Variability in Susceptibility of Some *Banksia* Species to *Phytophthora cinnamomi* and Their Distribution in Australia

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ABSTRACT

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The relative susceptibility of 1-mo-old seedlings of 29 *Banksia* species to *Phytophthora cinnamomi* was compared in two greenhouse pot studies. Eight species, *B. asplennifolia, B. collina, B. ericifolia, B. integrifolia, B. paludosa, B. robur, B. serrata*, and *B. serratifolia*, were resistant to *P. cinnamomi*. All eight species are native to the coastal regions of eastern Australia. Nine species, *B. ashybi, B. baxterii, B. candolleana, B. hookerana, B. laricina, B. lindeleyana, B. prionotes, B. sceptrum*, and *B. speciosa*, were highly susceptible to the fungus. In a separate experiment, no difference in susceptibility between cuttings and seedlings of the same species was detected.

Additional key words: Proteaceae, root rot

A relatively new and promising floricultural industry is developing in Hawaii using members of the Proteaceae family. Since its introduction nearly 10 yr ago, several plantings of these plants have been made in the Kula district of Maui and the Kohala and Kona districts of the island of Hawaii.

In the past 10 yr, *Phytophthora cinnamomi* Rands has been identified as the cause of severe root rots in Australia (6,7), South Africa (10), California (8), and Hawaii (2). In Hawaii, the genus *Banksia* is more severely affected than other genera of the Proteaceae. Banksias planted along drainage lines and other moist sites tended to be more severely affected, whereas those on drier sites generally survived longer. In a previous report (2), three major *Banksia* species planted in Hawaii were found susceptible to *P. cinnamomi*. This is especially disturbing because about 16% of Hawaii's Proteaceae plantings consist of the genus *Banksia*. Two *Banksia* species were determined resistant to *P. cinnamomi*. As cut flowers, however, they have little value. Graft compatibility between disease-susceptible and -resistant species thus far have met with limited success (*unpublished*). The current experiments were initiated to identify other banksias resistant to *P. cinnamomi*. The origin of *P. cinnamomi* is discussed in relationship to the distribution of susceptible *Banksia* species in Australia.

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MATERIALS AND METHODS

An isolate of *P. cinnamomi* obtained from a naturally infected plant of *B. speciosa* R. Br. was used in all pathogenicity studies. Inoculum produced using Hwang's modification (4) of the Chen-Zentmyer technique (1) consisted of 1×10^3 zoospores per pot. Inoculations were made at the soil surface adjacent to each seedling hypocotyl. Sterile distilled water was used for the untreated controls. Relative susceptibility was based on the percentage of diseased plants 3 mo after inoculation. Infection was confirmed by reisolation of the fungus from the crown and/or root tissues. Diseased seedlings were collected, washed free of potting media, surface-sterilized in 0.5% sodium hypochlorite for 10 min, rinsed in sterile distilled water, and placed on vegetable juice agar (20% Campbell's V-8 juice, 0.2% $CaCO_3$ and 20% agar) and incubated at 24 C under constant illumination.

Because of the difficulties in obtaining seeds of the various species used and variability in germination rates, two separate experiments were conducted to test all major species. In experiments 1 and 2, seedlings of 29 different *Banksia* species of Australian origin were compared for susceptibility to *P. cinnamomi*. *Banksia ashbyi* E. G. Bak. and *B. prionotes* Lindl., shown previously to be highly susceptible, and *B. serrata* L. F., shown previously to be highly resistant (2), were included as controls.

In a third experiment, seedlings of *B. collina* R. Br. and *B. integrifolia* L. F., representing resistant species, and *B. burdettii* I. G. Bak., representing a susceptible species, were compared with rooted cuttings of the same species with respect to their reaction to *P. cinnamomi*. Seeds were germinated in a pot

Table 1. Susceptibility of 18 *Banksia* species to *Phytophthora cinnamomi* evaluated 3 mo after inoculation (test 1)

Plant species	Days to 50% mortality[w]	Percent diseased[w]	
		Dead	Infected
Banksia ashbyi	8	90.0 g[x]	90.0 e
B. baueri	60	60.0 cd	70.0 cd
B. brownii	17	60.0 cd	63.3 c
B. candolleana	13	73.3 def	67.7 cd
B. collina	···	0.0 a	16.7 a
B. ericifolia	···	0.0 a	16.7 a
B. grandis	60	50.0 c	53.3 bc
B. integrifolia	···	0.0 a	16.7 a
B. hookerana	9	90.0 g	93.3 e
B. laricina[y]	8	76.2 e	80.9 de
B. marginata	···	26.7 b	43.3 b
B. media	···	20.0 b	56.7 b
B. prionotes	8	90.0 g	90.0 e
B. robur	···	0.0 a	0.0 a
B. sceptrum	17	63.3 cde	70.0 cd
B. serrata	···	0.0 a	0.0 a
B. serratifolia[z]	···	0.0 a	5.6 a
B. speciosa	7	86.7 fg	93.3 e

[w] Average of three replicates, each comprising 10 plants for each species.
[x] Numbers followed by the same letter are not significantly different ($P = 0.05$) according to Duncan's multiple range test.
[y] Only seven plants evaluated per replicate.
[z] Only six plants evaluated per replicate.

containing a peat moss and perlite mix and seedlings were transplanted upon production of the first true leaves. Cuttings were dipped in a indole butyric acid rooting hormone and placed individually in 1-in.-square cells of the transplanting tray (Speedling Inc., Sun City, FL 33586) containing peat moss and perlite. The cuttings were then placed in a propagation mist room for 2 mo before transplanting. All plants were transplanted into 2-in. pots that contained a medium-grade vermiculite. Transplanted seedlings were maintained for 1 mo and cuttings for 3 mo in a greenhouse before inoculations. Plants were fertilized twice, 1 and 2 mo after transplanting, with a foliar fertilizer (Gaviota Foliar 60, 7.4 ml/3.8 L) by drenching.

Each experiment included three replicates that contained six to 10 plants. All data were analyzed by analysis of variance and Duncan's multiple range test.

RESULTS

In experiments 1 and 2, susceptibility of 29 *Banksia* species to *P. cinnamomi* was assessed (Tables 1 and 2). Several species, *B. ashybi, B. baxterii* R. Br., *B. candolleana* Meisn., *B. hookerana* Meisn., *B. laricina* C. A. Gardn., *B. lindleyana* Meisn., *B. prionotes, B. sceptrum* Meisn., and *B. speciosa*, were rated highly susceptible to the fungus. Percentage mortality ranged from 60 to 100% after 3 mo, with 50% or greater mortality occurring within 23 days of inoculation. Eight species, *B. asplenifolia* Salib., *B. collina, B. ericifolia* L. F., *B. integrifolia, B. paludosa* R. Br., *B. robur* Cav., *B. serrata*, and *B. serratifolia* Salib., were considered highly resistant. None of the control seedlings died during the experiment.

In experiment 3, no differences in the percentage of dead or infected plants were observed between rooted cuttings and seedlings of the same species (Table 3). None of the resistant species, *B. collina* and *B. integrifolia*, died during this experiment compared with 40–46% mortality for *B. burdettii*.

P. cinnamomi was readily reisolated from the roots and crowns of plants that showed dark discoloration. The fungus was not reisolated from *B. robur* and *B. serrata* in experiment 1.

DISCUSSION

This study extends previous research in determining the susceptibility of species in the genus *Banksia* to *P. cinnamomi* (2). In view of the limited success achieved with interspecific graft compatibility thus far, it would be useful to select surviving seedlings from susceptible species for further testing with respect to susceptibility and horticultural desirability.

A comparison of resistant and susceptible species with respect to their distribution in their native habitat in Australia (3) is shown in Table 4. Species that were identified as resistant are distributed along the eastern and southeastern sections of Australia, whereas the susceptible species generally came from Western Australia.

P. cinnamomi is widely distributed in Australia and is commonly associated with root diseases in native plant communities along the eastern coast (7), southern Victoria (11), southern Queensland (5), and Western Australia (6). Pratt and Heather (7) suggested that *P. cinnamomi* is indigenous to eastern Australia. They provided as evidence the presence of the fungus as a common component of the soil microflora in both disturbed and undisturbed areas, a wide variation in fungal isolates with respect to growth rates, a wide distribution of both mating types, and development of resistant plant species in moist habitats. Although this hypothesis has been refuted by Routley and Routley (9), this study provides evidence that supports

Table 2. Susceptibility of 13 *Banksia* species to *Phytophthora cinnamomi* evaluated 3 mo after inoculation (test 2)

Plant species	Days to 50% mortality[w]	Percent Diseased[w]	
		Dead	Infected
Banksia ashbyi	18	70.0 bc[x]	50.0 cde
B. asplenifolia	···	0.0 f	3.3 g
B. baxterii	18	80.0 ab	83.3 ab
B. coccinea	23	60.0 bcd	70.0 bc
B. lindleyana	19	79.2 ab	87.5 ab
B. nutans	···	33.3 de	43.3 de
B. ornata	41	53.3 bcd	73.3 bc
B. paludosa	···	3.3 f	10.0 fg
B. prionotes	11	100.0 a	96.7 a
B. pulchella[y]	···	38.1 de	57.1 cd
B. serrata	···	16.7 ef	30.0 ef
B. sphaerocarpa	···	43.3 cde	50.0 cde
B. violacea[z]	32	54.2 bcd	70.8 bc

[w] Average of three replicates, each comprising 10 plants for each species.
[x] Numbers followed by the same letter are not significantly different ($P = 0.05$) according to Duncan's multiple range test.
[y] Only seven plants evaluated per replicate.
[z] Only eight plants evaluated per replicate.

Table 3. Susceptibility of three *Banksia* species seedlings and rooted cuttings to *Phytophthora cinnamomi* evaluated 3 mo after inoculation

Plant species	Plant material	Percent dead[y]	Percent infected[y]
Banksia burdettii	Seedling	40 b[z]	40 b
	Rooted cutting	46 b	46 b
B. collina	Seedling	0 a	0 a
	Rooted cutting	0 a	3.3 a
B. integrifolia	Seedling	0 a	0 a
	Rooted cutting	0 a	3.3 a

[y] Average of three replicates, each composed of 10 plants.
[z] Numbers followed by the same letter are not significantly different ($P = 0.01$) according to Duncan's multiple range test.

Table 4. Susceptibility[a] to *Phytophthora cinnamomi* of 33 *Banksia* species and their distribution[b] in Australia

Plant species	Distribution
Resistant	
B. asplenifolia	E. Australia
B. collina	E. Australia
B. ericifolia	E. Australia
B. integrifolia	E. Australia
B. paludosa	E. Australia
B. robur	E. Australia
B. serrata	E. Australia
B. serratifolia	E. Australia
Moderately susceptible	
B. baueri	W. Australia
B. brownii	W. Australia
B. caleyi	W. Australia
B. coccinea	W. Australia
B. grandis	W. Australia
B. marginata	S. E. Australia
B. media	W. Australia
B. nutans	W. Australia
B. ornata	S. E. Australia
B. pulchella	W. Australia
B. sceptrum	W. Australia
B. sphaerocarpa	W. Australia
B. violacea	W. Australia
Highly susceptible	
B. ashbyi	W. Australia
B. baxterii	W. Australia
B. burdettii	W. Australia
B. candolleana	W. Australia
B. hookerana	W. Australia
B. laricina	W. Australia
B. lindleyana	W. Australia
B. menziesii	W. Australia
B. occidentalis	W. Australia
B. occidentalis	W. Australia
B. prionotes	W. Australia
B. speciosa	W. Australia

[a] Determined in this study and from Cho (2).
[b] Determined from Holliday and Watton (3).

Pratt and Heather's claim. *Banksia* species identified as resistant are commonly found growing along the northeastern coast of Australia (3). Furthermore, similar to the evidence presented by Pratt and Heather, our data show a decrease in resistance of species distributed in southeastern Australia (ie, *B. marginata* and *B. ornata*). Pratt and Heather (7) recovered *P. cinnamomi* from two *Banksia* species, *B. integrifolia* and *B. serrata*, in eastern Australia that showed symptoms of the disease. They noted the difficulty, however, in distinguishing damage caused by *P. cinnamomi* from other causes. Because these species have been identified as resistant in this study, it is possible that other causes were involved.

Pratt and Heather (7) suggested that *P. cinnamomi* is a recent introduction in Western Australia. As evidence, they noted the large number of susceptible plant species and the widespread dieback caused by *P. cinnamomi* in the *Eucalyptus marginata* Sm. forest stands from that region. Our research also shows that *Banksia* species native in Western Australia were highly susceptible to *P. cinnamomi*.

In our research, no differences were observed between rooted plant material and 1-mo-old seedlings of the same species with respect to *P. cinnamomi* susceptibility.

It is disappointing to find that, in general, the *Banksia* species with spectacular inflorescences belong to the moderately susceptible or highly susceptible group of plants. Therefore, disease management for the grower will have to rely on proper cultural management practices and the development of chemical controls and compatible resistant rootstock materials.

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