

flowers opened. The yellow pollen from *A. schottii* was found on the stigmas of the saguaro flowers, among the cream-colored pollen from the saguaro, indicating that flowers of both species were visited. There were no claw marks or other signs of damage to the saguaro flowers by the bats. Details of their activity in the cage are described elsewhere (5).

Honey bees (*Apis mellifera* L.) have been in the Southwest only since 1872 (6), which is not long enough to account for many of the more mature saguaros. However, the bees are attracted to saguaro flowers (7) and were included in this test because of their versatility as pollinators.

The bees were housed in a single-body Langstroth hive, well stocked with bees of all stages, honey, and pollen. Water was constantly available. Bee activity in the cage was similar to that in the open (7).

Appropriate checks of hand cross-pollinated and of naturally self-pollinated flowers were made intermittently. Such flowers were covered with cheesecloth if pollinating agents were in the cage; otherwise the flowers were left open. For comparison, records were also kept on some naturally pollinated "field" blossoms.

Each day throughout the flowering period all caged flowers were tagged and the fruits that were shed were recorded. As fruits matured they were harvested and the seeds were removed and counted. Germination percentages were determined under conditions of natural light and temperature in the laboratory (8).

The results (Table 1) show that viable seeds were produced in considerable quantity by both night- and day-pollinating agents. The effectiveness of the agents tested and the probable effectiveness of similar pollinators in the area (7) would indicate that failure of the saguaro to repopulate is not due to lack of cross-pollination.

To our knowledge this is the first experimental proof (9) that honey bees can pollinate the saguaro or that white-winged doves and *Leptonycteris* bats can pollinate any plant (10).

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3. Appreciation is expressed to John C. Cook, superintendent, Saguaro National Monument, for permission to collect saguaro arms and conduct this experiment on the Monument grounds; to Gilbert Ray, executive director, Pima County Parks and Recreation Department, for permission to collect saguaro arms in Tucson Mountain Park; to the University of Arizona Arid Lands Program for incidental funds; to Lyle K. Sows and E. Lendell Cockrum for cooperation in collecting the white-winged doves and bats, respectively.
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9. This report is Arizona Agricultural Experiment Station technical paper No. 616.
10. A report giving greater details of these tests is in preparation.

3 January 1961

Chemotaxis of Zoospores for Root Exudates

Abstract. A chemotactic response of the zoospores of a soil-inhabiting plant pathogenic fungus, *Phytophthora cinnamomi*, for roots of avocado seedlings was observed. The chemotaxis of the zoospores and chemotropy of their germ tubes were directly related to infection and disease production. Indications were obtained of specificity of the pathogen-attracting root exudate, and interesting implications are evident with regard to mechanisms of invasion and pathogenicity, and to disease resistance.

Plant pathogenic fungi invade their hosts by means of several different avenues including natural openings in host tissue, wounds, and direct penetration of living tissue (1). Invasion through such avenues has been attributed to (i) attraction of the fungus mycelium or the germinating spore to stimulatory chemicals exuding from roots or leaves, (ii) attraction of the fungus to a nutrient gradient, or (iii) mere chance.

The complex medium of soil provides a difficult but highly interesting area in which to study attraction of plant roots for plant pathogenic microorganisms. The motile infective zoospores of pathogenic species of the fungus genus *Phytophthora* serve as an excellent tool for investigating such phenomena. Other investigators have demonstrated chemotaxis of motile plant units such as bracken spermatozoids and gametes of *Allomyces* (2) to specific chemicals, electric fields, or hormones. Goode (3) reported a nonspecific attraction of zoospores of *Phytophthora fragariae* to the root tips and the root-hair zones of strawberry roots. Flentje (4) has recently summarized information on means by which plant pathogenic fungi

reach their host, and on subsequent invasion of the host.

In the present investigation young, vigorously growing roots of avocado (*Persea americana* Mill.) seedlings were found to have strong attraction for the zoospores of the avocado root pathogen, *Phytophthora cinnamomi* Rands. A preliminary report has been published (5). Young excised root tips (1 to 2 cm long, 1 to 2 mm in diameter) from avocado varieties highly susceptible to the pathogen were placed in petri dishes containing actively swimming zoospores of *P. cinnamomi*. Zoospores were liberated from sporangia produced at 24°C for these tests by a method previously described (6), involving use of a nonsterile soil extract.

Chemotaxis of the zoospores for the roots occurred within a few minutes and was demonstrated by an obvious accumulation of the motile spores in the immediate vicinity of the root pieces. The concentration of spores was notably greater in the region of elongation on the root, just above the root tip, than it was at the root tip or in the region of differentiation. The avocado root does not produce root hairs.

In 30 to 60 minutes, as a rule, spores settling on and in the vicinity of the roots began to encyst and to germinate. Quantitative evidence of the attraction was then obtained by examining the root pieces under a dissecting microscope and counting numbers of spores settling on different areas of the root and adjacent to the root. These observations showed:

1) That the zoospores were particularly attracted to the region of elongation above the root tip, and that spores also encysted at different distances from the root as if in response to a concentration gradient of some stimulatory chemical exuding from the root (Table 1).

2) Positive chemotropism of germ

Table 1. Attraction of zoospores of *Phytophthora cinnamomi* to roots of a host plant (avocado) and a nonhost plant (citrus).

Distance from root (mm)	Av. No.* of zoospores settling in areas 0.5-mm square
<i>Avocado</i>	
0-0.5	34.0
0.5-1.0	14.7
1.0-1.5	11.1
1.5-2.0	8.7
2.0-2.5	5.0
2.5-3.0	4.2
<i>Citrus (mandarin orange)</i>	
0-0.5	0.6
0.5-1.0	1.4
1.0-1.5	0.9
1.5-2.0	1.3
2.0-2.5	0.9
2.5-3.0	1.3

* Figures represent mean of ten fields counted.

tubes of germinating zoospores for the avocado roots was evident in the case of spores settling on the bottom of the petri dish at distances of up to 2 to 3 mm from the root. The germ tubes were uniformly directed toward the avocado root pieces.

3) Shortly after spore germination occurred, invasion of the root took place through unwounded tissue, and within 24 hours a brown lesion was visible in the region of elongation, identical in appearance to lesions observed in the case of infection of intact plants.

4) Evidence was obtained that the attractive substance is specific to the susceptible living avocado root, since roots killed by boiling or by propylene oxide did not attract zoospores. No chemotaxy of zoospores occurred toward actively growing roots of several other types of plants (tomato, tobacco, mandarin orange) (Table 1). There was also evidence of decreased attraction in the case of avocado varieties with some resistance to *Phytophthora cinnamomi*. Some roots of other plants (macadamia nut, sweet orange, pea) exhibited attraction for the zoospores, but this was primarily to root tips and cut ends of roots rather than to the region of elongation. As further evidence of specificity, zoospores of *Phytophthora citrophthora*, a citrus pathogen, were not attracted to avocado roots, but were attracted to citrus roots (7).

5) Zoospores of *P. cinnamomi* and their germ tubes showed chemotactic and chemotropic activity for aqueous extracts of susceptible avocado roots taken up on filter-paper disks. The nature of the substance is under investigation.

These results obviously have interesting implications with respect to resistance and susceptibility of plant roots to pathogens, as well as to various basic aspects of mechanisms of invasions and pathogenicity (8).

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7. The culture of *P. citrophthora* was provided by L. J. Klotz of this department.
8. This report is paper No. 1246 from the Citrus Experiment Station, University of California, Riverside.

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Patterns of Corticosteroid and Pepsinogen Change Related to Emotional Stress in the Monkey

Abstract. In association with conditioned avoidance sessions of 72 hours' duration, monkeys showed a response pattern characterized by increased levels of 17-hydroxycorticosteroids and decreased levels of pepsinogen during the stress period, with a marked and prolonged elevation of pepsinogen levels occurring during the recovery period.

Previous reports from this laboratory have described long-term studies of the effects of behavioral conditioning procedures upon the pituitary-adrenal cortical system in the monkey (1, 2). A major objective in the extension of this work has been the measurement of additional endocrine or visceral functions so that centrally integrated patterns of visceral activity might be brought under investigation. In recent

studies concerned primarily with the effects upon adrenal cortical activity of repeated conditioned emotional stress in the monkey, a high incidence of gastric or duodenal ulceration was observed (3). These observations prompted us to include, along with determinations of pituitary-adrenal cortical activity, the measurement of pepsinogen levels as a means of indirectly evaluating gastric function in animals under stress. The present report, then, describes a preliminary effort that was carried on to compare plasma 17-hydroxycorticosteroid (17-OH-CS) and pepsinogen responses during and following periods of sustained emotional stress associated with avoidance behavior in the monkey.

Four adult rhesus monkeys, two of each sex, were placed in an experimental chair-type restraining apparatus and allowed several days for adaptation according to previous studies (4).

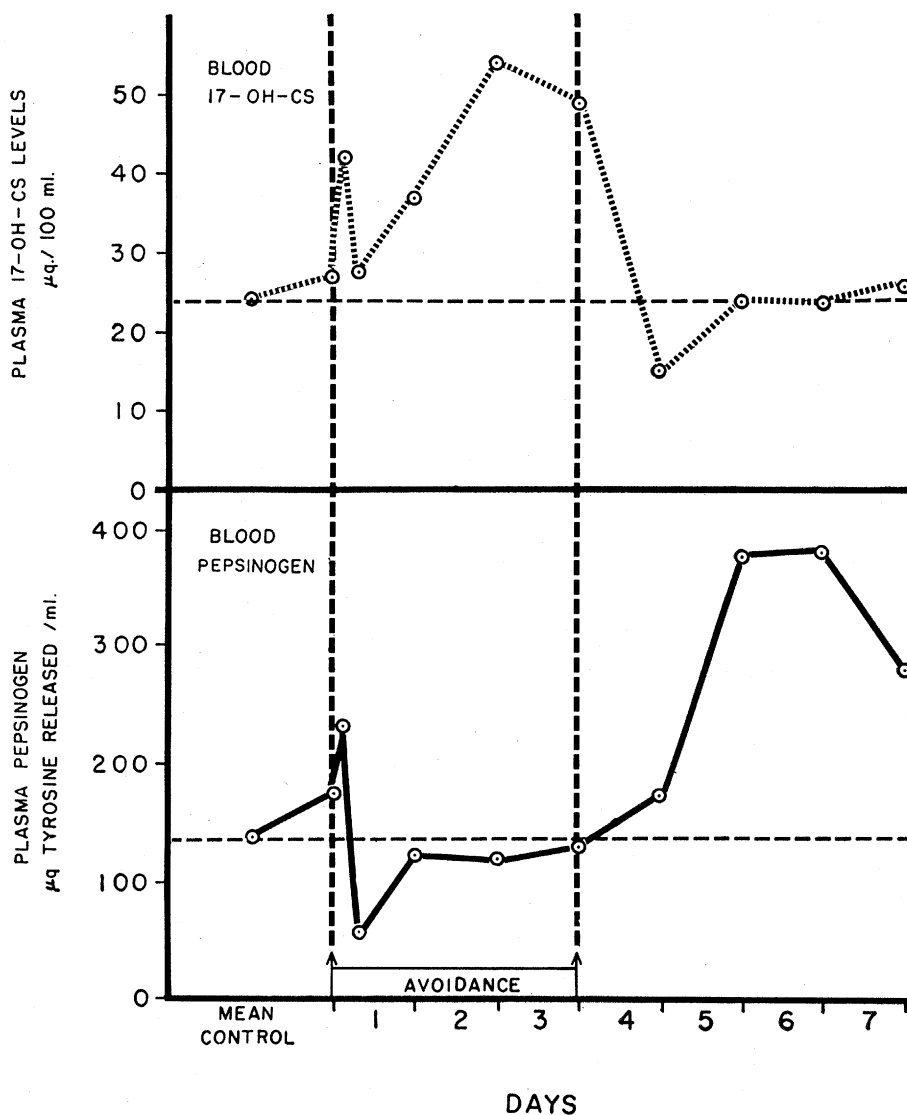


Fig. 1. Mean blood levels of 17-OH-CS and pepsinogen during 72-hour continuous avoidance sessions.