as well as human involvement, should be investigated as potentially important vectors of MY. Saprophytic, parasitic or mycorrhizal fungi were also seen to be potential vectors, although none have been reported in the literature. Field studies of the pattern and rate of spread of MY under different environmental conditions will also be required.

3.2.3 Environmental Factors and Predisposition

Environmental factors can predispose plants to be more easily infected by a pathogen, or to show symptoms more rapidly or more severely after infection by a pathogen. Individual elements of the "Disease Triangle" (host, pathogen, and environment) should not be studied in isolation. Understanding their interaction and its balance may provide the key to effective management, based on the disruption of the disease cycle and the exploitation of weaknesses.

Environmental (abiotic) factors such as nutrient imbalances or herbicide applications have been suggested as possible causes of MY. Trials should be carried out to investigate these possibilities further.

The presence of environmental stressors, such as nutrient imbalances, deficiencies or toxicities and herbicide inputs that could predispose trees to MY should be documented. Surveys should capture data to determine whether or not such stressors vary between roadsides, scattered trees in paddocks or parklands, and intact native vegetation.

3.2.4 Systematic Characterisation of Symptoms

Symptom expression in differing site types and host species will need to be systematically documented once a routine diagnostic test for MY has been developed.

3.2.5 Effect of Disease on Host – Biochemical, Physiological and Anatomical

Detailed knowledge of the effects of MY on host plants at the macroscopic, microscopic and sub-cellular level, the mechanisms of infection, invasion and symptom expression, and of how these processes are mediated, should give some useful indications of possible approaches for devising appropriate control strategies.

3.2.6 Systematic Determination of Local and National Distributions and Host Range

Further research will first be required to determine whether or not symptom expression varies amongst different host species, and between site types (see 3.2.4). A systematic and stratified regional survey of MY across Australia would incorporate estimates of the prevalence of symptomatic trees and would capture sufficiently accurate geospatial data to provide for estimates of the change in prevalence over time. This survey should also incorporate measures of potential environmental correlates with the expression of MY-like symptoms across a range of ecosystem types and land uses. A systematic survey encompassing a broader diversity of vegetation communities and soil types is required in a region exhibiting the full spectrum of MY expression before correlations with land-use, disturbance factors and soils can be made.

The reported link between roadsides and the occurrence of MY needs to be better established. The May 2003 Risk Management Workshop participants recommended a fine scale survey focused in a landscape where full expression of MY occurs, such as the South East of South Australia, to document environmental correlates of the disease. It is critical to document how much of the dieback in rural trees throughout Australia can be attributed to MY. This would also provide important data on the scale of general dieback diseases in Australia and could suggest the breadth of potential causes.

3.2.7 Genetic Resistance or Tolerance

The possibility of locating genetically resistant or tolerant trees amongst natural populations to provide source material for management and rehabilitation options in affected sites should be explored.

3.2.8 Risk Assessment

A comprehensive risk analysis for MY will need to be carried out once the cause has been elucidated. This analysis/assessment will need to include estimates of the environmental, social and economic costs, incurred by Government (local, State or Territory and Australian), industry and the Australian community.

Management strategies and procedures specific to MY will then be able to be developed. The introduction of such procedures may involve significant cost to land managers and industry, and therefore it will be essential that adequate and appropriate validation trials of the proposed measures are first carried out.

3.3 Current Research Underway

The University of Adelaide (Waite Campus) which had previously carried out molecular investigations of MY, is continuing this work under a 3 year linkage grant by the Australian Research Council (ARC) commencing in early 2004. Molecular research will be carried out to develop a routine diagnostic test for MY, identify causal organism(s), and investigate modes of spread and the disease cycle. The project is based on biotechnology developed at the Waite Campus and extensive previous experience of the team in determining the causes of new plant diseases. The University of Adelaide (Waite Campus) research team comprises:

Project Team: Prof John W Randles (Plant Virologist) and Dr Dagmar Hanold (Molecular Biologist); Industry Partners: Department of Conservation and Land Management WA, State Forests NSW, Transport SA, District Council of Tatiara, Barossa Council, and Coorong Council.

Research Scientists from the Victorian Department of Primary Industries (DPI, Knoxfield) and Department of Sustainability and Environment (DSE, Heidelberg), have been undertaking research funded by the Australian and South Australian Departments of Environment and Heritage since March 2003. They are systematically examining both biotic and abiotic factors and their interactions. The project provides a multidisciplinary approach with strong capabilities in plant disease diagnostics, plant disease epidemiology, molecular biology and survey design. The Victorian research team consists of:
Project team: Dr. Rosa Crnov (Molecular Virologist), Barbara Czerniakowski (Agronomist/Soil Pathologist), Ian Smith (Forest Pathologist), Dr. David Cheal (Flora Ecologist), Dr. Steve Sinclair (Flora Ecologist/Molecular Biologist), Fiona Thomson (Biometrician), Geoff Sutter (Flora Ecologist), Peter Franz (Biometrician) and Dr.Jo Luck (Molecular Plant Pathologist).
Collaborators: Dr. Karen Robb, (Entomologist, UC Riverside); Linda Semeraro, (Entomologist, DPI); Ian Pascoe (Mycologist), Lila Nambiar, (Nematologist, DPI); Mark Imhof (Soil Chemist, State Chemistry Laboratories).

Research on local distribution and spread of MY has been undertaken by Dr. David Paton (University of Adelaide, School of Earth and Environmental Sciences). He is continuing investigations into possible associations between changes in spread, land-use, disturbance and the presence of MY.

4. RECOMMENDATIONS

4.1 Research

The Task Group is unanimous in advising that, based on the available information, MY is a serious new plant disease threat that must be addressed. This is consistent with a prudent approach to a widespread new disease.

Basic work is urgently required to define the problem and identify its cause(s) and contributing factors, investigate its mode(s) of spread, and develop a rapid, routine diagnostic test. This work is a pre-requisite to:

1. carrying out a comprehensive risk assessment, which will enable individual jurisdictions to estimate the level of threat posed to different categories of vegetation in their regions;
2. developing cost-effective management strategies to deal with the disease where it is already established, to minimise its impact, and to prevent it from spreading to new areas.

There are two primary areas of research underway by The University of Adelaide (Waite Campus) (SA) and the Department of Primary Industries, Knoxfield (Vic) that are yielding promising results and which the MYTG believes should continue. The MYTG recommends every effort be made to promote synergies between the two research programs and minimise unnecessary duplication to maximise the utility of the research.

This will be best facilitated through the continuation of the MYTG with revised terms of reference and possibly expanded membership. This facilitation should be supported by the establishment of a dedicated website which promotes links between MY stakeholders.

Once more is known about the disease, it will be appropriate for regions where Mundulla Yellows is identified as a problem to seek funding for further research, and for their management of the problem, through existing channels. At that time it would be useful to revisit the composition of a multi-jurisdictional review group to coordinate research and facilitate communication and the development of management strategies.

Realistic estimates of the threat posed to different stakeholders, which will directly determine their willingness to fund research, are not possible until a causal agent(s) is identified. The MYTG believes under these circumstances it is difficult to identify further non-government funding sources.

4.2 Management

The MYTG is of the view that no definitive recommendations concerning disease management can be made until the causative agent(s) has been determined.

Acknowledgements

The MYTG acknowledges the work of the four peer reviewers Drs Adrian J Gibbs (Primatrix), Ross E Beever (Landcare Research, NZ), Caroline Mohammed (CSIRO) and Charlma Phillips (Forestry SA) for their critical analysis of the Broadleaf Capital International Pty Ltd Risk Management Report. The contributions of the workshop participants are also gratefully acknowledged.

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Appendix 2: The Mundulla Yellows Task Group – Terms of Reference and Membership

Terms of Reference of the Mundulla Yellows Task Group

(a) Provide a status report on current research into the cause and spread of Mundulla Yellows and current risk management activities;

(b) Devise a broad and flexible short and medium term research strategy that identifies multiple pathways to improve knowledge of the cause and control of Mundulla Yellows;

(c) Establish a risk management framework that takes into account the current incomplete state of knowledge of the epidemiology of Mundulla Yellows, and identifies roles for specific groups in establishing appropriate hygiene and quarantine procedures for containing Mundulla Yellows.

(d) Recommend a funding strategy for the research and management activities identified in (b) and (c).

Members of the Mundulla Yellows Task Group

Ms Christine Schweizer (**Chair**) Australian Government Department of the Environment and Heritage, Canberra	Mr Mike Stukely Department of Conservation and Land Management, Western Australia
Dr Michael Cole Australian Government Department of Agriculture, Fisheries and Forestry, Canberra	Dr Stephanie Williams Department of Environment and Heritage South Australia
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Mr Jack Simpson State Forests New South Wales	Corresponding member: Mr Tim Rudman Department of Primary Industry, Water and Environment, Tasmania
Mr Ian Smith Dept of Sustainability and Environment Victoria	

Appendix 3: Species Affected by Mundulla Yellows

Data below on plants known to be affected is derived from Paton (2000) and Sarneckis (2000). The data from Sarneckis (2000) was sourced opportunistically, not systematically. Species native to South Australia are in bold and species introduced from other countries or from other Australian states are in italics.

Acacia sp.
Acacia *cyclops*
Acacia farinosa
Acacia longifolia* subsp *sophorae
Acacia pycnantha
Acacia rivalis
Allocasuarina littoralis
Allocasuarina verticillata
Angophora costata
Bursaria sp.
Callistemon harkness
Callistemon sp.
Corymbia ficifolia
Dianella sp.
Eucalyptus sp.
Eucalytpus arenacea
Eucalyptus baxteri
Eucalyptus camaldulensis
Eucalyptus camaldulensis* var. *camaldulensis
Eucalyptus cladocalyx
Eucalyptus cneorifolia
Eucalyptus diversifolia
Eucalyptus fasciculosa
Eucalyptus gomphocephala
Eucalyptus incrassata
Eucalyptus intertexta
Eucalyptus leptophylla
Eucalyptus leucoxylon
Eucalyptus leucoxylon* subsp. *Stephaniae
Eucalyptus obliqua
Eucalyptus occidentalis
Eucalyptus platypus
Eucalyptus sargentii
Eucalyptus sideroxylon
Eucalyptus stoatei
Hakea sericea
Kunzea pomifera
Lecopogon parviflorus
Lophostemon confertus
Melaleuca sp.

Melaleuca hypericifolia
Melaleuca brevifolia
Melaleuca nesophylla
Xanthorrhoea sp.

Amenity Species Affected in Unley Council Area

Acacia baileyana
Acacia decurrens
Acacia floribunda
Acacia iteaphylla
Acacia longifolia
Acacia pendula
Acacia podalyraeifolia
Acacia sp.
Acacia vestita
Acmena smithii
Agonis flexuosa
Albizzia sp.
Angophora costata
Angophora sp.
Banksia sp.
Callistemon Captain Cook
Callistemon citrinus
Callistemon Kings Park
Callistemon pinifolius
Callistemon sp.
Callistemon viminalis
Casuarina cunninghamiana
Chamelaucium uncinatum
Conifer
Cordyline australis
Corymbia citriodora
Corymbia sp.
Eucalyptus botryoides
Eucalyptus caesia
Eucalyptus camaldulensis
Corymbia citriodora
Eucalyptus cladocalyx
Corymbia eximea
Corymbia ficifolia
Eucalyptus leucoxylon
Eucalyptus luehmanniana
Corymbia maculata
Eucalyptus nicholii
Eucalyptus platypus
Eucalyptus sideroxylon
Eucalyptus spathulata
Eucalyptus torquata
Eucalyptus sp.
Grevillea robusta
Grevillea rosmarinifolia
Grevillea sp.
Hakea laurina
Hibiscus sinensis
Hymenosporum flavum
Jacaranda mimosifolia
Lophostemon confertus
Melaleuca armillaris
Melaleuca decussata
Melaleuca hypericifolia
Melaleuca incana
Melaleuca nesophylla
Melaleuca styphilioides
Melaleuca sp.
Protea neriifolia
Thryptomene paynii

Appendix 4: Research Undertaken

1975	First documentation of MY symptoms	Geoff Cotton, an apiarist based in Keith, South Australia
Early 1990's.	Short-term distributional study in South Australia's Upper South East.	Dr's David Paton and Steven Eldridge
1992	Investigation for the presence of known fungal plant pathogens	David Paton and Steven Eldridge
1992 and 1993	Mapping of the distribution of MY in South Australia's Upper South East	David Paton and Steven Eldridge
	Soil samples at five diseased sites around Mundulla taken	Dr. Frank Podger and Geoff Cotton
1994 - 1999	Field trials using treatments of fungicide, antibiotics, insecticide and a nutrient solution.	David Paton and Jo Cutten
1997 – present 1999	Investigations into MY Disease description (early, medium and late stages)	Drs John Randles, and Dagmar Hanold D. Hanold & JW Randles
1995 and 1998	Recording of MY-affected sites over time in the Adelaide metropolitan area	John Choate of the South Australian Department for Environment and Heritage (DEH).
1999	Analysis of nutrients of symptomatic leaves from MY trees and green leaves from normal trees.	Dr's Hanold & Randles
1999 and 2000	Roadside transects in the South East of SA documenting the presence and absence of MY	John Choate
1998	Bark patch grafting experiment	Prof. John Randles, Mr Geoff Briant
1999	Collection of detailed distributional data on roads in the Keith-Bordertown area in five 1-km transects extending out to 100m into adjacent paddocks.	Dr Paton and Cutten
2000	Survey to identify affected species of plants.	Paton and Sarneckis (2000)
2001	Systematic survey to document the association between landuse, disturbance and the presence of MY.	Dr David Paton, third year University of Adelaide undergraduates and Hafiz Stewart of DEH

2001-2002	Testing of the phytoplasma hypothesis	Dr Karen Gibb, Prof John Randles, Dr.s N. Habili & D. Hanold
2000	Tissue culture studies.	Dr Dagmar Hanold and Prof John Randles
2000	Identification of saprophytes in cultures of bacteria and fungi from MY field trees	Barbara Hall, from the South Australian Research and Development Institution.
1999 - 2002	Electron microscopy	Prof Randles, Dr Gowanlock and Dr Hanold
2001-2002	Preliminary test of the soil borne hypothesis.	Department of Environment and Heritiage
2000 - 2001	molecular assay for eucalypt tissue	D. Hanold & jwr
2000 - present	Testing of hypothesis that MY is caused by a virus-like organism.	Dr Hanold and Prof Randles
2000 - 2001	Opportunistic, qualitative survey of occurrence of trees expressing MY symptoms across southern Australian states.	Dr Hanold and Prof Randles
2002 - present	Characterisation and cloning of MY-RNAs	Dr Hanold & Prof Randles
	Herbicide theory and opportunistic surveys	Jim McNamara

Appendix 5. Identified Stakeholders

The following have been identified as beneficiaries of MY management or as possible sources of funding:

Local government
Agencies responsible for road transport
Regional management groups including catchment management bodies, water authorities, and Natural Resource Management groups
Industry Associations
NGOs
Research Institutions
Government
Philanthropy Institutions or Organisations
Farming Associations
Tourism Associations
Traditional Owners