Influence of the Matric and Osmotic Components of Water Potential on Zoospore Discharge in Phytophthora

J. D. MacDonald and J. M. Duniway

Assistant and Associate Professors, respectively, Department of Plant Pathology, University of California, Davis, CA 95616.

Portion of a thesis submitted by the senior author in partial fulfillment of the requirements for the Ph.D. degree, University of California, Davis.

Supported in part by National Science Foundation Grant BMS75-02607.

Accepted for publication 24 October 1977.

ABSTRACT

MACDONALD, J. D., and J. M. DUNIWAY. 1978. Influence of the matric and osmotic components of water potential on zoospore discharge in Phytophthora. Phytopathology 68: 751-757.

Mycelial disks from agar plate cultures of *Phytophthora cryptogea* and *P. megasperma* incubated in soil at −150 millibars (mb) matric potential (ψ_m) on tension plates formed abundant sporangia within 3-4 days. The effect of ψ_m on zoospore discharge was then determined by changing ψ_m from −150 mb, where sporangia failed to release zoospores, to 0, −1, −5, −10, or −25 mb ψ_m. Sporangia typically discharged large numbers of zoospores within 60 to 90 min in completely saturated soil (ψ_m = 0) and at −1 mb ψ_m. Discharge was impaired at −5 mb, greatly restricted at −10 mb, and fully prevented at −25 mb ψ_m. Similar results were obtained with sporangia formed at −50, −150, or −300 mb. Use of different textured fractions of soil revealed that discharge is governed more by ψ_m than by soil water content. Shifts in temperature between 16 and 24 C failed to induce zoospore discharge at limiting ψ_m values. The influence of osmotica on zoospore discharge was evaluated by removing mycelial disks bearing sporangia from soil at $\psi_m = -150$ mb and placing them in solutions of known solute potential (ψ_s). Zoospores were discharged in solutions of KCl and $MgSO_4$ at $\psi_s > -4.5$ bars and in solutions of sucrose, NaCl, sea salts, and polyethylene glycol (PEG) 300 with ψ_s values as low as −6 to −9 bars. Discharge in PEG 6000 occurred only in solutions of $\psi_s > -1.3$ bars. These results indicate that zoospore discharge is extremely sensitive to the ψ_m component relative to the ψ_s component of soil water potential. Possible mechanisms of zoospore discharge are discussed.

Although zoospores of soilborne *Phytophthora* spp. are considered to be important in dissemination and infection (13, 17, 18, 19), the factors influencing the release of zoospores from sporangia in soil are not well defined. Zoospore release usually is induced in the laboratory by subjecting sporangia in aqueous systems to rapid temperature shifts (31). However, rapid temperature changes of the magnitudes usually employed are not likely to occur in a natural soil environment. Because of the aquatic affinities of this genus and the well known effects of wetness on sporulation by its aerial members, soil water has long been suspected to greatly influence sporangium formation and zoospore release by the soilborne species of *Phytophthora*. Indeed, such an influence recently has been demonstrated for sporangium formation by several *Phytophthora* spp. in soil (6, 7, 20, 21, 23, 27). However, despite its epidemiological significance, there is still little quantitative information on the levels of moisture conducive to zoospore release from sporangia in soil.

Most of the studies that have examined the influence of water status on zoospore release have employed solutes to vary water potential (ψ). For example, Katsura (16) reported that sporangia of *P. capsici* released zoospores when placed in sucrose solutions with solute potentials (ψ_s) as low as −8 bars. At lower ψ_s values sporangia germinated directly. Working with *Aphanomyces euteiches*, Hoch and Mitchell (15) determined that spore formation, extrusion, and the final release of motile spores, all had very exacting ψ_s requirements. Unfortunately, results obtained by manipulating ψ_s may not apply directly to the soil environment where changes in ψ are not correlated as much to changes in ψ_s as to changes in matric potential (ψ_m). Furthermore, several investigations suggest that zoospore release may be more sensitive to changes in ψ_m than the reported ψ_s limitations would indicate. Hoch and Mitchell (14) presented limited evidence that zoospore production in *A. euteiches* required a ψ_m value higher than −0.01 bar, as compared with the −2.0 bars ψ_s which permitted normal spore release. Working with *P. megasperma*, Pfender et al. (20) found that 95% of the sporangia in flooded soil ($\psi_m = 0$) germinated indirectly after 3 days, whereas less than 10% did so after 12 days at −0.05 bar ψ_m, and none did so at −0.1 or −0.6 bar ψ_m. Duniway (8) counted empty sporangia of *P. cryptogea* before and after adjusting the soil ψ_m from −0.25 bar to higher values, and found that the percentage of sporangia that released zoospores was decreased significantly when the ψ_m was decreased only slightly from zero.

In the present study, the influence of water potential on zoospore release by two *Phytophthora* spp. is examined quantitatively and, although emphasis is given to the influence of soil ψ_m, the separate effects of the ψ_m and ψ_s components of ψ on zoospore discharge are compared and contrasted.

00032-949X/78/000 127$03.00/0

MATERIALS AND METHODS

Two species of *Phytophthora* were studied; an isolate of *P. cryptogea* Pethyb. and Laff. described previously (6, 7, 8), and an isolate of *P. megasperma* Drechs. pathogenic to alfalfa, that was obtained from S. M. Mircetich, USDA-ARS, Department of Plant Pathology, University of California, Davis, CA 95616. Both isolates were grown on pea-dextrose agar prepared by soaking 100 g of dried peas overnight in 800 ml of distilled water and then bringing them to a boil for 10 min. The boiled peas were comminuted in a Waring Blendor and poured into a flask with 10 g dextrose and 17 g Difco agar. The mixture was adjusted to 1 liter, autoclaved, and dispensed into petri plates. Seven- to 10-day-old cultures were used in all experiments.

Sporangia were produced by lifting disks of aerial mycelium cut from culture plates with a 7-mm-diameter cork borer and incubating the disks 3-4 days in soil maintained at −150 millibars (mb, 1,000 mb = 1 bar) on tension plates as described by Duniway (6, 8). To assure rapid equilibration of the soil when ψ_m was manipulated, the volume and depth of soil on tension plates were kept to a minimum. Soil was placed to a uniform depth of 2 mm on the porous glass plates of the Büchner funnels that served as tension plates. Mycelial disks then were laid directly on the soil surface and each disk was covered individually with 2 mm of soil. Nine disks were spaced evenly in each funnel. To minimize differences in equilibration times, tension plates were selected for uniformity of water flow through their porous glass plates. The bottom of the soil in the tension plates was the reference point for measuring the height of the water column controlling ψ_m.

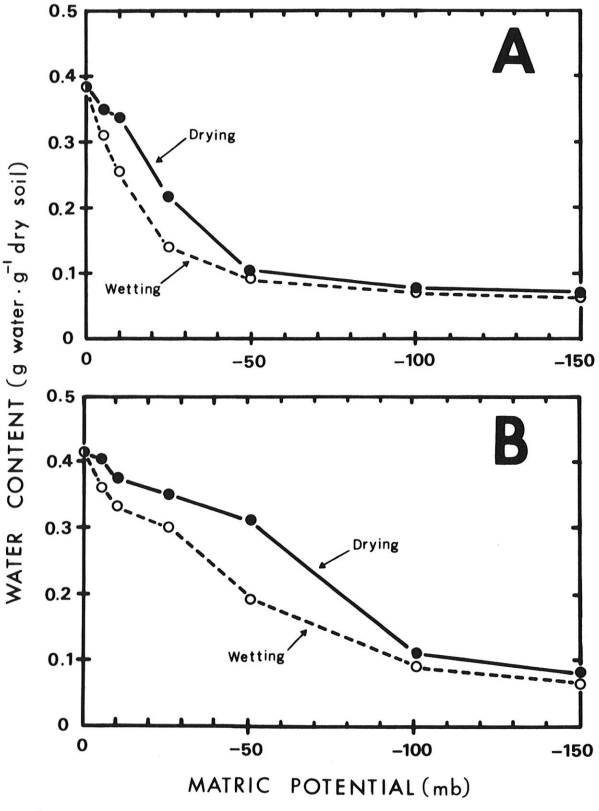

Fig. 1-(A, B). Relationship between water content and matric potential in the **A)** coarse (>250 μm) sand fraction and **B)** medium-fine (60-250 μm) sand fraction of Yolo fine sandy loam undergoing drying and wetting on tension plates.

The soil used in all experiments was a Yolo fine sandy loam (YFSL). The soil was wet-sieved after autoclaving to obtain coarse (> 250 μm) and medium-fine (60-250 μm) sand fractions. This was done because sporangium production in both the coarse and medium-fine sand fractions was superior to that in nonfractioned YFSL. Sieving also provided media derived from the same parent material with different water content-ψ_m relationships (Fig. 1).

Matric potential effects.—The influence of ψ_m on zoospore discharge was determined by adjusting the height of the water column in the tension plate apparatus to increase ψ_m from the −150 mb at which sporangia had formed to 0, −1, −5, −10, or −25 mb. Unless stated otherwise, the soil was wetted to the final ψ_m by water flow upwards through the porous glass plate rather than by adding water to the soil surface. This assured that ψ_m never exceeded the desired value. When temperature was not a variable, it was maintained constant at 22-24 C.

Zoospore release was observed by two methods. At 1-, 2-, 3-, 4-, and 6-hr intervals after the tension plates were reset to higher ψ_m values, three sporangia-bearing mycelial disks and the small volume of soil in which they were buried were removed from the funnels with a spatula. Each disk and its adjacent soil was suspended in 10 ml of distilled water, and the number of motile zoospores present in the soil sample was estimated by thoroughly mixing the suspension and placing an aliquot onto a counting grid mounted on a microscope slide. A glass coverslip was placed over the counting grid which consisted of a 15 × 10-mm piece of plastic window screen. The grid supported the weight of the coverslip so that the motile spores were not subjected to mechanical stresses which would cause them to encyst quickly (13). Each square of the counting grid filled a single field of view at ×100 magnification and enclosed a volume of 0.5 μliter. The number of motile spores in 20 squares was counted in each sample. Counts of zoospores under these conditions were done as rapidly as possible to assure that the only spores counted were those released by sporangia in the soil prior to sampling. The total number of zoospores present (motile and/or encysted) in a sample was estimated by spotting 50 μliter of the 10-ml soil suspension onto the surface of a selective antibiotic agar medium (7), and incubating the plates at 22-24 C for 18-24 hr. The adhering soil then was washed from the agar surface with a stream of distilled water, and one drop of acid fuchsin was added to each spot to fix and stain the germinating spores. The total number of zoospores in the 50 μliter spot was counted at ×100 magnification using bright-field microscopy.

Solute potential effects.—Discharge of zoospores from sporangia in osmotica was determined by removing mycelial disks from soil after sporangia had formed at −150 mb ψ_m, and placing them in solutions of various ψ_s which had been dispensed into partitioned petri plates.

Three disks were placed in each solution. The solutions were examined under ×40 magnification for swimming zoospores at intervals of 1, 2, and 4 hr. The relative numbers of zoospores discharged were rated on an arbitrary scale of 0-4 in which 0 represents no release and 4 represents the maximum release, such as was observed in water. The ψ_s values of sucrose, NaCl, and KCl solutions were obtained from published tables. The ψ_s values of solutions of $MgSO_4$, polyethylene glycol (PEG) 6000, PEG 300, and synthetic sea salts (Instant Ocean®, Aquarium Systems, Inc., Eastlake, OH 44094) were determined with an isopiestic psychrometer.

RESULTS

Matric potential effects.—Because there were differences between species and among experiments in the number of sporangia formed by mycelial disks in soil, the numbers of zoospores released at various ψ_m values were always compared with the numbers released in completely saturated soil ($\psi_m = 0$). To promptly saturate the soil, distilled water was added to the soil surface at the time tension plates were lowered to bring ψ_m to 0. Zoospores were first observed 1 hr after the soil was saturated. Release by both *P. cryptogea* (Fig. 2) and *P. megasperma* (data not shown) then proceeded rapidly over the next 3 hr and was essentially complete within 4 hr. Mycelial disks were removed from the soil before wetting and 4 hr after saturation and sporangium counts were made using the method described by Duniway (6) with the exception that fluorescent brighteners were not employed. These counts showed that 78-99% of the initially-full sporangia had emptied after 4 hr at $\psi_m = 0$. Zoospore release in completely saturated soil was considered optimal, and the numbers of zoospores released in all other treatments are expressed as percentages of the numbers released at saturation.

Zoospore release by both *P. megasperma* and *P. cryptogea* in soils allowed to rewet for 6 hr through tension plates to $\psi_m = 0$ or -1 mb also was found to be optimal (Fig. 3). However, in soil wetted to $\psi_m = -5$ mb, release was only 33 and 36% of the optimum for *P. megasperma* and *P. cryptogea*, respectively. At $\psi_m = -10$ mb, release was reduced to 8-10% of the optimum and there was no release of zoospores by either species at $\psi_m = -25$ mb.

There was a slight delay in the initiation of release in soils allowed to wet through the tension plates due to the time required for the soils to fully equilibrate. However, the 1-2 hr required for complete equilibration of the soil ψ_m had no apparent effect on the ultimate ability of sporangia to release zoospores within the 6-hr period. When additional observations of zoospores at intervals of 8, 12, and 24 hr were included in the -10 and -25 mb treatments, there was no change in the results (Fig. 3); i.e., release was severely impaired at -10 mb ψ_m, and completely prevented at -25 mb ψ_m. Mycelial disks were removed from tension plates after 24 hr at -25 mb ψ_m, and sporangium counts were made. There was no significant decrease in the number of full sporangia, or increase in the number of empty sporangia, as compared with counts of sporangia at -150 mb made just before wetting the soil. However, a limiting ψ_m value of -25 mb did not impair subsequent zoospore release because release proceeded in an optimum fashion when tension plates were set to $\psi_m = 0$ after 24 hr at -25 mb.

The coarse and medium-fine sand fractions of YFSL (Fig. 1), were used in additional experiments to determine the relative influence of soil ψ_m and soil water content on zoospore discharge by *P. cryptogea*. Zoospore release, as monitored by counts of motile spores, was nearly identical in the coarse and medium-fine sand fractions of

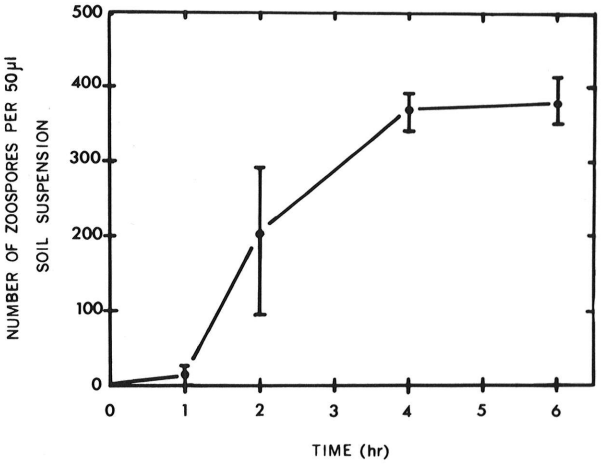

Fig. 2. Numbers of zoospores of *Phytophthora cryptogea* released in the coarse (>250 µm) sand fraction of Yolo fine sandy loam after saturation with water for various periods of time. Numbers of zoospores were determined by counting germinated spores on a selective medium and the vertical bars indicate the total variation.

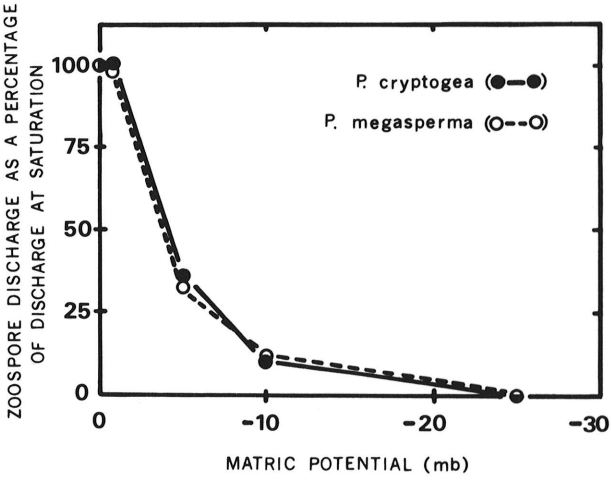

Fig. 3. Influence of soil matric potential (ψ_m) on zoospore discharge by *Phytophthora megasperma* and *P. cryptogea* in the coarse (>250 µm) sand fraction of Yolo fine sandy loam. The soil was allowed to wet from -150 mb ψ_m to the higher ψ_m values for 6 hr, and the total number of zoospores released was determined by counts of germinated spores on a selective medium.

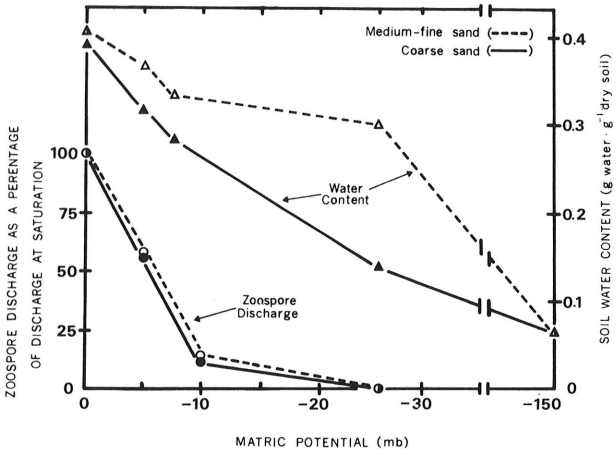

Fig. 4. Influence of matric potential (ψ_m) on soil water content and zoospore discharge by *Phytophthora cryptogea* in the coarse (>250 μm, closed symbols) and medium-fine (60-250 μm, open symbols) sand fractions of Yolo fine sandy loam. Soil water content was measured 6 hr after ψ_m was increased from −150 mb to the higher ψ_m values, and measurements of zoospore discharge are based on the maximum numbers of motile zoospores observed between 1 and 6 hr after ψ_m was increased.

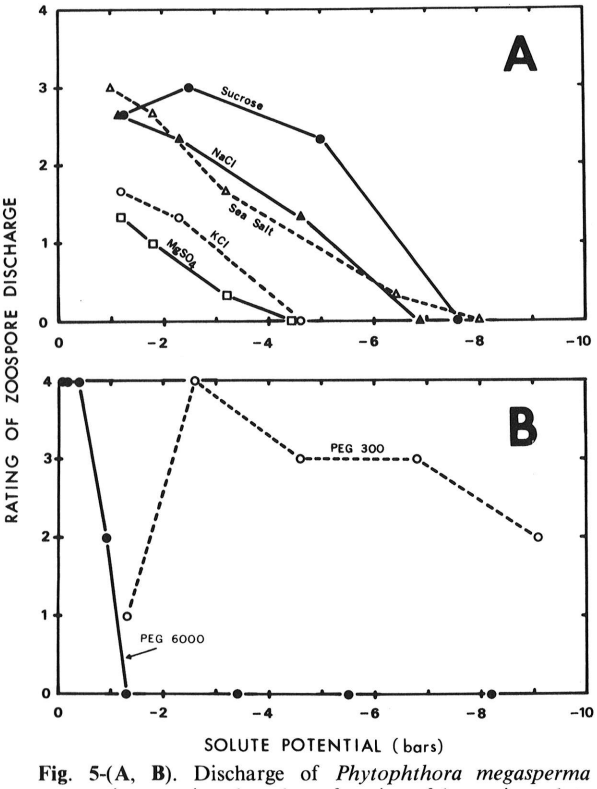

Fig. 5-(A, B). Discharge of *Phytophthora megasperma* zoospores in osmotica plotted as a function of decreasing solute potential. Sporangia were produced in the coarse (>250 μm) sand fraction of Yolo fine sandy loam at −150 mb matric potential and placed in the various solutions. Numbers of zoospores discharged were rated on an arbitrary scale of 0-4 after 2 hr in the solutions.

YFSL; i.e., release was somewhat inhibited at −5 mb, severely restricted at −10 mb, and totally prevented at −25 mb ψ_m (Fig. 4). Prevention of zoospore release at −25 mb in the medium-fine sand occurred even though the water content of the sand was nearly identical with that of the coarse sand at −5 mb, where zoospore release did occur (Fig 4). As in the preceding experiments, soil water content had equilibrated within 1-2 hr and all zoospore release was completed within 6 hr after ψ_m was increased.

Since all of the above experiments employed sporangia formed at $\psi_m = -150$ mb, additional experiments were conducted with *P. cryptogea* to determine whether the ψ_m at which sporangia formed influenced the ψ_m at which zoospore release would occur. Tension plates were prepared to maintain the coarse sand fraction of YFSL at $\psi_m = -50$ or −300 mb for sporangium formation. Following sporangium formation, tension plates were reset to 0, −5, −10, or −25 mb ψ_m and zoospore release was monitored by plating drops of soil suspension on selective agar medium. Zoospore release under these conditions was not significantly different from that by sporangia formed at −150 mb ψ_m.

To determine whether temperature shifts would stimulate zoospore release by *P. cryptogea* at limiting ψ_m values, mycelium disks were incubated in the coarse sand fraction of YFSL on tension plates maintained at −150 mb ψ_m and room temperature (22-24 C). After sporangia had formed, half of the tension plates were placed in a growth chamber at 16 C with the soil at approximately −100 mb ψ_m for 30 min to allow for temperature equilibration. Pairs of tension plates then were set to ψ_m values of 0, −10, or −25 mb in the growth chamber, and a duplicate series of paired tension plates were set to the same ψ_m values at room temperature. After 3 hr at 16 C, one tension plate at each ψ_m value was removed from the chamber and placed at room temperature. Likewise, tension plates were taken from room temperature and placed in the 16 C growth chamber. Observations of zoospore discharge were made by plating drops of soil suspensions on selective medium at 2-, 4-, and 6-hr intervals after ψ_m was increased to the final higher values.

Temperature shifts were not stimulatory to zoospore release in these experiments. At −10 mb ψ_m, zoospore release was severely restricted, regardless of whether the tension plates were maintained at a constant temperature, or moved from a low-to-high or high-to-low temperature. At −25 mb ψ_m, zoospore release did not occur in any of the temperature treatments. The only temperature effects observed were a delay in zoospore release and a general depression in the numbers of zoospores released in soil held at $\psi_m = 0$ and 16 C relative to the treatment, $\psi_m = 0$, at room temperature.

Solute potential effects.—Although only the data for *P. megasperma* are presented in full (Fig. 5), zoospore release by *P. megasperma* and *P. cryptogea* was nearly identical in the osmotica. The lower ψ_s limit for zoospore release in solutions of sucrose, NaCl and synthetic sea salts was −6 to −8 bars ψ_s (Fig. 5-A). Release in solutions of KCl^{-} and $MgSO_4$ was prevented at approximately −4.5 bars ψ_s (Fig. 5-A). Release in PEG 300 occurred at ψ_s values as low as −9.1 bars, whereas release in solutions of PEG 6000 occurred only at $\psi_s > -1.3$ bars, with the greatest release at $\psi_s \geq -0.4$ bar (Fig. 5-B).

DISCUSSION

Although water long has been recognized as a necessary factor for zoospore release by *Phytophthora* (3), the precise effects of soil water on zoospore release have not been characterized previously. Our results demonstrate that zoospore discharge by *P. cryptogea* and *P. megasperma* is extremely sensitive to matric forces in the soil. Whereas release proceeds normally at 0 or −1 mb ψ_m, it is greatly impaired at −5 mb ψ_m, a change of only 4 mb. Further, our observations indicate that a ψ_m of −25 mb is limiting to all stages of indirect germination. When counts of sporangia were made following 24 hr at −25 mb, it was noted that zoospore cleavage had not occurred at this ψ_m and apparently none of the initial steps of indirect germination had taken place, as evidenced by the continued presence of the large central vacuoles in the sporangia (4). Among the results in previous studies of sporangium formation by several species of *Phytophthora* (20, 27), zoospore movement in species of *Phytophthora* (8, 20), host infection by *Olpidium brassicae* (29), of zoospore discharge in *Aphanomyces*, *Phytophthora*, and *Olpidium* (11, 14, 16, 29), one can find some evidence that a variety of zoosporic fungi may have ψ_m requirements for zoospore release similar to those reported here.

Because of its extreme sensitivity to ψ_m, zoospore release appears to have the most exacting ψ requirement of any stage in the life cycle of *Phytophthora* spp. (6, 7, 16, 21, 23, 24, 25, 26), and zoospore release is probably unique in its sensitivity among the ψ requirements in the life cycles of all the fungi experimented with to date (1, 5, 12). Although Duniway (8) showed a similar limitation on zoospore movement through soil at relatively small matric tensions, this was believed to be the result of a draining of the large soil pores needed to accommodate swimming zoospores. In contrast to zoospore movement, the results obtained with different textured fractions of soil (Fig. 4) indicate that ψ_m has a greater influence on zoospore discharge than does soil water content. Likewise, the experiments with temperature shifts showed ψ_m has a much greater influence on zoospore release than changes in temperature within the range where release can occur. Although temperature manipulations do not appear to be stimulatory to zoospore release, temperatures of 33-36 C appear to represent an upper limit for release. Sporangia held at these temperatures under optimum ψ_m conditions do not release zoospores until removed to lower temperatures (MacDonald and Duniway, *unpublished*).

While the ψ_m limits on zoospore discharge described here (Figs. 3 and 4) refer to limits on the rapid, synchronous discharge by a population of young sporangia, it has been reported that a fraction of the sporangium population of *P. drechsleri* and *P. megasperma* held in soil at −50 to −300 mb ψ_m may undergo indirect germination after 7-12 days (6, 20). However, these reports did not employ methods of direct observation of zoospore discharge and it is not known whether discharge under these conditions occurred in a typical manner. Although longer periods of time may allow some zoospore discharge at marginal ψ_m values, an explanation of these observations in relation to our findings on ψ_m limitations is difficult without knowing the precise mechanism of discharge. Direct germination of sporangia, on the other hand, evidently can occur (6, 7, 20, 27) in soils at ψ_m values too low to permit indirect germination.

The observations of zoospore discharge in osmotica described here agree well with those reported by Katsura (16) for *P. capsici*. Zoospores were released in all solutions, except PEG 6000, at ψ_s values as low as −3, −5, or −9 bars (Fig. 5). This contrasts with the −25 mb ψ_m limit on zoospore discharge, and respresents a 120- to 360-fold difference in response to the ψ_m and ψ_s components of ψ. These results suggest that sporangia can take up certain solutes from surrounding solutions and thereby discharge zoospores at lower ψ_s values than would be predicted from the ψ_m effects. This view is supported by our observations of zoospore discharge in solutions of PEG. The lower- molecular-weight PEG 300 allowed zoospore discharge at ψ_s values as low as −9.1 bars (Fig. 5-B). On the other hand, the higher-molecular-weight PEG 6000, which would not so readily enter cells, completely prevented zoospore discharge at ψ_s values $\leqslant$ −1.3 bars (Fig. 5-B). Hoch and Mitchell (14, 15) also found that solutions of higher-molecular-weight PEG prevented zoospore formation in *A. euteiches* at much higher ψ_s values than did other solutes. The differential effects of PEG 300, sucrose, and inorganic ions on zoospore discharge (Fig. 5) are probably due to their differential uptake by sporangia, but specific ion toxicities also could be involved (26).

Even though zoospore discharge can occur in solutions of sea salts at ψ_s values as low as −6.5 bars, it is apparent that discharge would not be expected to occur in full-strength sea water, which has a ψ_s less than −20 bars. Apparently zoospore discharge by these presumably terrestrial or fresh water species is more sensitive to ψ_s than is discharged by marine species of the same genus (9).

Blackwell and Waterhouse (3) provided a generalized description of zoospore discharge in *Phytophthora*. They indicated that the zoospores may be expelled as a mass in a thin vesicle which ultimately bursts and allows them to swim away and that the mass appears to be forcibly pressed out of the sporangium by an inner vesicle. Although in some species of *Phytophthora* the zoospores exit the sporangium singly (4), only mass expulsion was observed in the species used in this study. The central question is, by what force are the zoospores expelled from the sporangium and how is it affected by ψ? Fuller (10) indicated that available evidence argues strongly for a pressure buildup within sporangium prior to zoospore release. While the source of such a pressure apparently has not been elucidated, one common explanation is that osmotic pressures are generated by the conversion of glycogen and/or other polysaccharides to glucose (4, 10). However, if zoospore discharge was dependent simply on osmotic adjustments, it should not be limited by small ψ_m deficits, particularly since it can operate in solutions of −5 to −9 bars ψ_s. Other mechanisms have been hypothesized by Webster and Dennis (28), who suggested a surface tension model for the discharge of cytoplasm into the sporangium vesicle of *Pythium middletonii*, and Fuller (10), who suggested that contractile proteins may function in zoospore discharge.

One possible mechanism which has not previously been hypothesized is that of a swelling gel matrix which may

generate at least some of the pressures necessary for zoospore discharge. Gel matrices exhibit very sharp dewatering curves when ψ_m is decreased (30), and in rewetting cycles, such as those employed in these experiments, would require a ψ_m of nearly 0 for complete hydration. While there is no direct evidence that a gel contributes to release, such a mechanism could explain the great sensitivity of zoospore discharge to slight ψ_m deficits. Swelling gel-like substances apparently occur among other fungi, such as the Sphaeropsidales. Although their response to ψ_m is not known, these fungi typically expel large numbers of conidiospores from pycnidia in a slime matrix or cirrhus when wetted (2).

In spite of the fact that ψ_m, which is a function of the adsorption of water onto soil particles and the capillarity of soil pores, can be a dominant component of soil ψ (12), much of the experimental work on the ψ requirements of soil fungi has used agar or liquid systems where ψ is controlled almost exclusively by ψ_s (15, 16, 24, 26). However, ψ_s and ψ_m are sufficiently different that care must be exercised when attempting to draw conclusions about one based on a response to the other. While Adebayo and Harris (1) found a consistent and predictable relationship between the ψ_m and ψ_s effects on growth by *P. cinnamomi* and *Alternaria tenuis*, the ψ_s limits on zoospore discharge described here and elsewhere (15, 16) seem insignificant compared to the limits imposed by a few thousandths of a bar ψ_m. Indeed, in nonsaline agricultural soils, ψ_s probably is insignificant in zoospore discharge, being rarely less than −2 bars (22) at the high ψ_m values which permit discharge. The high ψ_m requirement for zoospore discharge (Figs. 3 and 4) and movement through soil (8) may partially explain the frequent association of root and crown rots caused by *Phytophthora* spp. with saturated soil conditions. This requirement for near-saturated conditions indicates that the aquatic affinities of this genus have not been lost even by those members that appear well adapted to a soil habitat.

LITERATURE CITED

1. ADEBAYO, A. A., and R. F. HARRIS. 1971. Fungal growth responses to osmotic as compared to matric water potential. Soil Sci. Soc. Am. Proc. 35:465-469.
2. ALEXOPOLOUS, C. J. 1962. Introductory mycology. John Wiley and Sons, New York. 613 p.
3. BLACKWELL, E. M., and G. M. WATERHOUSE. 1930. Spores and spore germination in the genus Phytophthora. Trans. Br. Mycol. Soc. 15:294-310.
4. CHAPMAN, J. A., and R. VUJIČIĆ. 1965. The fine structure of sporangia of Phytophthora erythroseptica Pethyb. J. Gen. Microbiol. 41:275-282.
5. COOK, R. J. 1973. Influence of low plant and soil water potentials on diseases caused by soilborne fungi. Phytopathology 63:451-458.
6. DUNIWAY, J. M. 1975. Formation of sporangia by Phytophthora drechsleri in soil at high matric potentials. Can. J. Bot. 53:1270-1275.
7. DUNIWAY, J. M. 1975. Limiting influence of low water potential on the formation of sporangia by Phytophthora drechsleri in soil. Phytopathology 65:1089-1093.
8. DUNIWAY, J. M. 1976. Movement of zoospores of Phytophthora cryptogea in soils of various textures and matric potentials. Phytopathology 66:877-882.
9. FELL, J. W., and I. M. MASTER. 1975. Phycomycetes (Phytophthora spp. nov. and Pythium sp. nov.) associated with degrading mangrove (Rhizophora mangle) leaves. Can. J. Bot. 53:2908-2922.
10. FULLER, M. S. 1977. The zoospore, hallmark of the aquatic fungi. Mycologia 69:1-20.
11. GISI, U. 1975. Investigations on the soil phase of Phytophthora cactorum (Leb. et Cohn) Schroet. with fluorescent optical direct observation. Z. Pflanzenpathol. Pflanzenschutz. 82:355-377. (in German with English summary).
12. GRIFFIN, D. M. 1972. Ecology of soil fungi. Syracuse University Press, Syracuse, New York 193 p.
13. HICKMAN, C. J., and H. H. HO. 1966. Behaviour of zoospores in plant-pathogenic phycomycetes. Annu. Rev. Phytopathol. 4:195-220.
14. HOCH, H. C., and J. E. MITCHELL. 1970. The effects of water potential on zoospore production in Aphanomyces euteiches. Phytopathology 60:1296 (Abstr.).
15. HOCH, H. C., and J. E. MITCHELL. 1973. The effects of osmotic water potentials on Aphanomyces euteiches during zoosporogenesis. Can. J. Bot. 51:413-420.
16. KATSURA, K. 1971. Some ecological studies on zoospore of Phytophthora capsici Leonian. Rev. Plant Protection Res. 4:58-70.
17. KLIEJUNAS, J. T., and W. H. KO. 1974. Effect of motility of Phytophthora palmivora zoospores on disease severity in papaya seedlings and substrate colonization in soil. Phytopathology 64:426-428.
18. MC INTOSH, D. L. 1972. Effects of soil water suction, soil temperature, carbon and nitrogen amendments, and host rootlets on survival in soil of zoospores of Phytophthora cactorum. Can. J. Bot. 50:269-272.
19. MEHROTRA, R. S. 1972. Behavior of zoospores of Phytophthora megasperma var. sojae and P. drechsleri in soil. Can. J. Bot. 50:2125-2130.
20. PFENDER, W. F., R. B. HINE, and M. E. STANGHELLINI. 1977. Production of sporangia and release of zoospores by Phytophthora megasperma in soil. Phytopathology 67:657-663.
21. REEVES, R. J. 1975. Behaviour of Phytophthora cinnamomi Rands in different soils and water regimes. Soil Biol. Biochem. 7:19-24.
22. RICHARDS, L. A. (ed.). 1954. Diagnosis and improvement of saline and alkali soils. U.S. Dep. Agric., Agric. Handbook No. 60. 157 p.
23. SNEH, B., and D. L. MC INTOSH. 1974. Studies on the behavior and survival of Phytophthora cactorum. Can. J. Bot. 52:795-802.
24. SOMMERS, L. E., R. F. HARRIS, F. N. DALTON, and W. R. GARDNER. 1970. Water potential relations of three root-infecting Phytophthora species. Phytopathology 60:932-934.
25. STERNE, R. E. 1976. The relationship of soil and plant water status to Phytophthora root rot of avocado. Ph.D. Dissertation. Univ. of California, Riverside, 145 p.
26. STERNE, R. E., G. A. ZENTMYER, and F. T. BINGHAM. 1976. The effect of osmotic potential and specific ions on growth of Phytophthora cinnamomi. Phytopathology 66:1398-1402.
27. SUGAR, D. 1977. The development of sporangia of Phytophthora cambivora, P. megasperma, and P. drechsleri and severity of root and crown rot of Prunus mahaleb as influenced by soil matric potentials. M. S. Thesis, University of California, Davis. 56 p.
28. WEBSTER, J., and C. DENNIS. 1967. The mechanism of sporangial discharge in Pythium middletonii. New Phytol. 66:307-313.
29. WESTERLUND, F. V., JR. 1977. Environmental factors affecting Olpidium brassicae and transmission of the lettuce Big Vein agent. Ph.D. Dissertation. University of

California, Davis. 58 p.
30. WIEBE, H. H. 1966. Matric potential of several plant tissues and biocolloids. Plant Physiol. 41:1439-1442.
31. ZENTMYER, G. A., and D. C. ERWIN. 1970. Development and reproduction of Phytophthora. Phytopathology 60:1120-1127.