

Pathology

PART I. PRINCIPLES

R.K. MITTAL and S.B. MATHUR

Indian Council of Agricultural Research,
New Delhi, India, and
Danish Government Institute of Seed Pathology,
Denmark

PART II. PRACTICE

K. OLD and Z.Q. YUAN

CSIRO Forestry and Forest Products, Australia, and
University of Tasmania, Australia

PART I. PRINCIPLES

Seeds and seedlings are frequently affected by physical and physiological disorders and the diseases caused by fungi, bacteria, and viruses. Young plants or seedlings are particularly susceptible to a number of diseases because of their tender tissues and because they often have difficulty in establishing themselves. Health and vigor of seedlings and their further growth are to a considerable extent dependent on the quality of seeds. Since seedlings grown from the seeds are the primary source of planting stock and the improved seeds are expensive, it is necessary to investigate the seed and seedling pathogens and, if necessary, apply some control measures either before sowing the seeds or at seedling stage.

In the past, damping-off fungi, cone rust, a number of cone and seed insects in pines, and fungal damages to oak acorns, hazelnuts, chestnuts, walnuts, and seeds of birch and elm were considered the only major problems in seed and seedling production. Many fungi have now been isolated and studied for their effects on seeds of both conifer and hardwood tree species (Mittal and others 1990) and some have

caused considerable losses of seeds and seedlings. Loss of several kilograms of seed of *Pterocarpus indicus*, *Acacia auriculiformis*, and *Leucaena leucocephala*, at the Pantabangan Dam Watershed in the Philippines during 1979, has been reported by the National Irrigation Administration (Quiniones 1987). According to Chalermpongse and others (1984), in Thailand, the loss of seeds due to infection of *Botryodiplodia theobromae* on *Swietenia macrophylla* was 92 percent, *Colletotrichum gloeosporoides* on *Dalbergia cochinchinensis* was 4 percent, *Alternaria longissima* on *Bauhinia* sp. was 2 percent, *Pestalotiopsis* sp. on *Cassia bakeriana* was 6 percent, *Macrophoma* sp. on *Eucalyptus camaldulensis* was 1 percent, and *Fusarium* sp. on *Shorea obtusa* was 2 percent.

Deterioration of tree seeds by fungi involves problems differing in many aspects from those of grains. For example, tree seeds are exposed to many conditions before storage that permit the development of mold fungi. Cones are often collected in sacks and kept at collection points or processing plants for varying periods under conditions that favor fungal

development prior to seed extraction. After extraction, the seeds are dried and stored for varied periods until used in field or nursery. Hence, it is important to know the characteristics of fungi associated with important species, what damage they cause, where and when and under what circumstances the damage occurs, and what can be done to prevent the damage.

The science of tree seed pathology is still very young. The occurrence and distribution of most of the tree seed pathogens is well realized but there is very little understanding of their impact on seed production, seed quality, and seed viability. Interest in these problems has been growing steadily in the past few decades, but recently this interest has changed into serious concern mostly because of the problems encountered in the renewal and management of the forests, which are a major resource worldwide. Some information about the characteristics of seed-borne fungi is now available but is mostly related to temperate tree species. Therefore, in discussing the tropical tree seeds this information will also be briefly discussed at times for reference.

FUNGI OF FOREST TREE SEEDS

Fungi associated with tree seeds vary in different host species, in different regions, and in different years. Many of them are molds and develop on the seed surface only; some cause internal infections too. Nearly all seeds carry spores of various microscopic fungi either on the surface or within the seed. A superficial mycoflora is almost always found because of the ready adhesion of spores to the uneven surface of the seeds. Although the number of spores occurring varies considerably, as in Norway spruce and Scotch pine, it can be as high as 50 to 150,000 spores, and, in some seedlots, several hundred thousand spores per 1 g seed (Urosevic 1961). Under favorable conditions, some spores germinate, the mycelium penetrating into the cotyledons of the seed and feeding on the embryo.

Several kinds of fungi can be associated with tree seeds. Thus, there are species causing decay and reducing the germination of stored seeds, species attacking germinating seeds and seedlings, and other species that are more or less harmless, or at least appear to be so. Present knowledge does not permit precise separation of individual species of fungi occurring on seeds. However, it is clear that many species usually considered as unimportant and harmless can cause considerable damage under certain conditions, for example: unsuitable storage conditions, seeds of poor quality (immature, low vigor, or heavily molded), unsuitable growing conditions (involving moisture, temperature, or aeration), etc. Therefore, while eval-

uating the importance of these fungi, it is essential to consider the biology of the individual species of fungi.

Depending upon their location, the seed-borne fungi can, in general, be classified in two groups: externally seed-borne and internally seed-borne. The first group includes species of *Botryosphaeria*, *Botrytis*, *Fusarium*, *Mucor*, *Phialophora*, *Rhizopus*, and *Trichothecium*. They are not usually host specific and may involve more than one species. Some of the well-known internally seed-borne fungi include species of *Alternaria*, *Aspergillus*, *Botrytis*, *Botryodiplodia*, *Caloscypha*, *Cephalosporium*, *Fusarium*, *Phoma*, *Schizophyllum*, and *Sirococcus*. These may cause deterioration of seed quality and pre- or post emergence mortality of seedlings (Singh and Mathur 1993).

Urosevic (1961) provided instructions for health testing of oak acorns, including a key for distinguishing fungi from acorns, and divided the acorn mycoflora into two groups: (i) parasites and semiparasites such as *Ciboria batschiana*, *Ophiostoma spp.*, *Gloeosporium quercinum*, *Phomopsis quercella*, *Cytospora intermedia*, *Botrytis cinerea*, and *Pestalotia sp.*; and (ii) saprophytes including *Alternaria*, *Aspergillus*, *Fusarium*, *Penicillium*, *Trichoderma*, and others.

Based upon their pathogenicity, Sutherland (1995) classified the seed-borne fungi of conifers as (i) saprophytes or weak pathogens; (ii) pathogens such as the cold fungus *Caloscypha fulgens* which consistently kills seeds; (iii) pathogens mainly important as seedling pathogens, e.g. *Sirococcus conigenus*; and (iv) fungi, e.g. *Fusarium spp.*, whose pathogenicity depends upon factors including fungus species and pathogenic strain, and host and host stress.

DISEASES AND DAMAGES BY SEED-BORNE FUNGI

Symptoms of seed-borne diseases are usually divided into pre- and post emergence damping-off. The former consists of reduced emergence and decay of the radicle just emerged from the seedcoat; the latter is subdivided into root rot, cotyledon rot, and basal stem rot after the seedlings emerge from the soil. Reduction in seed germination, decay and loss of viability of seeds during storage, and the diseases of seedlings are among the major problems wrought by fungal pathogens.

GERMINATION REDUCTION

Inhibition of coniferous seed germination by widespread contaminants (Garbowski 1936, Rathbun-Gravatt 1931, Ten Houten 1939) and through artificially inoculated fungi (Fish-

er 1941, Timonin 1964) has been reported. Huss (1956) observed that the molding had virtually no effect on pine seeds of high viability, but poor quality seeds suffered a substantial reduction of germination. It has also been observed that the destruction of seeds largely depended on their growth rate, and as germination progressed, the resistance to destruction increased (Gibson 1957). The extremely common and numerous species of mold fungi, viz. species of *Mucor*, *Rhizopus*, *Trichothecium*, *Botrytis*, *Penicillium*, and others, which colonized the surface of *Quercus* acorns or got into the surface tissues, were found of secondary importance in the loss of germinability of acorns (Potlaichuk 1953). Gibson (1957), however, reported that the saprophytic fungi, viz. *Aspergillus* spp., *Mucor* spp., *Rhizopus* sp., *Trichoderma* sp., and *Cladosporium* sp., of the seedcoat microflora could, under favorable conditions, invade tissues of the germinating seeds and kill the seedlings of *Pinus patula*. The seedcoat microflora could thus be directly responsible for the weakening of seed vigor, predisposing it to the attack of soil-borne pathogenic fungi. Shea (1957) supported the view and mentioned that the influence of molds on seeds could vary considerably and their mere presence did not mean that they were harmful. However, Prisyazhnyuk (1960) mentioned that the greater the infection by fungi of seeds, the lower the germinability of seeds of *Pinus sylvestris*, *Larix sibirica*, *Picea abies*, and *Abies sibirica*. Rowan and De Barr (1974) observed extensive molding of three seed lots of slash pine during germination tests. Following the standard testing procedures, the seeds were cracked individually and almost 90 percent of them were found full, although the germination ranged from 31 to 79 percent only. *Fusarium solani* was obtained from the ungerminated seeds.

Out of the 12 *Fusarium* isolates tested through inoculation on *Pinus patula* seeds (Pawuk 1978), 3 reduced germination, whereas 9 increased percent damping-off but did not affect seed germination or seedling growth. *Leucaena* seeds infected by *Colletotrichum graminicola* failed to germinate, and if the infection was carried to the nursery, seedlings under moisture stress succumbed to the damping-off disease (Quiniones 1987). *Botryodiplodia theobromae*, which causes black dry rot in mahogany (*Swietenia macrophylla*) and caused 92 percent seed deterioration in Thailand (Chalermpongse and others 1984), did not affect germination but, after potting in the nursery, the rot developed during the hardening-off period of seedlings (Quiniones 1987).

Stratification, also known as moist cold prechilling of seeds, is commonly used to break dormancy in seeds, and to attain vigorous, speedy, maximum, and uniform germination for laboratory testing and green house and nursery sowing (Wang 1986). It is a common practice for most conifer and several hardwood seeds. Sutherland (1979) reported spread of

Caloscypha fulgens on seeds of several conifer species at low (3 to 5 °C) temperatures of stratification. The fungus mummifies the seeds, resulting in poor germination. Mittal and others (1987) observed the development and spread of fungi on *Pinus strobus* seeds during stratification. These fungi, generally, did not lower seed germination but diseased the germinants.

DECAY AND LOSS OF VIABILITY DURING STORAGE

Large quantities of high-quality seeds are required annually for artificial regeneration. In view of the lack of uniformity and predictability of cone and seed crops, bulk quantities of seeds are collected in good seed years and stored for use in intervening years to ensure a continuous supply of seeds for annual production of planting stock and for direct seeding. Storability of seeds is dependent upon temperature, time, relative humidity, and method of storage, as well as the moisture content of, and initial fungal inoculum on, seeds to be stored. Improper storage of cones, as reported by Shea (1960), caused heating of cones as a result of biological activity and these cones suffered more damage by fungi. Sixty percent of the *Pinus sylvestris* seeds stored in sacks; about 30 percent of seeds stored in boxes, bins, and tin drums; and only 10 percent stored in hermetically sealed vessels were infected with various fungi (Prisyazhnyuk 1960).

Quercus acorns lost up to 70 percent of their germination capacity during storage due to fungal infections (Potlaichuk 1953). Achenes of *Platanus occidentalis* stored at 2 °C showed no loss in germinability even after 7 months at 20 and 30 °C; however, germinability decreased and most fungi on achenes increased with increasing temperature, relative humidity, and time of storage (Fakir and others 1971).

In conifers, Lavender (1958) found no loss in germination capacity of *Pseudotsuga menziesii* seeds stored in cones up to 4 months at normal fall temperatures in an unheated warehouse. Bloomberg (1969) reported that seeds in *Pseudotsuga menziesii* cones stored for 225 days under operational conditions were free of diseases but, during germination tests after extraction, up to 56 percent of them became diseased. Rediske and Shea (1965) observed significant reduction in the same seed viability when freshly picked cones of high moisture content (60 percent) were sacked and stored in outside bins in the fall. However, full viability of these seeds could be retained for 3 years when stored in sealed cones at 0 °F (Barton 1954). When the same seeds were stored in canvas bags, germination was somewhat reduced within 6 months, and severely reduced after 12 months of storage. Gordon (1967) advised refrigeration of extracted seeds of *Pseudotsuga menziesii* immediately after their removal from the cone, to inhibit further fungal

activity within the seed. However, Rediske and Shea (1965) supported Schubert's (1960) observation on *Pinus monticola* seeds: that fungi remained active in pine seed at temperatures below the freezing point.

There have been suggestions for storing tree seeds at subfreezing temperatures to maintain their germinability (Willan 1985). Cryopreservation of seed germplasm at or near the temperature of liquid nitrogen has the potential for reducing deterioration of seed to such a low level that essentially perpetual preservation can be achieved (Stanwood 1985). However, based on artificial inoculations of *Fusarium sporotrichioides* and *Mucor hiemalis* on water-soaked *Pinus strobus* seeds before storing at -18, -80, -145, and -197 °C for 35 days, Mittal and Wang (1989) inferred that storing tree seeds at ultra-low temperatures will not eliminate contaminating fungal pathogens.

SEEDLING DISEASES

Damping-Off

Pre- and post emergence damping-off caused by various fungi are the most dangerous diseases affecting conifers as well as hardwood species. Quiniones (1987) reported establishment of *Fusarium solani*, a soil inhabitant, in the seeds of *Leucaena* and *Agathis* which caused post emergence damping off in the nursery and in the outplanted seedlings.

Seedling Blight

Sirococcus blight caused by a seed-borne fungus *Sirococcus strobilinus* is an important disease of seedlings of several spruce and pine species, and of *Pseudotsuga menziesii* throughout the northern Temperate Zone (Sutherland 1985). In this case, the pathogen attacked very young seedlings, killing the primary needles from the base upward. Dead seedlings remained upright, and small, black pycnidia usually formed at the base of infected needles. Diseased seedlings usually occurred randomly, characteristic of seed-borne diseases.

Seedling Wilt

Another important disease transferred by seeds is the tracheomycosis wilting of plants (Urosevic 1964). This symptom can be elucidated as a reaction of the host to the irritation by the parasite wherein the typical blocking of tracheae by thalli, and a yellowish brown, rubber-like substance filling the adjacent parenchymatous cells, are produced.

Reduced seedling height and leaf symptoms (chlorotic and necrotic lesions and malformed leaves) are also sometimes observed in seedlings raised from the fungus-inoculated seeds of *Acer saccharum* (Janerette 1979), *Picea glauca*, and *Pinus strobus* (Mittal and Wang 1986, 1993).

TESTING OF SEED-BORNE FUNGI

Seed-borne fungi testing includes isolation and study of fungi during cone collection and processing, seed extraction, processing, storage, germination, and seedling growth. Unlike with agricultural crops, methods for health testing of most forest tree seeds have not been standardized, and the testing is usually done using normal procedures of moist blotter and agar plates. Singh and Mathur (1993) have elaborated on the seed-health testing methods including direct observation, washing test and incubation methods (blotter and agar plate) for seeds, and seedling symptom test and growing-on test for seedlings. Some special methods like dilution plate method, ultrasound technique, isozyme patterns, seed tissue excision, seed sectioning, radiography, and ELISA technique have also been discussed. The ISTA recommendations for germination testing, which are more clearly available for agricultural crop seeds, are usually followed as standard practice. Seed size and, sometimes, unavailability of tree seeds in large quantities, makes it difficult to use large numbers of seeds in testing. Therefore, it is important to find out how many seeds of a tree species should be tested in how many replicates.

For many years it has been difficult to secure accurate, maximum germination of all viable seeds or achieve the true planting value of forest tree seeds. For example, the dormant nature of the seeds of *Abies balsamea*, *A. fraseri*, and other *Abies* species, which required a moist prechill treatment of 21 to 28 days or more at 3 to 5 °C, together with fungal contamination and growth during the 2-month overall test duration, have been responsible for sometimes erroneous, erratic, or negative germination results. Another problem generally encountered in seed testing involves seed pretreatment. For this purpose, Wall (1974) and several others used 0.1-percent mercuric chloride solution for 2 minutes, following the ISTA recommendations for seed-health testing of agricultural crop seeds. Wall (1974) also used 0.5-percent sodium hypochlorite solution for 2 to 3 minutes for surface sterilization of diseased red pine seedlings. Mittal (1995) recommended treatment with 2-percent sodium hypochlorite solution for 10 minutes for

Picea glauca and *Pinus strobus* seeds for greenhouse sowing as well as for laboratory testing. Thus, there existed different opinions on the type and duration of treatment that seeds should receive before testing.

For testing pathogenicity through artificial inoculation of seeds with some seed-borne fungi, different methods for seed inoculations are employed. Several workers have attempted rolling of seeds on fresh fungal cultures, whereas others used spore suspensions. At several platforms, a controversy existed over the method of inoculation and the testing environment, which need to be standardized for different types of seeds.

The reduction of germination under conditions of artificial infection does not correspond exactly to the reduction of germination that occurs under conditions of natural infection. Under natural conditions, the various microorganisms on seeds interact within themselves and with the microorganisms present in the soil or growing media. Such an interaction is often even antagonistic, which affects the ability of individual microorganisms to develop rapidly and to infect seeds. Often, in artificial inoculation studies, the conditions for facilitating microbial growth are provided. This suggests a need for testing the pathogenicity of various fungi in natural soils or growing media under greenhouse or field conditions (Mittal and Wang 1990).

FACTORS AFFECTING DEVELOPMENT AND SPREAD OF SEED-BORNE FUNGI

ABIOTIC FACTORS

Collection, Extraction, and Processing

The time, place, and method of collection of cones or seeds, and their subsequent handling during transport, extraction, and processing, affect the development and spread of mycoflora on tree seeds. It is generally presumed that cones and seeds acquire various fungi, including molds, while they are still on trees. *Pinus pinea* cones contained discolored, powdery seeds, in some of which the kernel was still sound but in others was blackened and completely destroyed by the grayish mycelial growth of *Alternaria alternata* (Sibilia 1927). Similarly, a pathogen *Coniothyrium* sp., established itself in and on the *Betula aleghanensis* seed before it fell to the ground (Shigo and Yelenosky 1963). However, Salisbury (1955) and Prisyazhnyuk (1960) reported that most of the individual seeds from tightly closed cones of conifers were completely free from molds. Seeds should be extracted from the cones immediately after harvest to minimize seed infection from the microbial popula-

tion already present on cones.

The later the oak acorns were collected from the fields, the more infected they were with fungi (Potlaichuk 1953). *Penicillium*, *Fusarium*, *Alternaria*, and *Trichothecium roseum* were found to be the most common fungi during acorn development. Development of *Cladosporium herbarum*, which was frequently encountered on the acorns in the first and last samples during their development, depended on the amount of rainfall; the highest occurrence was during the period of heavy rainfall.

Dewinging of the seeds can lead to considerable reduction in germination, presumably through damage to the testa and subsequent fungal invasion (Gordon 1967, Harding 1952, Huss 1956, James and Genz 1981). While studying the fungi associated with seeds of *Picea glauca* and *Pinus strobus* during cone processing and seed extraction, Mittal and Wang (1987) observed that the contamination occurred and spread during air drying and seed extraction processes, and that considerably more fungi occurred on both types of seeds after they were left on the forest floor for 15 days. Mojtahedi and others (1978) found that the wash water was a source of fungus contamination when fresh, uncracked pistachio nuts were tested for *Aspergillus flavus* and aflatoxin before and after a commercial washing treatment.

Storage Conditions

The moisture content of seeds, initial fungal inoculum on seeds, and the method of storage, all affect the spread of fungi on seeds during storage. Full viability of extracted Douglas fir seeds was retained for 3 years when stored in sealed cans at 0 °F. However, when stored in canvas bags, germination was somewhat reduced within 6 months and seriously reduced after 12 months (Barton 1954). Significant reduction in Douglas fir seed viability when freshly picked cones of high moisture content (60 percent) were sacked or stored in outside bins in the fall, was observed by Rediske and Shea (1965). Immediate gentle drying or refrigeration maintained viability.

While orthodox seeds, which can tolerate low moisture content and low storage temperatures and, therefore, can be stored successfully for often longer periods, the recalcitrant tree seeds are fast-perishable. Their high moisture content and storage often at ambient or relatively higher temperatures help in establishing several storage fungi. Fast deterioration of the recalcitrant seeds by these storage fungi is expected to be due to the debilitation of seeds caused by internal moisture stress generated within the cells or tissues, primarily as a result of the water-requiring process of vacuolation (Berjak 1996). This debilitation impaired phytoalexin synthesis by the seeds, facilitating proliferation of associated fungi or bacteria. Prevention of fungal activity during storage can be more easily achieved

by controlling the moisture content of the seeds than by controlling the storage temperature, because fungal activity is possible between -8°C and $+80^{\circ}\text{C}$ when the seed moisture content and the relative humidity of storage are high enough (Roberts 1972b). It is therefore important to find out the optimum moisture content and the storage temperature requirements for individual forest tree species.

Stratification

The killing of viable seeds of some conifer species has been particularly serious during the long, cold, prechill treatment at 3 to 5°C (Sutherland 1979). Mittal and others (1987) studied the development and spread of fungi on *Pinus strobus* seeds during stratification, though the former did not, usually, lower the seed germination in further testing. The prechilled seeds germinated vigorously and speedily, and therefore probably escaped the damages, supporting the views of Gibson (1957). However, some germinants failed to emerge completely from the seedcoat and others were damaged by top decays caused by *Alternaria alternata*, *Fusarium oxysporum*, and *Penicillium variabile* in the laboratory, possibly due to the high moisture content of the prechilled seeds and the high environmental temperature: the two important factors contributing to fungal development, spread, and infection (Mittal and Wang 1986). This suggested a need for treatment by some surface cleaning or sterilizing agent before stratifying the seeds, especially the highly dormant seeds.

Cultural Practices

Surface-sterilized seeds with pierced testa sowed in normal moist soil completely failed to germinate, presumably due to invasion by soil saprophytes (Gibson 1957). It seems possible that the destruction of seeds was facilitated by the proximity of a relatively large food base, the seed reserves adjoining the small volume of living tissue.

Sitka spruce seeds incubated at 10°C were highly susceptible to fungal attack because they remained dormant while the pathogen was growing at near its maximum rate (Salt 1967). Losses in nurseries are not necessarily related to the time taken for seedlings to emerge, but they are likely to be greater in places where fluctuations in temperatures over 10°C are usually less prevalent. Losses are expected to increase with earlier dates of sowing. Autumn sowing is most unreliable because in warm soil, the seeds germinate early and escape damage, whereas in cold soil they do not germinate until the following spring, and suffer maximum loss.

Most soil mixes for containerized conifers contain vermiculite or perlite incorporated with sphagnum peat. This

type of mix is usually well drained and acidic, the two factors that help reduce diseases (James 1985). Major groups of pathogens associated with nursery diseases are species of *Fusarium* and watermolds, such as *Pythium* and *Phytophthora*. Although watermolds may be seed-borne, they are more often introduced into container nurseries through contaminated irrigation water. These fungi cause disease on very young seedlings, and are favored by poorly drained soil mixes and prolonged wet conditions in the greenhouse.

BIOTIC FACTORS

Fungal colonization of conifer seeds is facilitated by insects like seedbugs, and by squirrel damages to seeds (James 1985, Rowan and De Barr 1974, Sutherland 1979). Elaborating on the insect-fungal interaction during infection of oak acorns, Urošević (1959) reported that at the time of oviposition, various fungi were introduced by the insects into the acorns. These fungi then penetrated into the mature acorn tissues. Death of the acorns was thus brought about not only by the damage caused by the larvae of the weevil, but also by fungi accompanying the weevil. The penetration of the acorn by the fungal mycelium entailed negative effects more rapidly and more injurious than did the maturation feeding of the larvae. Acorns thus affected might represent a dangerous focus of infectivity during both initial storage and long-term storage of the acorns.

MANAGEMENT OF SEED PATHOGENS

SEED COLLECTION, PROCESSING, AND STORAGE

Collection of seeds from healthy, disease-free areas or orchards; collection from healthy trees, healthy cones or acorns at the appropriate time; collection from tree and not from ground or from squirrel caches, etc.; transport of cones or seeds in well-aerated, clean, dry containers or bags; avoiding damage to seeds during extraction and processing; and use of optimum seed extraction and storage conditions, all need be studied for different forest tree species and considered for prevention of fungal infections on seeds.

SURFACE TREATMENT

Although adverse effects on seed germination have sometimes been reported, seed treatment with sterilants to reduce or

eliminate fungal contamination has been considered necessary for production of healthy seedlings at several nurseries. For sterilizing conifer seeds with minimal stimulation or retardation to them, an immersion of the seeds in a commercial detergent followed by treatment with 30-percent hydrogen peroxide has been recommended (Gordon 1967). Hydrogen peroxide treatment (30 percent for 45 minutes) improved the total germination from 47 to 80 percent and from 25 to 61 percent in the 2, poor-quality, unstratified seeds of *Pinus taeda* (Mason and Van Arsdel 1978). Water treatment at 57 °C for 10 minutes was found quite effective in eliminating large numbers of seed-borne fungi of *Pinus roxburghii* and *P. wallichiana* (Munjaj and Sharma 1976). Delatour and others (1980) also suggested hot water (44 °C for 8 hours) soak treatment for killing *Ciboria batschiana* in *Quercus* acorns.

CHEMICAL CONTROL

Coating seeds with a repellent against birds and small rodents, and a fungicide against damping-off has been a common practice in forest tree nurseries at several places. Although a lot of literature on chemical seed treatment control of seed-fungi has accumulated, most studies were made on conifers. The sulphuric acid treatment to *Araucaria excelsa* seeds, which has been prescribed by quarantine regulations against *Cryptospora longispora*, was found effective in eradicating the seed-borne fungi but the acid was detrimental to seed germination (Khan and others 1965). A 70 to 75 percent dust of PCNB applied to the seed of balsam, Fraser, and grand firs gave excellent control (100 percent) of *Rhizoctonia solani* without any injury to the germinating seedlings. Mittal and Sharma (1981), based on their observations with different tree species (*Cedrus deodara*, *Eucalyptus citriodora*, *E. hybrid*, *Pinus roxburghii*, *P. wallichiana*, and *Shorea robusta*), suggested that Brassicol, Bavistin SD, and Dithane M-45, as seed dressers, could be used to effectively control most of the common seed-borne fungi of these tree species. For control of a common fungus, *Aspergillus niger*, on the seeds of *Shorea robusta*, seed treatment with Bavistin SD or Brassicol was most effective (Mittal and Sharma 1982).

Effective control of several fungi, such as *Botryodiplodia theobromae*, *Colletotrichum gloeosporoides*, *Fusarium* spp., *Macrophomina phaseolina*, *Pestalotia* sp., *Phoma* sp., and *Phomopsis* sp., on the seeds of *Acacia auriculiformis*, *Albizia* spp., *Gmelia arborea*, *Leucaena leucocephala*, several *Pinus* spp., *Pithecelobium dulce*, *Pterocarpus indicus*, *Cedrella odorata*, and *Grevillea robusta*, has been successfully achieved through the combined use of Benlate (0.15 percent) and Dithane M 45 (0.15 percent) (Cortiguerra 1985, Pacho 1985).

Seed treatments with oil, talc, and dye have also been found beneficial but much less so than treatment with Thiram

(a fungicide) for Sitka spruce (*Picea sitchensis*) (Salt 1967).

Since detrimental effects of chemical seed treatment on seed germination and seedling quality have also been reported (James 1983), it is desirable that lower concentrations, which should not be phytotoxic, be tried. Kozlowski (1986) reported that Captan at concentrations up to 2500 ppm did not affect seed germination of *Pinus resinosa*; however, concentrations of 500 ppm or higher injured roots, stems, and cotyledons within 13 days. Root injury consisted of collapse of root hair cells, epidermal cells, and cortical cells, and the cotyledon injury included the collapse of epidermal and mesophyll cells. Similar observations were made earlier by Cram and Vaartaja (1956) and Vaartaja (1964).

LEGISLATION

Vigorous implementation of the seed laws, such as the Seed Acts and Seed Certification Programmes for quality evaluation and management, and the standards for seed collection, extraction, storage, and movement, is needed to avoid the seed problems.

CONCLUSION

There is an increasing awareness worldwide that unless we intensify efforts at gene conservation, reforestation, and intensive forest management, serious depletion of the world's forests will result. Although reforestation is recognized as an essential activity, an adequate supply of seeds of high quality and high genetic potential is often a limiting factor in many countries. This emphasizes the need for organized seed production and seed research to resolve many problems related to reforestation.

Several fungi have been studied on tree seeds; they vary in different host species, in different regions, and in different years. Even the detrimental effects to seeds during germination and storage, and to seedlings in nurseries, vary in different host species and environments. With the favorable environment in the Tropics, viz. high atmospheric temperatures coupled with high humidity, damage to seeds and seedlings is greater there. Biotic factors like squirrel and seedbug damages, and abiotic factors like time and method of collection, shipment, extraction, processing, testing, and storage of seeds, all affect the occurrence of fungi in seeds. Improvement in these practices, use of surface sterilants and/or fungicides, and following legislated practices like quarantine will help in the worldwide management of seeds.

PART II. PRACTICE

Australia is the origin of a unique and extensive resource of tree and shrub species which have proven to be of great value for the establishment of plantations in many parts of the world. Examples include approximately 5 million ha fast-growing eucalypt plantations in Brazil; the *Acacia mangium* resource, almost 1 million ha, recently established in Indonesia; *Acacia saligna*, planted extensively in north Africa and the Middle East as a fodder tree; *Acacia colei*, planted around villages in semiarid Niger to provide edible seed to supplement inadequate diets (Harwood 1994), and the extensive plantings of *Casuarina equisetifolia* on sandy shorelines of southern China and Vietnam for typhoon protection and a wide range of timber and nonwood benefits (Nguyen 1996).

Although Australian native trees, especially eucalypts, have been grown as exotics for more than a century, the area of plantations has expanded rapidly during the last 30 years. This expansion has been driven by the development of hardwood pulp as a major international commodity to meet the increasing demand for paper, and the widespread adoption of Australian trees for community forestry in Asia and parts of Africa. Having evolved in a continent characterized by climatic extremes and infertile soils, Australian native trees have proved to be well adapted to cultivation as multipurpose trees on degraded soils and provide a wide range of products including timber, poles, fuel, and oils.

The Australian Tree Seed Centre (ATSC), part of CSIRO Forestry and Forest Products, has acted for 35 years as a national seed bank, supplying seed to researchers in Australia and over 100 other countries. The seed originates in natural forests but over the last decade ATSC has been complementing these collections with seed-orchard seed. Seed orchards have been established in tropical and temperate Australia, and in several overseas countries in southern and Southeast Asia and Oceania, in collaboration with a wide range of agencies.

In 1987 research was initiated by ATSC to investigate the presence of fungal pathogens in stored seed. Although seed dispatched overseas by ATSC is routinely treated to meet the phytosanitary requirements of the recipient country, there was little information on the seed pathology of the three most important Australian native genera grown in plantations, domestically and overseas, namely *Eucalyptus*, *Acacia*, and *Casuarina*.

This contribution to the chapter on seed pathology reviews the world literature on the seed pathology of those eucalypts, acacias, and casuarinas which are grown on a significant scale as plantation species in the tropics, and highlights some issues related to quarantine and the movement of pathogens internationally.

STORAGE FUNGI AND SEED-BORNE PATHOGENS

Most seeds carry spores of various fungi, either on the surface or within the tissues, and counts as high as 150,000 spores per tree seed have been reported (Anderson 1986). Some seed-borne fungi can cause the death of seeds and seedlings whereas other fungi, for example species of *Aspergillus*, *Penicillium*, *Chaetomium*, *Rhizopus*, and *Trichoderma*, which are the genera most often isolated from seed samples of a wide range of species (Mohan and Sharma 1991, Yuan and others 1990), are saprophytes. If improperly stored, the growth of saprophytic fungi on seed can drastically reduce viability, but with a few exceptions, e.g. *A. niger* (Yuan and others 1997), they are rarely implicated in causing the death of seedlings.

Much of the literature on seed-borne fungi of these three tree genera consists of lists of fungal species with little information as to their pathogenic status. There are relatively few reports in the literature where isolation of putative pathogens from seed of eucalypts, acacias, and casuarinas has been complemented by inoculation tests to establish their pathogenicity. Some examples where this has been carried out include Bhawani and Jamaluddin (1995) who tested the pathogenicity of *Curvularia lunata* to *Acacia nilotica*; Harsh and others (1992) who found that a *Verticillium* sp. present in seed samples caused a post-emergence damping-off of seedlings; Saxena (1985) who investigated seedling mortality of *Eucalyptus* sp.; and Yuan and others (1990) who isolated 25 fungal genera representing at least 38 species from seed lots of *Acacia* spp., *Casuarina* spp., and *Eucalyptus* spp. and tested the pathogenicity of 14 fungal species by inoculating *A. auriculiformis*, *C. cunninghamiana*, and *E. camaldulensis*. A similar study was subsequently carried out on 10 seedlots of *E. pellita*, a tree species of increasing importance for plantations in the humid Tropics (Yuan and others 1997).

EUCALYPTUS SEED MYCOFLORA AND SEED-BORNE PATHOGENS

Lists of fungi isolated from samples of eucalyptus seed are often included with records from a range of other tropical tree species (table 1). Examples include Mohan and Sharma

Table 1

Pathogenic Fungi Associated with Tropical Eucalypt Seeds

Fungus	Host	Country	Reference(s)
<i>Botryodiplodia</i> sp.	<i>E. grandis</i>	Uruguay	Mittal and others, 1990
<i>Botrytis cinerea</i>	<i>E. grandis</i>	India	Mohan and Sharma, 1991
<i>Colletotrichum</i> sp.	<i>E. citriodora</i>	India	Mohan and Sharma, 1991
<i>Coniella australiensis</i>	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Curvularia eragrostidis</i>	<i>E. alba</i>	Thailand	Pongpanich, 1990
	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Curvularia fallax</i>	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Curvularia geniculata</i>	<i>E. tereticornis</i>	India	Reddy and others, 1982
<i>Curvularia inequalis</i>	<i>E. citriodora</i>	India	Mittal and others, 1990
<i>Curvularia lunata</i>	<i>E. camaldulensis</i>	Thailand	Pongpanich, 1990
	<i>E. grandis</i>	Thailand	Pongpanich, 1990
	<i>E. tereticornis</i>	Thailand	Pongpanich, 1990
	<i>E. globulus</i>	India	Mohan and Sharma, 1991
	<i>E. grandis</i>	India	Mohan and Sharma, 1991
	<i>E. tereticornis</i>	India	Mohan and Sharma, 1991
	<i>E. camaldulensis</i>	Australia	Yuan and others, 1990
	<i>E. grandis</i>	Australia	Yuan and others, 1997
	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Curvularia pallescens</i>	<i>E. alba</i>	Thailand	Pongpanich, 1990
	<i>E. camaldulensis</i>	Thailand	Pongpanich, 1990
	<i>E. robusta</i>	Thailand	Pongpanich, 1990
	<i>E. globulus</i>	India	Mohan and Sharma, 1991
	<i>E. grandis</i>	India	Mohan and Sharma, 1991
<i>Curvularia pubescens</i>	<i>E. citriodora</i>	India	Mittal and others, 1990
<i>Curvularia senegalensis</i>	<i>E. camaldulensis</i>	Australia	Yuan and others, 1990
	<i>E. nitens</i>	Australia	Yuan and others, 1990
	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Curvularia verruculosa</i>	<i>E. grandis</i>	India	Mohan and Sharma, 1991
<i>Cylindrocladium clavatum</i>	<i>E. tereticornis</i>	India	Mohan and Sharma, 1991
<i>Drechslera australiensis</i>	<i>E. grandis</i>	India	Mohan and Sharma, 1991
	<i>E. tereticornis</i>	India	Mohan and Sharma, 1991
	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Drechslera halodes</i>	<i>E. saligna</i>	India	Reddy and others, 1982
	<i>E. tereticornis</i>	India	Reddy and others, 1982
<i>Drechslera rostrata</i>	<i>E. grandis</i>	India	Mohan and Sharma, 1991
	<i>E. tereticornis</i>	India	Mohan and Sharma, 1991
<i>Fusarium equiseti</i>	<i>E. grandis</i>	India	Mohan and Sharma, 1991
	<i>E. tereticornis</i>	India	Mohan and Sharma, 1991
	<i>E. deglupta</i>	Philippines	Mittal and others, 1990
<i>Fusarium moniliforme</i>	<i>E. camaldulensis</i>	Thailand	Pongpanich, 1990
	<i>E. grandis</i>	India	Mohan and Sharma, 1991
	<i>E. tereticornis</i>	India	Mohan and Sharma, 1991
	<i>E. grandis</i>	Uruguay	Mittal and others, 1990

Table 1 (continued)

Fungus	Host	Country	Reference(s)
<i>Fusarium oxysporum</i>	<i>E. deglupta</i>	Thailand	Mittal and others, 1990
<i>Fusarium poae</i>	<i>E. alba</i>	India	Mohan and Sharma, 1991
<i>Fusarium semitectum</i>	<i>E. camaldulensis</i>	India	Mohan and Sharma, 1991
	<i>E. camaldulensis</i>	Egypt	Mittal and others, 1990
<i>Fusarium solani</i>	<i>E. citriodora</i>	India	Mittal and others, 1990
<i>Fusarium</i> sp.	<i>E. camaldulensis</i>	Australia	Yuan and others, 1990
	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Harknessia fumaginea</i>	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Harknessia hawaiiensis</i>	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Macrophomina phaseolina</i>	<i>E. grandis</i>	India	Mohan and Sharma, 1991
	<i>E. tereticornis</i>	India	Mohan and Sharma, 1991
<i>Macrophomina</i> sp.	<i>E. camaldulensis</i>	Thailand	Pongpanich, 1990
<i>Pestalotiopsis disseminata</i>	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Pestalotiopsis funerea</i>	<i>E. alba</i>	India	Mittal and others, 1990
	<i>E. grandis</i>	Uruguay	Mittal and others, 1990
<i>Pestalotiopsis mangiferae</i>	<i>E. tereticornis</i>	India	Reddy and others, 1982
<i>Pestalotiopsis neglecta</i>	<i>E. pellita</i>	Australia	Yuan and others, 1997
<i>Phomopsis</i> sp.	<i>E. citriodora</i>	India	Mohan and others, 1991
<i>Ramularia</i> sp.	<i>E. crebra</i>	Australia	Drake, 1974
	<i>E. melanophloia</i>	Australia	Drake, 1974
<i>Verticillium albo-atrum</i>	<i>E. grandis</i>	India	Mohan and others, 1991
<i>Verticillium</i> sp.	<i>E. grandis</i>	Uruguay	Mittal and others, 1990
	<i>E. hybrid</i>	India	Harsh and others, 1992

(1991), who also highlighted issues of seed collection processing, storage, seed health testing, treatment and certification and indicated where improvement was needed; Agmata (1979), who provided the first list of tree seed-borne fungi in the Philippines; Mittal and others (1990), who compiled a world checklist of microorganisms associated with tree seed; and Richardson (1983) who provided an annotated list of seed-borne diseases. Lists of seed mycoflora for *Eucalyptus* spp. alone are provided by Sharma and Mohan (1980), Tiwari and Sharma (1981), and Reddy and others (1982).

Several authors have demonstrated pathogenesis of seed-borne fungi, either by isolation of putative pathogens from both seed and blighted seedlings, or by inoculation studies. Mittal (1986) and Mittal and Sharma (1982) studied the mycoflora of *Eucalyptus* hybrid (predominantly *E. tereticornis*) and *E. citriodora* (syn. *Corymbia citriodora*) and also the means of controlling pathogenic fungal species. Saxena (1985) detected 30 species of fungi on seed of *E. grandis* and related seedling mortality to seed mycoflora, and Michail and others (1986) report-

ed on fusarium postemergence damping-off of *Eucalyptus* spp. and its control. Four potentially pathogenic species, *A. niger*, *Fusarium* sp., *Penicillium canadense*, and *Rhizopus oryzae*, were found on seed of *E. citriodora* (Mittal and Sharma 1982). Drake (1974) found that a species of *Ramularia* infected unopened capsules of *E. crebra* and *E. melanophloia* while still borne on the tree, and caused up to 50-percent sterility.

Yuan and others (1990) inoculated *E. camaldulensis* seed with 14 putative pathogens isolated from seed and recorded seedling emergence compared to controls. All fungi except *Botrytis cinerea* and *Cytospora* sp. reduced emergence, with *Fusarium*, *Curvularia*, and *Pestalotiopsis* spp. being the most pathogenic. In a later study with *E. pellita* (Yuan and others 1997), *A. niger*, *Dreschlera australiensis*, *Harknessia fumaginea*, and *Pestalotiopsis disseminata* all reduced germination and/or caused seedling mortality postemergence compared to controls. *Coniella australiensis*, a widespread leaf pathogen of eucalypts, was also isolated for the first time from seed. It was suggested that these fungi may have originated from leaf debris

within the samples and that such tissue offers a possible means for dissemination of foliar and stem pathogens with seed.

RISKS FROM THE DISSEMINATION OF PATHOGENS WITH EUCALYPT GERmplasm

In a publication outlining technical guidelines for the safe movement of germplasm of *Eucalyptus* (Ciesla and others 1996), 32 pathogenic fungi were listed as being associated with eucalyptus seed. A revised list is given as table 1. All these fungi are generally regarded as being widely distributed geographically, and the presence of such pathogens in seed lots represents little in the way of quarantine hazard. Foliar pathogens, such as *Mycosphaerella* spp., *Kirramyces* spp., and *Aulographina* sp. (which are specialized pathogens found in most parts of the world where eucalypts are grown) were either absent from seed lots or escaped detection. It can be reasonably assumed that these pathogens have originated in Australia and have been disseminated internationally with seed or vegetative plant material, perhaps many decades ago. Another possibility is that fungal pathogens of Myrtaceae, which are widely grown as crop plants, e.g. guava and cloves; or form part of the indigenous flora, e.g. in Southeast Asia, South America, and Africa; could adapt as pathogens of eucalypts and may pose a threat to a wide spectrum of native vegetation if introduced in Australia. The occurrence of guava rust, *Puccinia psidii*, on eucalypts in South and Central America (Ferreira 1983) has created a particular risk as the fungus is damaging to eucalypt plantations. Stringent controls are in place governing the importation of eucalypt seed to Australia from South America.

ACACIA SEED MYCOFLORA AND SEEDBORNE PATHOGENS

As noted above for eucalyptus, records of the mycoflora of seeds of *Acacia* spp. are commonly combined with lists for other tree species (Dayan 1986). In a recent report on surveys of tropical acacia diseases by Old and others (1997b) some reference is made to seedling diseases, notably those recorded in Thailand (Pongpanich 1997), but the emphasis of the report is on diseases of plantations and native stands.

There are very few reports in the literature of seed-borne

fungi causing diseases of acacia seedlings. *Curvularia lunata* has been recorded by Bhawani and Jamaluddin (1995) as causing shoot dieback of seedlings of *A. nilotica*. This fungus has been recorded previously from seed of *A. auriculiformis* and recently from *A. crassicarpa* and *A. aulacocarpa*. Several of the pathogens recorded by Mohanan and Sharma (1988) as causing diseases of exotic acacias in India are putative seed-borne organisms. Table 2 lists plant pathogenic species recorded on seed lots of tropical acacias.

Yuan and others (1990) inoculated *A. auriculiformis* with the same 14 seed-borne fungi used to inoculate eucalypt seed, with similar results. *Fusarium* spp., *Dreschlera* sp., *Curvularia* spp., and *Pestalotiopsis* spp. caused the most severe pre-emergence blight of inoculated seedlings. An unpublished study of the mycoflora of *A. aulacocarpa*, *A. auriculiformis*, *A. crassicarpa*, and *A. mangium* seed by Yuan and others (1997) has indicated the presence of *Botryodiplodia* (syn. *Lasiodiplodia*) spp., *Curvularia* spp., *Dreschlera* spp., *Fusarium* spp., and *Pestalotiopsis* spp.

The lists of pathogenic fungi on acacia trees in native stands and plantations in northern Australia and in India presented by Old and others (1997a) and Sharma and Maria Florence (1997) include species of *Pestalotiopsis*, *Phoma*, *Phomopsis*, *Lasiodiplodia*, and *Curvularia*, which can be seed-borne. The detailed etiology, however, of the diseases of acacias recorded in both reports is largely unknown. Old and others (1997a) gave a brief account of a severe disease outbreak on *A. mangium* in 1992 in northern Australia caused by a *Cercospora* spp. The pattern of disease incidence suggested that the origin of the outbreak was in the nursery where seedlings were raised, and plants grown from seed imported from Papua New Guinea initially showed the highest level of disease. Although severe damage occurred in several plantations, the disease was present at very low levels during the following year and was undetectable subsequently. Detailed analysis of all seed lots used in the plantings failed to isolate *Cercospora* sp. (Old and others 1993). In view of several well-known examples of seed-borne *Cercospora* spp., this could have been the origin of the disease. Alternatively, the pathogen is indigenous and occurring undetected on native *Acacia* species in the region.

RISK FROM THE DISSEMINATION OF ACACIA PATHOGENS WITH GERmplasm

As with eucalypts, the species recorded so far on the seed of tropical acacias are regarded as being already widely distrib-

uted throughout the regions where tropical acacias are grown. Also, they are generally pathogens with a broad spectrum of hosts. Unlike eucalypts, the seed of many acacia species are relatively large (10 mg to 1 g) and the presence of retained floral parts such as the aril offers niches for the carriage of saprophytic growth of facultative pathogens. (Table 2)

The recent report of an international workshop on diseases of tropical acacias (Old and others 1997b) gave an account of the status of a range of pathogens and the diseases they cause in Australia, India, and Southeast Asia. The stimulus for the workshop was the rapid expansion of plantations of tropical acacias, especially in Indonesia, Malaysia, and Thailand. Every year several tons of seed of *A. mangium*, the most widely grown species, are collected from native stands or seed orchards and dispatched between countries of the region. There seems little doubt that this practice carries significant risk of transporting pathogens on seed or associated plant debris. The practice of immersing seeds in boiling water prior to sowing to break dormancy may reduce the likelihood of chance contaminants being spread, but there is an urgent need for more information on the diseases of these species and the possibilities of seed-borne spread.

CASUARINA SEED MYCOFLORA AND PATHOGENS

There is very little information in the literature on seed-borne diseases of tropical *Casuarina* spp. Mittal and others (1990) listed 18 fungi associated with *C. equisetifolia*, 10 of these being putative pathogens (table 3). Sahai and Mehrotra (1982) examined the mycoflora of forest tree seeds including *Casuarina* (species not stated) but only one of those recorded (*Fusarium semitectum*) can be regarded as a putative pathogen. Yuan and others (1990) sampled the mycoflora of *C. equisetifolia* seed but recorded only four recognized pathogens including *A. niger*, *Botrytis cinerea*, *Curvularia senegalensis*, and *Pestalotiopsis* spp. *Alternaria alternata*, *B. cinerea*, *Pestalotiopsis* spp., and a *Phoma* spp. were isolated from *C. cunninghamiana*. The most useful summaries of diseases of *Casuarina* are those of Mohanan and Sharma (1989, 1993). In the latter publication they noted that information on seedling diseases in India is meager despite the raising of casuarina seedlings in that country for nearly a century. They reported that the soil-borne pathogens *Rhizoctonia solani* and *Macrophomina phaseolina* are the main pathogens of seedlings. Of these, *M. phaseolina* has been listed as seed borne but is actually a very common

soil-borne fungus with a very wide geographical distribution, causing the well-known charcoal root rot of a broad range of tree species (Srivastava and Kalyani 1990).

Of more interest to seed pathology is the record by Mittal and others (1990) of *Phomopsis casuarinae* as being seed borne. This is a well-known pathogen, recorded in Portugal (Dos Santos 1966) and in India (Mohanan and Sharma 1993). The fungus causes stem cankers and foliar lesions, and is probably more severe on stressed trees, sharing this characteristic with *Botryosphaeria* spp (Pongpanich and others 1996) and *Trichosporum vesiculosum* (Mohanan and Sharma 1993, Narayanan and others 1996). Table 3 presents a list of diseases of Casuarina for which an association with seed has been established or may be inferred.

RISKS FROM THE DISSEMINATION OF PATHOGENS WITH CASUARINA GERMPLASM

One of the more significant diseases of *Casuarina equisetifolia* is the blister-bark pathogen *Trichosporum vesiculosum* (*nom. illegit.*). The status of this organism as a pathogen has been questioned (Boa and Ritchie 1995). However, Narayanan and Sharma (1996) and Narayanan and others (1996) summarized the available information which strongly points to the fungus being a damaging pathogen of stressed trees. The fungus spreads by production of very large quantities of sooty spores within blisters under the outer bark, which rupture to release spores. The etiology of the disease is imperfectly understood, but it is likely that wound infection is the major means of spread with further movement within an affected stand by root-to-root contact (Narayanan and others 1996).

Until 1994 the fungus was known as a pathogen of casuarina only in India (Bakshi 1951) and in Mauritius (Orlan 1961). However, blister-bark disease has been found in central Vietnam (Sharma 1994) and was identified at two locations in Thailand in 1995 in replicate plantings of an international *Casuarina* provenance trial (Pongpanich and others 1996). The seed for this trial had originated in many countries, including India, and although there is no information as to the origin of the outbreak, it is possible that the fungus was seed borne. *T. vesiculosum* would not be detected by standard seed testing methods; however, its fecundity suggests that chance contamination of seed could have been the origin of the simultaneous appearance of this disease in two locations in Thailand separated by approximately 200 km. There is a need for further

Table 2

Pathogenic Fungi Associated with Tropical Acacia Seeds

Fungus	Host	Country	Reference(s)
<i>Botryodiplodia theobromae</i>	<i>A. confusa</i>	Philippines	Agmata, 1979
(<i>syn. Lasiodiplodia theobromae</i>)	<i>A. auriculiformis</i>	Thailand	Chalermpongse and others, 1984
<i>Botryodiplodia</i> sp.	<i>A. auriculiformis</i>	Australia	Yuan and others, 1997
<i>Colletotrichum gloeosporioides</i>	<i>A. auriculiformis</i>	India	Mohanan and Sharma, 1988
<i>Colletotrichum</i> sp.	<i>A. auriculiformis</i>	Thailand	Pongpanich, 1997
<i>Curvularia brachyspora</i>	<i>A. auriculiformis</i>	Thailand	Chalermpongse and others, 1984
<i>Curvularia eragrostidis</i>	<i>A. auriculiformis</i>	Australia	Yuan and others, 1997
<i>Curvularia lunata</i>	<i>A. auriculiformis</i>	Australia	Yuan and others, 1990
	<i>A. catechu</i>	India	Vijayan, 1988
	<i>A. confusa</i>	Philippines	Agmata, 1979
	<i>A. crasscarpa</i>	Australia	Yuan and others, 1997
<i>Curvularia pallescens</i>	<i>A. auriculiformis</i>	Philippines	Mittal and others, 1990
<i>Curvularia senegalensis</i>	<i>A. auriculiformis</i>	Australia	Yuan and others, 1990
<i>Curvularia</i> sp.	<i>A. auriculiformis</i>	Thailand	Pongpanich, 1997
<i>Cylindrocladium</i> sp.	<i>A. auriculiformis</i>	Thailand	Pongpanich, 1997
<i>Diplodia</i> sp.	<i>A. crasscarpa</i>	Australia	Yuan and others, 1997
<i>Drechslera</i> spp.	<i>A. auriculiformis</i>	Thailand	Pongpanich, 1997
	<i>A. crasscarpa</i>	Australia	Yuan and others, 1997
<i>Fusarium equiseti</i>	<i>A. catechu</i>	India	Vijayan, 1988
<i>Fusarium moniliforme</i>	<i>A. raddiana</i>	Israel	Mittal and others, 1990
<i>Fusarium oxysporum</i>	<i>A. catechu</i>	India	Vijayan, 1988
<i>Fusarium semitectum</i>	<i>A. auriculiformis</i>	Philippines	Mittal and others, 1990
	<i>A. auriculiformis</i>	India	Mohanan and Sharma, 1988
	<i>A. modesta</i>	India	Mittal and others, 1990
<i>Fusarium solani</i>	<i>A. catechu</i>	India	Vijayan, 1988
	<i>A. holosericea</i>	Australia	Yuan and others, 1990
<i>Fusarium</i> sp.	<i>A. auriculiformis</i>	Thailand	Pongpanich, 1997
	<i>A. auriculiformis</i>	Australia	Yuan and others, 1990
	<i>A. auriculiformis</i>	Australia	Yuan and others, 1997
<i>Hansfordia</i> sp.	<i>A. auriculiformis</i>	Thailand	Pongpanich, 1997
<i>Helminthosporium</i> sp.	<i>A. mearnsii</i>	Australia	Yuan and others, 1990
<i>Lasiodiplodia</i> sp.	<i>A. auriculiformis</i>	Thailand	Pongpanich, 1997
<i>Pestalotia</i> sp.	<i>A. mearnsii</i>	China	Liu, 1988
<i>Pestalotiopsis disseminata</i>	<i>A. auriculiformis</i>	Australia	Yuan and others, 1997
<i>Pestalotiopsis neglecta</i>	<i>A. auriculiformis</i>	Australia	Yuan and others, 1997
<i>Pestalotiopsis phoenicis</i>	<i>A. auriculiformis</i>	Australia	Yuan and others, 1997
<i>Pestalotiopsis</i> sp.	<i>A. auriculiformis</i>	Australia	Yuan and others, 1990
<i>Phoma</i> sp.	<i>A. auriculiformis</i>	India	Mathur, 1974
	<i>A. auriculiformis</i>	India	Mohanan and Sharma., 1988
	<i>A. confusa</i>	Philippines	Agmata, 1979
	<i>A. modesta</i>	India	Mittal and others, 1990
	<i>A. raddiana</i>	Israel	Mittal and others, 1990
	<i>Acacia</i> sp.	Egypt	Mittal and others, 1990
<i>Phomopsis</i> sp.	<i>A. auriculiformis</i>	Thailand	Pongpanich, 1997

Table 3

Pathogenic Fungi Associated with Tropical Casuarina Seeds

Fungus	Host	Country	Reference(s)
<i>Botryodiplodia theobromae</i>	<i>C. equisetifolia</i>	Philippines	Mittal and others, 1990
<i>Botryodiplodia</i> sp.	<i>C. equisetifolia</i>	Philippines	Mittal and others, 1990
<i>Curvularia brachyspora</i>	<i>C. equisetifolia</i>	Philippines	Mittal and others, 1990
<i>Curvularia lunata</i>	<i>C. equisetifolia</i>	Philippines	Mittal and others, 1990
	<i>C. equisetifolia</i>	Thailand	Mittal and others, 1990
<i>Curvularia pallescens</i>	<i>C. equisetifolia</i>	Philippines	Mittal and others, 1990
<i>Curvularia senegalensis</i>	<i>C. equisetifolia</i>	Australia	Yuan and others, 1990
<i>Fusarium moniliforme</i>	<i>C. equisetifolia</i>	Philippines	Mittal and others, 1990
<i>Fusarium semitectum</i>	<i>Casuarina</i> sp.	India	Sahai, 1982
<i>Macrophomina phaseolina</i>	<i>C. equisetifolia</i>	Philippines	Mittal and others, 1990
<i>Pestalotia</i> sp.	<i>C. equisetifolia</i>	Philippines	Mittal and others, 1990
<i>Pestalotiopsis</i> sp.	<i>C. equisetifolia</i>	Australia	Yuan and others, 1990
	<i>C. cunninghamiana</i>	Australia	Yuan and others, 1990
<i>Phoma</i> sp.	<i>C. equisetifolia</i>	Philippines	Mittal and others, 1990
	<i>C. cunninghamiana</i>	Australia	Yuan and others, 1990
<i>Phomopsis casuarinae</i>	<i>C. equisetifolia</i>	India/Australia	Bose, 1947

research on this pathogen to determine its taxonomic affinities, its pathogenic status, and the etiology of blister-bark disease. Until further information is available it would be imprudent to disregard the possibility that the disease may be seed borne. (Table 3)