ROOT ROT OF RANUNCULUS ASIATICUS CAUSED BY PYTHIUM DEBARYANUM

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INTRODUCTION

A destructive root disease of Persian buttercup (Ranunculus asiaticus L.) was first observed in commercial field plantings at Inglewood and Pacific Palisades, Calif., during the winter of 1937–38. Subsequently the disease has also been found in Santa Cruz, San Mateo, and San Francisco Counties. The losses are reflected not only in decreased field stands of plants, but in a smaller cut-flower crop and yield of tubers. Because of its economic importance, an investigation of this disease was conducted during the past four seasons, the results of which are briefly recorded in this paper. A preliminary note has previously been published.

SYMPTOMS OF THE DISEASE

Ranunculus plants, whether produced from seeds or tubers, appear to be highly susceptible to root rot infection when grown in the field. The most striking symptoms of the disease consist of a general wilting followed by rapid collapse and death of the plant. The roots, tubers, stems, and petioles may be invaded. The roots and tubers of infected plants are dark brown, water-soaked, and flaccid. The stem plate is discolored and dark, but usually not water-soaked or flaccid. Occasionally the petioles become infected, in which event the diseased parts are dark brown, frequently exhibiting necrotic streaks which are 1 to 3 mm. wide and about 30 to 60 mm. long, extending outward from the base of the petiole and parallel to its long axis. In the field, plants are most commonly affected when 6 to 8 inches high, but, under experimental conditions, plants of all ages appear to be highly susceptible. The disease occurs on widely different soil types, ranging from coarse sand to heavy clay, and is favored by excessive rainfall or irrigation, poor drainage, cool weather, and crowding of plants.

THE CAUSAL FUNGUS, PYTHIUM DEBARYANUM

Isolations made from diseased material on water or malt-extract agars have consistently yielded a fungus which has been identified as Pythium debaryanum Hesse. The four isolates of the fungus studied, representing two districts in southern and two in central California where the disease occurs, are readily cultivated on potato-

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dextrose, malt, corn-meal, and other agars, on which they develop a
copious, dense, aerial mycelium. The asexual and sexual reproduc-
tive organs are sparingly produced in the surface growth but develop
rather profusely intramatrically. Both types of fruiting bodies are
abundant in the diseased tissues of tubers of infected plants. Per-
haps the best medium for observing the development of the repro-
ductive structures is plain water agar. On this substrate, the
sporangia are usually spherical, thin and smooth-walled, and aerog-
genous; numerous intercalary sporangia are produced which may be
spherical or barrel-shaped. Sporangia germinate either by the produc-
tion of zoospores in a vesicle or by several germ tubes. Oogonia are
similar to sporangia and have an average diameter of 22.2µ. An-
theridia are usually plural, varying in number from one to six,
predominantly two to three per oogonium, monoclinous and diclinous,
when monoclinous arising some distance below the oogonium and
not adjacent to it. One fertilization tube is usually produced by
each antheridium applied to the oogonial wall. The oospores are
spherical and smooth-walled, with a mean diameter of 18.2µ. Ger-
mination of oospores has not been observed.

The temperature-growth relations of the fungus have been deter-
mined for two of the four isolates. They are: Minimum, 1° C.,
optimum, 28°, and maximum 37°.

All four isolates of the fungus proved pathogenic to healthy
ranunculus plants grown from seeds and tubers. Inoculum was
prepared by growing the fungus on sterilized cracked wheat in 8-inch
test tubes. When ready for use, this was added to autoclaved soil
contained in 6-inch pots in such a manner as to avoid injuring the
root system of the young plants. Sterile cracked wheat was added
to the pots serving as controls. All pots were heavily watered each
day in order to keep the soil very moist. The incubation period

![Figure 1.—Root rot of Persian buttercup: A, Symptoms produced on young Persian buttercup plant by Pythium debaryanum 12 days after inoculation in the greenhouse; B, noninoculated control.](image-url)
ranged from 11 to 25 days, but all infected plants died within a day or two after the foliage commenced to wilt (fig. 1). Of 20 plants inoculated with each isolate, none escaped infection, while the 20 control plants remained healthy. The fungus was reisolated from each infected plant and proved to be identical with the original isolates. When tested in parallel series, the reisolates again proved highly pathogenic. In these tests, the infected plants exhibited symptoms identical with those of naturally infected plants. The appearance of the root system of artificially infected plants, as contrasted with healthy roots, is shown in figure 2.

Although *Pythium ultimum* Trow and *P. irregulare* Buis, are commonly associated with root rots of various herbaceous ornamentals in California, they have not been found associated with this disease. Inoculation experiments have shown that *P. ultimum* is strongly pathogenic to *Ranunculus* while *P. irregulare* is weakly pathogenic but likewise capable of inducing disease. The symptoms produced by these two species are similar to those produced by *P. debaryanum*.

**EXPERIMENTAL HOST RANGE**

In studies on the host range, one isolate of the fungus from southern California was used along with the no-wound technique described for the pathogenicity tests. Healthy plants to be inoculated had four to six leaves. Reisolations were made from all experimentally infected plants, and the reisolate of the fungus from a particular host was then tested by inoculation into healthy specimens of that host.
Since it is generally believed that *Pythium deharyanum* is cosmopolitan in its attacks, a large variety of plants including many of those grown in rotation with *Ranunculus* were experimentally inoculated. The experimental host range proved to be very limited and includes Iceland poppy (*Papaver nudicaule* L.), columbine (*Aquilegia caerulea* James), fibrous-rooted begonia (*Begonia semperflorens* Link and Otto), butterflyflower (*Schizanthus pinnatus* Ruiz and Pav.), and cucumber (*Cucumis sativus* L.). If seeds had been sown in experimentally infested soil the host range might have been extended. If this technique were employed the results would be indicative of the ability of *P. deharyanum* to cause damping-off and would not indicate its potentialities as a root rotting agent of more mature plants. Infection of these hosts, characterized by sudden wilting without change in color of the foliage and water-soaked lesions on the fibrous roots, occurred in 11, 18, 27, 9, and 17 days, respectively.

No infection was obtained in 41 species of plants representing 37 genera in 23 families, as follows:

### Aizoaceae:  
*Iceplant* (*Mesembryanthemum crystallinum* L.)

### Amaranthaceae:  
*Cockscomb* (*Celosia argentea* L. var. *cristata* Kuntze)

### Campanulaceae:  
*Canterbury-bells* (*Campanula medium* L.)

### Caryophyllaceae:  
*Sweet-william* (*Dianthus barbatus* L.)
*Carnation* (*Dianthus caryophyllus* L.)

### Chenopodiaceae:  
*Spinach* (*Spinacia oleracea* L.) var. Bloomsdale

### Compositae:  
*Transvaal daisy* (*Gerbera jamesonii* Hook. var. *transvaalensis* Hort.)
*China-aster* (*Callistephus chinensis* Nees) var. Giant Branching White, wilt resistant
*French marigold* (*Tagetes patula* L.)
*African marigold* (*Tagetes erecta* L.)
*Gaillardia pulchella* Fouq. var. *picta* Gray
*Hybrid cineraria* (*Senecio cruentus* DC.)

### Cruciferae:  
*Cabbage* (*Brassica oleracea* L. var. *capitata* L.) var. Winter Colma
*Cauliflower* (*Brassica oleracea* var. *botrytis* L.) var. February
*Radish* (*Raphanus sativus* L.) var. White Icicle
*Dames violet* (*Hesperis matronalis* L.)
*Annual stock* (*Malthiola incana* R. Br. var. *annua* Voss) var. Fiery
*Blood Red*

### Dipsacaceae:  
*Mourning bride or pine cushion flower* (*Scabiosa atropurpurea* L.)

### Labiatae:  
*Flowering sage* (*Salvia farinacea* Benth.)

### Leguminosae:  
*Broadbean* (*Vicia faba* L.)

### Malvaceae:  
*Hollyhock* (*Althaea rosea* Cav.)

### Onagraceae:  
*Godetia grandiflora* Lindl.

### Papaveraceae:  
*California poppy* (*Eschscholtzia californica* Cham.)
*Oriental poppy* (*Papaver orientale* L.)

### Polemoniaceae:  
*Phlox drummondii* Hook.

### Ranunculaceae:  
*Poppy anemone* (*Anemone coronaria* L.)
*Love-in-a-mist* (*Nigella damascena* L.)
*Rocket larkspur* (*Delphinium ajacis* L.)
DISCUSSION

In California, Persian buttercup plants are grown commercially in the field from seeds and tubers for seed, tuber, and cut-flower production. When grown for cut flowers, growers generally prefer to plant seeds, which are usually drilled in rows in August. The young plants, which commence to flower early in January if not infected, are frequently invaded by the fungus in the December preceding, or soon after the winter rains commence. When grown on poorly drained land, the mortality may reach 50 percent or more. Since limited tests of various strains of seeds and tubers from different sources failed to disclose any evidence of resistance to the disease, control is dependent upon the selection of well-drained sites.

SUMMARY

A destructive root disease of Persian buttercup (Ranunculus asiaticus) is described.

The disease is prevalent in commercial plantings in four coastal counties of California. It is favored in its development and spread by excessive moisture, poor drainage, cool weather, and crowding of plants.

The chief symptoms of the disease consist of a general wilting followed by rapid collapse and death of the plant. The roots, tubers, stems, and petioles may be affected.

Pythium debaryanum has been consistently isolated from infected plants and has proved pathogenic in greenhouse tests. The morphology and temperature relations of the fungus are discussed.

The incubation period for the disease ranges from 11 to 25 days.

In greenhouse tests, the fungus proved pathogenic to young plants of Iceland poppy, columbine, fibrous-rooted begonia, butterflyflower, and cucumber.

Persian buttercup seedlings were also experimentally infected with Pythium ultimum and P. irregularare. The symptoms were similar to those produced by P. debaryanum.