Effect of Ion Concentration and Sodium:Calcium Ratio of a Nutrient Solution on Phytophthora Root Rot of Tomato and Zoospore Motility and Viability of *Phytophthora parasitica*

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ABSTRACT

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In two sets of greenhouse experiments, the effect of four Na:Ca equivalent ratios (0, 1, 5, and 10) on Phytophthora root rot of tomato was studied at two ionic concentrations (2.5 vs. 25 meq L^{-1} or 25 vs. 50 meq L^{-1}) of a modified Hoagland's solution. Two weeks after planting, the plants were either kept at the same ionic concentration or were shifted from low to high or high to low concentration, and half of the plants in each treatment were inoculated with zoospores of *Phytophthora parasitica*. The percentage of root rot was assessed visually 2 wk after inoculation. Root rot severity increased significantly with increasing Na:Ca ratios at ion concentrations of 2.5 and 25 meq L^{-1} before or after inoculation. Salt stress at 50 meq L^{-1} before inoculation increased root rot. Salt stress at 50 meq L^{-1} after inoculation reduced root rot caused by an isolate of *P. parasitica* originating from nonsaline soil, particularly at higher Na:Ca ratios of 5 and 10. Root rot caused by an isolate originating from saline soil was not reduced. Percentages of motile and germinated zoospores decreased in vitro, and those of encysted and lysed zoospores increased with increasing salt concentrations and Na:Ca ratios. These effects were more pronounced for the isolate from nonsaline soil than for the isolate from saline soil. The isolate from saline soil lost its relative salt tolerance after 2 mo in culture. Inoculation of tomato seedlings with this isolate after 2 mo in culture resulted in root rot severity similar to that caused by the isolate from nonsaline soil when salt stress was applied during and after inoculation.

Phytophthora root and crown rot, which is incited by several *Phytophthora* species but primarily by *P. parasitica* Dastur (16), is a major disease of tomatoes in California (2). Disease severity depends on soil factors, such as aeration, irrigation, drainage, and salinity (2,28,31,34).

Soil salinity is an increasing problem in the arid areas of California (4,22), and several root diseases are enhanced under saline soil conditions (9,28,29,34). Phytophthora root rot of citrus (9), chrysanthemum (25,26), tomato (34), and tobacco (3), and Pythium blight of creeping bentgrass (29) were shown to be more severe under saline conditions. Damping-off of tomato seedlings by *Rhizoctonia solani* Kühn and wilt by *Fusarium oxysporum* Schlechtend.:Fr. f. sp. *lycopersici* (Sacc.) W. C. Snyder & H. N. Hans. were enhanced in increasingly concentrated nutrient solutions (5). In contrast, high salt concentrations decreased Phytophthora root rot of pineapple caused by *P. cinnamomi* Rands (1) and damping-off of tomato and cucumber seedlings by *Pythium ultimum* Trow (5).

The mechanisms by which salinity affects plant growth and susceptibility to root rots and wilts are not known exactly (20,25, 26). Early workers postulated that salt stress exerted its influence on the plant by means of the more negative osmotic potential of the soil environment (6). However, ion imbalance and toxicity have also been implicated in the effects of salinity on plant growth (12,18,20). Ion imbalance can occur when high concentrations of Na compete for uptake with Ca, K, and Mg (18). Once in cells, Na can have toxic effects by interfering with enzymes or the function of membranes (21). Several authors (12,18) have postulated that Na displaces Ca from membranes, resulting in a loss of selective permeability. This may, in turn, lead to increased root exudation (12).

In earlier studies with Phytophthora root rot, a distinction was made between predisposition of the plant to salt stress before inoculation and the reaction to salt stress after inoculation (25,26, 34). Salt stress applied before inoculation of chrysanthemum with *P. cryptogea* Pethybr. & Lafferty increased the number of zoospore cysts attached to the root surface, which probably resulted from increased root exudation (25). Colonization of chrysanthemum roots by *P. cryptogea* was also more rapid when salt stress was applied within 24 hr after inoculation, indicating that the host defense mechanisms were impaired (26). Similarly, Phytophthora root rot of tomato was more severe when young plants were exposed to chronic salt stress both before or after inoculation with *P. parasitica* (34).

In the experiments with *P. cryptogea* and *P. parasitica* (25,26,34), salt solutions were applied at relatively high concentrations (electrical conductivity [EC] = 7–25 dS m^{-1}) and had fixed Na:Ca ratios (equivalent ratio Na:Ca = 5:1 or 1:1). However, under field conditions, several ions may be present in various ratios (6,30), and the Na:Ca ratio may be as important as the overall ion concentration in determining the effect of salinity on root rot severity (10). For example, Fusarium wilt of tomato developed faster in nutrient solutions at increasing Na:Ca ratios (32,33). The influence of Na:Ca ratios on disease severity has not been determined for Phytophthora root rots.

Ion concentration and ratio can also affect zoospore motility and viability of *Phytophthora* spp. Various researchers (7,11,14) have shown increased encystment with increasing ion concentrations of 1–30 mM. Sodium reduced the ability of encysted zoospores to germinate, but calcium stimulated cyst germination (11,14). In addition to a reduction in germination, Na concentrations between 10 and 30 mM caused more zoospore lysis than similar concentrations of Ca (14). Similar studies have not been performed for *P. parasitica*.

Thus far, only the effects of relatively high levels of salinity stress at fixed Na:Ca ratios have been studied in relation to Phytophthora root rot. Our study was undertaken to determine the more subtle effects of slightly saline nutrient solutions at various Na:Ca ratios on Phytophthora root rot of tomato and motility, encystment, germination, and lysis of zoospores of *P. parasitica*.

MATERIALS AND METHODS

Nutrient solutions. Two basic solutions were prepared—a slightly modified Hoagland's solution (15) and another solution similar to the first but with all Ca ions replaced by equivalent amounts of Na (Table 1). The two solutions were mixed together in various proportions to obtain four Na:Ca equivalent ratios of 0:10, 1:1, 5:1, and 10:1. The pH was adjusted to 6.0 by adding 0.1 M KOH. To obtain two levels of ion concentrations, the concentrated solutions at four Na:Ca ratios were diluted to the required levels. Thus, differences in electrical conductivity (EC) were attributable to differences in concentrations of all ions, not just NaCl and $CaCl_2$. The EC was determined with a La Motte multirange conductivity meter (La Motte Chemical Products Co., Chestertown, MD), and the water potential (ψ) with a thermocouple psychrometer (model SC-10A, Decagon Devices Inc., Pullman, WA). The EC ranged from 0.2 to 4.0 dS m^{-1} at ion concentrations ranging from 2.5 to 50 meq L^{-1}. A soil with an EC of a saturated paste extract (EC_e) of 4.0 dS m^{-1} or higher is considered saline (4,22,30). The corresponding ψ values ranged from 0 to 250 kPa. At the end of each experiment, the pH and EC of the solutions were determined again for all treatments.

Preparation of zoospore suspensions. Most experiments were conducted with one isolate of *P. parasitica* (S−) isolated in 1987 from a commercial tomato field northwest of Davis, CA. (This isolate was obtained from Deborah Neher, Department of Plant Pathology, University of California-Davis.) Other experiments were performed with an isolate of *P. parasitica* (S+) recovered from a university experimental field at Davis, in which saline irrigation water (Na:Ca equivalent ratio = 2:1) had been applied for 3 yr. The EC_e from this latter soil was 6 dS m^{-1}. Isolates were maintained on cornmeal agar (CMA) slants at room temperature and were transferred once every 6 mo. Zoospore suspensions were prepared as follows: mycelial squares (5 × 5 mm) were cut from 7-day-old colonies grown on V-8 juice agar (200 ml of V-8 juice, 2 g of $CaCO_3$, and 17 g of Difco agar in 1 L of distilled water) and incubated in the light in petri dishes containing distilled water at room temperature for 3 days. After sporangia had formed, the petri dishes were chilled for 1 hr at 4 C and returned to room temperature for 1 hr to stimulate zoospore release. Zoospores were separated from V-8 agar and hyphae by filtration through cheesecloth, and spore concentrations were determined with the aid of a hemacytometer.

Greenhouse experiments. Seeds of tomato (*Lycopersicon esculentum* Mill. 'Royal Flush') were sown in vermiculite and moistened with deionized water. After 1 wk, seedlings were transplanted into nutrient solutions with four Na:Ca ratios (0:10, 1:1, 5:1, or 10:1 meq:meq) at a low or high ionic concentration (2.5 or 25 meq L^{-1}) in 1-L glass jars wrapped with aluminum foil. Sponge stoppers at the lower stem were used to support the plants in a hole in the jar lids. Nutrient solutions were aerated by constant bubbling of humidified air into each jar, and solutions were replaced with fresh solutions weekly. Experiments were conducted in a greenhouse with supplemental lighting provided by fluorescent tubes for 14 hr per day. Daily temperatures fluctuated between 29 and 31 C during the day and 18 and 24 C at night.

After 2 wk in nutrient solutions, half the plants in the 2.5 meq L^{-1} solution were transferred to 25 meq L^{-1} and half of those in 25 meq L^{-1} solution were shifted to 2.5 meq L^{-1}. Immediately after changing the solutions, half of the plants in all solution treatments were inoculated with zoospores (2–5 × 10^4 zoospores per jar) of *P. parasitica* (S−); the other half were kept as controls. The shifts from high to low and low to high ion concentrations just before inoculation allowed for distinction between the effects of ion concentrations before and during or after inoculation. Plants were harvested and visually evaluated for root rot severity (percentage of the root system necrotic) 2 wk after inoculation. To confirm that the necrosis evident on the roots was caused by *P. parasitica*, five root sections (1-cm long) of six inoculated and six uninoculated plants were rinsed in sterile distilled water, blotted dry, and placed on PARP medium (17). Remaining shoot and root tissues were dried at 80 C for 48 hr, and plant dry weights were determined. Dry weights of inoculated plants (mean of six replicates) were expressed as percentages of corresponding uninoculated plants. The percent of reduction in dry weight attributable to *P. parasitica* was calculated and subjected to statistical analysis.

Another experiment was performed as described above, but with higher ionic concentrations (25 and 50 meq L^{-1}). All plants were first exposed to 25 meq L^{-1} for 1 wk and then transferred to fresh solutions at either 25 or 50 meq L^{-1} for another week. Thus, the preinoculation salt stress at 50 meq L^{-1} lasted 1 wk. After that week, half the plants at 25 meq L^{-1} were shifted to 50 meq L^{-1} and half those at 50 meq L^{-1} were shifted to 25 meq L^{-1}. Half of the plants in each solution treatment were inoculated with a zoospore suspension of 10^4 zoospores per jar. Plants were harvested 2 wk after inoculation as described for the first experiment. This experiment was conducted with each of two isolates of *P. parasitica* (S− and S+) in separate experiments.

Each experiment was arranged in a completely randomized design, with six or four replicates in the first (2.5 and 25 meq L^{-1}) and second (25 and 50 meq L^{-1}) series of experiments, respectively. All experiments were conducted twice. The percentage of the root system discolored and shoot and root dry weights were first subjected to regression analyses (SAS Institute, Inc., Cary, NC) for each individual experiment (10). The residual values were checked for homogeneity and normality (SAS Institute, Inc., Cary, NC). When the residual values were not normally distributed, the means of six (or four) observations per experiment were subjected to regression analyses in which the experiments were considered as blocks. Analysis of the means normalized the residual values as determined by the Shapiro-Wilk statistic (SAS Institute).

Zoospore motility, encystment, germination, and lysis. The effects of four ion concentrations (0, 2.5, 25, and 50 meq L^{-1}) in nutrient solutions on zoospore motility, encystment, and germination were compared for isolates S− and S+. Nutrient solutions had a constant Na:Ca equivalent ratio of 5:1. Zoospores were released in distilled water as described above, and 1 ml of zoospore suspension was transferred to 100 ml of each of the four nutrient solutions. Final zoospore suspensions were kept at room temperature for 30 min. A counting grid of a 15 × 17 mm piece of plastic window screen was mounted on a microscope slide (27) to facilitate counting of motile, germinated, encysted, and lysed zoospores in 50 μl of the final suspension. Fifty microliters of suspension contained about 10–20 zoospores. Each treatment was replicated five times so that 50–100 zoospores were counted for each treatment. The eight treatments (four ion concentrations × two isolates) were assigned randomly to the experimental units

TABLE 1. Mineral salts added (ml L^{-1}) and ion concentrations (meq L^{-1}) of modified full-strength Hoagland's solutions with equivalent amounts of Ca (solution 1) or Na (solution 2)[a]

Salt	ml L^{-1}		Ion	meq L^{-1}	
	Solution 1	Solution 2		Solution 1	Solution 2
1 M KH_2PO_4	1	1	K^+	6	6
1 M KNO_3	3	5	H^+	2	2
1 M $MgSO_4$	2	2	Mg^{2+}	4	4
1 M NH_4NO_3	3	3	NH_4^+	3	3
EDTA FeNa	1	1	NO_3^-	12	12
Micronutrients[b]	1	1	SO_4^{2-}	6	6
1 M $Ca(NO_3)_2$	3		PO_4^{3-}	3	3
1 M $CaCl_2$	2		Cl^-	4	4
1 M K_2SO_4	1		Ca^{2+}	10	
1 M $NaNO_3$		4	Na^+		10
1 M NaCl		4			
1 M $NaSO_4$		1			

[a] Solutions 1 and 2 were mixed together in varying proportions to obtain various Na:Ca ratios.

[b] Micronutrients: 2.86 g of H_3BO_3, 1.81 g of $MnCl_2 \cdot 4H_2O$, 0.22 g of $ZnSO_4 \cdot 7H_2O$, 0.08 g of $CuSO_4 \cdot 5H_2O$, and 0.02 g of $H_2MoO_4 \cdot H_2O$ per liter of water.

(beakers with nutrient solution) in each of five blocks over time.

In another experiment, two ion concentrations (25 and 50 meq L^{-1}) and four Na:Ca equivalent ratios (0:10, 1:1, 5:1, and 10:1) were used. This experiment was conducted with isolates S− and S+ in separate tests. Zoospores were released in distilled water. The suspensions were diluted 100-fold in the eight nutrient solutions and kept for 30 min at room temperature. There were five replicates in a randomized complete block design. Motile, germinated, encysted, and lysed zoospores were counted as described above.

Each experiment was conducted twice. Percentages of zoospores in each category were analyzed by regression analysis (SAS Institute) in which the replicates were considered as blocks. Residual values were tested for homogeneity and normality.

RESULTS

Greenhouse experiments. The pH, EC, and ψ of freshly prepared nutrient solutions were not affected by Na:Ca ratios. The pH was slightly lower ($P = 0.05$) at higher ion concentrations (5.9 at 50 meq L^{-1} vs. 6.1 at 25 and 2.5 meq L^{-1}), and EC values were proportional to the ion concentrations (0.3, 2.2, and 4.2 dS m^{-1} at 2.5, 25, and 50 meq L^{-1}, respectively). The ψ also reflected the ion concentrations (240 kPa at 50 meq L^{-1} and 180 kPa at 25 meq L^{-1}). At the end of each experiment, the pH of the nutrient solutions was still slightly lower ($P = 0.01$) at the higher ion concentrations (6.5, 6.4, and 6.2 at 2.5, 25, and 50 meq L^{-1}, respectively). The EC and ψ of 1-wk-old nutrient solutions still reflected the initial ion concentrations, but the EC was, on average, 0.3 dS m^{-1} lower than that of fresh solutions, which may be attributable to daily replacement of water in the jars. The pH EC, and ψ of the solutions were unaffected by plant inoculation with *P. parasitica*.

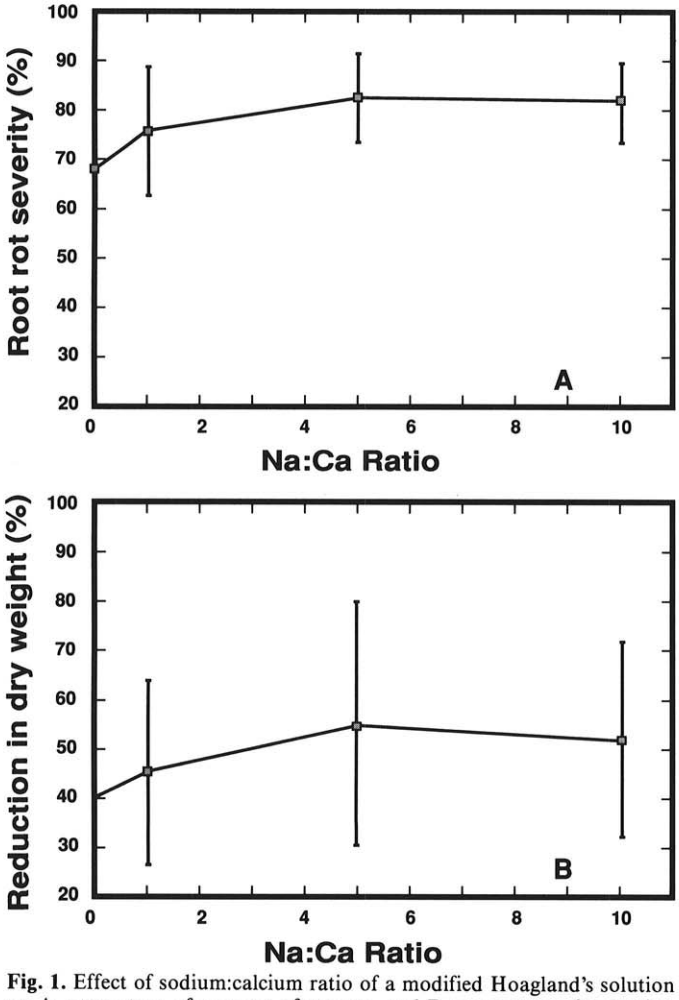

Fig. 1. Effect of sodium:calcium ratio of a modified Hoagland's solution on **A,** percentage of root rot of tomato, and **B,** percentage of reduction in root dry weight attributable to infection by isolate S− of *Phytophthora parasitica* from nonsaline soil. Treatments with ionic concentrations of 2.5 and 25 meq L^{-1} before and after inoculation were combined. Vertical bars denote standard deviations.

In the absence of *P. parasitica*, roots exposed to solutions with high Na:Ca ratios (5 or 10) developed a light reddish brown discoloration, especially when the overall ionic concentration was low (2.5 or 25 meq L^{-1}). This discoloration was more pronounced at 2.5 than at 25 meq L^{-1}. Infection by *P. parasitica* resulted in grayish brown root discoloration, which could be distinguished easily from the reddish discoloration caused by high Na:Ca ratios. Isolations from root tissues confirmed that *P. parasitica* was present only in inoculated and not in uninoculated roots.

In the experiments with 2.5 and 25 meq L^{-1} and four Na:Ca ratios, no significant interactions occurred between ion concentrations before and after inoculation and Na:Ca ratio for root rot severity or the percent of reduction in shoot and root dry weights (P ranged from 0.15 to 0.80). Main effects of total ion concentrations before or after inoculation on root rot severity were also not significant. However, averaged across ion concentrations, disease severity increased ($P = 0.03$) as the Na:Ca ratio increased (Fig. 1A). Shoot and root dry weights were reduced ($P = 0.001$) by Phytophthora root rot, and the mean percent reduction in dry weight varied from 12 to 81%. Percent reduction in root dry weight attributable to *P. parasitica* was slightly higher as the Na:Ca ratio increased (Fig. 1B), but the effect was not significant. Shoot weight was not affected by Na:Ca ratios.

When ionic concentrations were 25 and 50 meq L^{-1}, significant interactions occurred between pre- and postinoculation salt stress with respect to root rot severity for either isolate S− or S+ ($P = 0.05$ and < 0.01, respectively). Exposure of the roots to salt stress (50 meq L^{-1}) before inoculation increased root rot severity from 79 to 93% for isolate S−, and from 77 to 88% for isolate S+, if the postinoculation ion concentration was 25 meq L^{-1} (Table 2). However, preinoculation stress had no effect on root rot severity when salt stress of 50 meq L^{-1} was continued after inoculation. The interaction between preinoculation salt stress and Na:Ca ratio was significant for isolate S+ (Table 2). Root rot severity increased significantly with increasing Na:Ca ratios when the preinoculation ion concentration was 25 meq L^{-1}, but not when it was 50 meq L^{-1}.

Averaged over all Na:Ca ratios, salt stress of 50 meq L^{-1} during and after inoculation reduced root rot severity induced by isolate S− from 90 to 77%. However, there was a significant interaction between postinoculation salt stress and Na:Ca ratio with respect

TABLE 2. Effect of ion concentration (meq L^{-1}) and Na:Ca ratio before inoculation[a] with isolates S− and S+ of *Phytophthora parasitica* on root rot severity (%) of 5-wk-old tomato plants

Na:Ca ratio	Root rot severity (%)			
	Isolate S−		Isolate S+	
	25 (meq L^{-1})	50 (meq L^{-1})	25 (meq L^{-1})	50 (meq L^{-1})
0	80 ± 8[b]	94 ± 10	62 ± 4	84 ± 3
1	90 ± 9	93 ± 7	74 ± 11	89 ± 5
5	72 ± 13	96 ± 3	85 ± 4	83 ± 3
10	74 ± 30	89 ± 9	86 ± 6	95 ± 4
Means	79 ± 17	93 ± 7	77 ± 12	88 ± 6

Analysis of variance for root rot severity

Source	df[c]	MS[d]	$P > F$	df	MS	$P > F$
Pre[e]	1	1,512	0.01	1	979	<0.01
Ratio[f]	3	146	0.50	3	418	<0.01
Pre × ratio	3	155	0.50	3	219	<0.01
Error	24	178		28	31	

[a] Ion concentration after inoculation = 25 meq L^{-1}.
[b] Standard deviation.
[c] Degrees of freedom.
[d] Mean squares.
[e] Preinoculation ion concentration.
[f] Na:Ca ratio.

to root rot severity ($P = 0.05$). Root rot severity was progressively reduced as the Na:Ca ratio increased when the ion concentration after inoculation was 50 meq L^{-1} (Fig. 2A) but was not affected by Na:Ca ratio at 25 meq L^{-1}. Similar interactions between postinoculation salt stress and Na:Ca ratio were obtained for the percent reductions in shoot and root dry weight attributable to infection by isolate S− of *P. parasitica* ($P = 0.05$ and 0.11, respectively [Fig. 2B]). Analogous to root rot severity, the percent reduction in shoot and root dry weight decreased at increasing Na:Ca ratio, and this decrease was more pronounced at 50 than at 25 meq L^{-1} after inoculation.

Unlike isolate S−, isolate S+, when freshly isolated from saline soil, induced more root rot ($P = 0.01$) as the Na:Ca ratio increased from 0 to 10, when salt stress of 50 meq L^{-1} was applied during and after inoculation (Fig. 2A), indicating that isolate S+ was more tolerant to high Na:Ca ratios than isolate S−. There was no significant interaction between postinoculation ion concentration and Na:Ca ratio. Nevertheless, the average root rot severity was slightly but significantly ($P = 0.02$) lower at 50 than at 25 meq L^{-1} after inoculation (78 and 82% infection, respectively). When the experiment with isolate S+ was repeated 1 mo later, results were almost identical to those obtained with isolate S− (Fig. 2A). There was a significant ($P = 0.10$) interaction between postinoculation ion concentration and Na:Ca ratio. Postinoculation salt stress of 50 meq L^{-1} decreased root rot severity more at Na:Ca ratios of 5 and 10 than at ratios of 0 and 1 (Fig. 2A). Percent reduction in shoot and root dry weights caused by Phytophthora root rot again reflected root rot severity. The reduction in root dry weight by isolate S+ was slightly larger when S+ was recently obtained from saline soil (Fig. 2B) but decreased when it had been in culture for 2 mo.

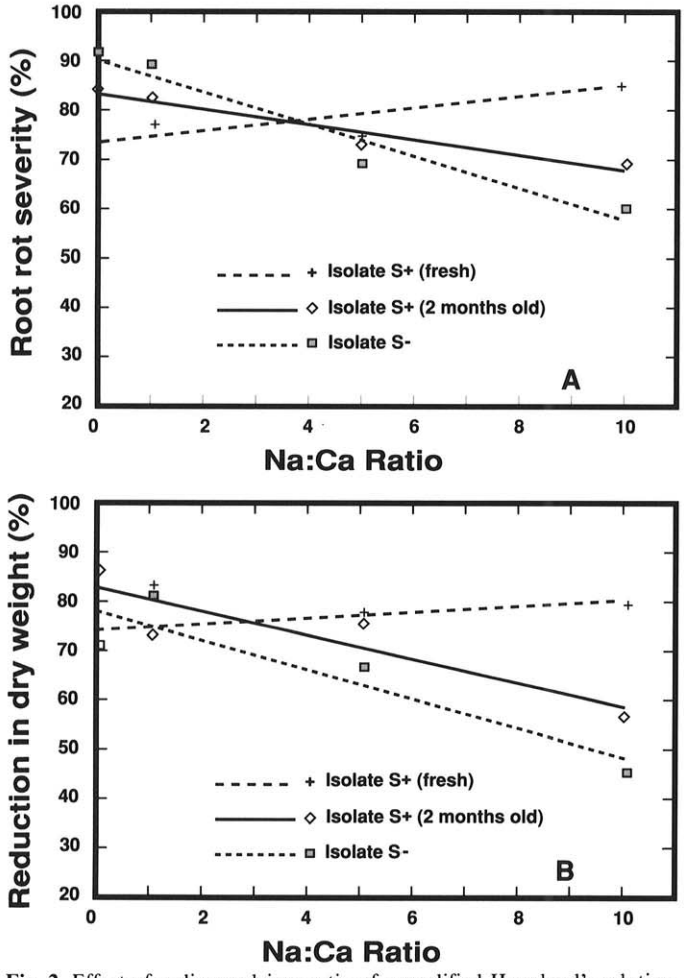

Fig. 2. Effect of sodium:calcium ratio of a modified Hoagland's solution at 50 meq L^{-1} after inoculation of tomato seedlings with *Phytophthora parasitica* on **A,** percentage of root rot, and **B,** percentage of reduction in root dry weight attributable to infection by isolate S− of *P. parasitica* from nonsaline soil, isolate S+ recently obtained from saline soil, and isolate S+ after 2 mo in culture. Treatments with ion concentrations of 25 and 50 meq L^{-1} before inoculation were combined.

Zoospore motility, encystment, germination, and lysis. The percentages of motile or germinated zoospores of isolate S− from nonsaline soil and isolate S+ recently obtained from saline soil decreased ($P = 0.001$) at increasing ion concentration from 0 to 50 meq L^{-1} (Fig. 3A). However, the decrease in percentage of motile zoospores was less ($P = 0.001$) for isolate S+. Conversely, percentages of encysted and lysed zoospores increased ($P = 0.001$) with increasing ion concentrations, but significantly ($P = 0.001$) less so for isolate S+ (Fig. 3B). Zoospores of isolates S− and S+ were compared again at the same four ion concentrations when the isolates had been on CMA slants for 4 mo. During that time period, isolate S+ had lost its tolerance to high ion concentrations, and zoospores of both isolates S− and S+ reacted similarly to the different ion concentrations (Fig. 3C and D).

Besides the effect of total ion concentration, the Na:Ca ratio also had a pronounced effect on zoospore motility and viability, and there were significant interactions between Na:Ca ratios and ion concentrations ($P = 0.01$). Percentages of motile and germinated zoospores of isolate S− decreased ($P = 0.001$), whereas percentages of encysted and lysed zoospores increased ($P = 0.001$) as Na:Ca ratios increased from 0 to 10 (Fig. 4A and B). Moreover, the percentages of motile and germinated zoospores were lower and those of encysted and lysed zoospores higher at 50 meq L^{-1} than at 25 meq L^{-1} (Fig. 4A and B). For isolate S+, freshly isolated from saline soil, percentages of motile and germinated zoospores also decreased ($P = 0.001$) with an increasing Na:Ca ratio and ion concentration (Fig. 4C). However, at 50 meq L^{-1} and Na:Ca of 10, the percentage of motile zoospores decreased less than that of isolate S− (Fig. 4A and C). Percentages of encysted and lysed zoospores increased ($P = 0.002$ and 0.001, respectively) with increasing Na:Ca ratio and ion concentration in a similar manner as those of isolate S− (Fig. 4D). When the experiment with isolate S+ was repeated after the isolate had been in culture for 2 mo, the decrease in zoospore motility and germination and increase in encystment and lysis with increasing Na:Ca ratios and ion concentrations were similar to those of isolate S−. These results are consistent with the loss of salt tolerance at increasing ion concentrations mentioned above.

DISCUSSION

Preinoculation exposure of tomato roots to even slightly saline (EC = 4 dS m^{-1}) solutions increased the severity of Phytophthora root rot. Although statistically significant, the magnitude of increase was not as great as has been reported (9,25,26,34), probably because we used lower salinity levels than used in other studies. Earlier researchers (9,25,26,34) employed salinity levels of 16–25 dS m^{-1} to show the effects of stresses that may be encountered in highly saline soils. Experiments described here were designed to show subtle effects of low salinity. In addition, we used a different tomato cultivar compared with earlier researchers (9,25,26,34) and assessed root rot severity after 14 rather than after 7–10 days (9,26,34).

Postinoculation salt stress (EC = 4 dS m^{-1}) reduced Phytophthora root rot caused by isolate S− of *P. parasitica* from nonsaline soil but not that caused by isolate S+ recently obtained from a saline soil. The reduction in root rot severity could be explained by the sensitivity of zoospores of *P. parasitica* to increasing ionic concentrations. Percentages of zoospores of either isolate that were encysted or lysed were significantly higher at 25 and 50 meq L^{-1} (2 and 4 dS m^{-1}) than at 0 and 2.5 meq L^{-1} (0 and 0.26 dS m^{-1}), but this effect was more pronounced for the isolate from nonsaline soil. Contrary to these results, Swiecki (34) observed an increase in Phytophthora root rot of tomato when salt stress ($NaCl + CaCl_2$ in a 5:1 equivalent ratio to a final EC of 16 dS m^{-1}) was applied to potted tomato seedlings during or after inoculation with *P. parasitica*. Production of sporangia of the isolate used in those experiments was stimulated in salt

solutions with EC levels ranging from 2 to 16 dS m^{-1} (Na:Ca = 1:1 [34]). However, zoospore release and motility were reduced at the same salt concentrations, and net zoospore activity in soil, as evidenced by infected tomato seedlings used as baits, was reduced at 7 compared with 0.2 dS m^{-1} (34). Reduced zoospore motility and increased encystment and lysis at increasing ion concentrations (including NaCl and CaCl at 1–30 meq L^{-1}) were also reported for *P. palmivora* and *P. cinnamomi* (7,11,14). The increase in root rot severity observed in previous studies (26,34), when high levels of salt stress were applied during or after inoculation, indicates that increased susceptibility of the plant and production of sporangia by the pathogen may have outweighed the reduction in zoospore viability. The level of salt stress used in these experiments (4 dS m^{-1}) may not have induced sufficient damage to the plant to compensate for the reduction in zoospore viability at that salt concentration.

When an isolate of *P. parasitica* from saline soil was exposed to salt solutions within 2 wk after isolation, it was relatively tolerant to ion concentrations of 25 and 50 meq L^{-1} (2 and 4 dS m^{-1}, respectively) compared with an isolate from nonsaline soil. However, after 2 mo in culture on CMA, the isolate from saline soil lost salt tolerance and induced less root rot in tomato plants exposed to salt stress (4 dS m^{-1}) than in unstressed plants. Differences in salt tolerance among isolates of *P. parasitica* were

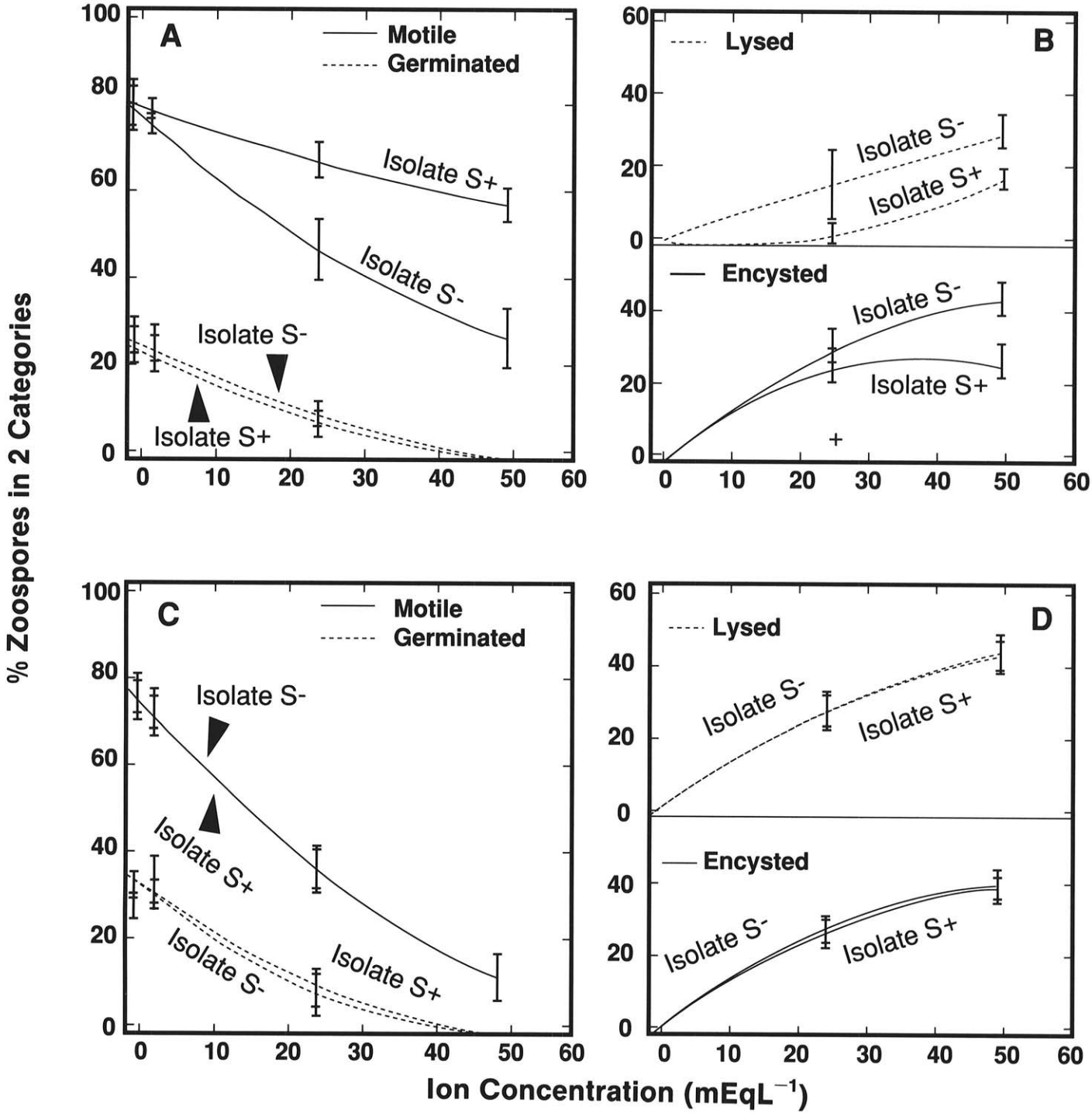

Fig. 3. Effect of ion concentration of a modified Hoagland's solution (Na:Ca ratio = 5:1) on percentage of zoospores of *Phytophthora parasitica* isolate S− from nonsaline soil, isolate S+ recently isolated from saline soil in the categories **A,** motile or germinated, and **B,** encysted or lysed, and of isolate S− and isolate S+ after 2 mo in culture in the categories **C,** motile or germinated, and **D,** encysted or lysed. Vertical bars denote standard deviations.

also reported by Blaker and MacDonald (8), but loss of salt tolerance in culture was not observed. However, the sources of the salt-tolerant isolates used in the two studies differed; the one used in these studies originated from a salinized experimental field infested 1 yr previously with four isolates from nonsaline soil, whereas the salt-tolerant isolate of Blaker and MacDonald (8) was isolated from a highly saline soil naturally infested with *P. parasitica*.

Previous studies on the effects of salinity on Phytophthora root rots were conducted with a fixed Na:Ca ratio (9,25,26,34). In the study under discussion, we have shown that the Na:Ca ratio in nutrient solutions may be as important in disease development as the total ion concentration. Root rot severity increased with increasing Na:Ca ratios at all ion concentrations when tomato seedlings were inoculated with an isolate of *P. parasitica* from saline soil but only at a low ion concentration (2.5 meq L^{-1}) with an isolate from nonsaline soil. A combination of salt stress (4 dS m^{-1}) and high Na:Ca ratios (5 or 10) applied during and after inoculation inhibited Phytophthora root rot induced by an isolate from nonsaline soil but not that by an isolate from saline soil. This was explained by a difference in sensitivity of these two isolates to high ion concentrations as well as high Na:Ca ratios. Zoospore viability of both isolates decreased at increasing Na:Ca ratios and ion concentration, but the decrease was less for the isolate from saline soil than for the isolate from nonsaline soil. The fact that the isolate from nonsaline soil induced more severe root rot at high Na:Ca ratios only when the overall ion concentration was low (2.5 meq L^{-1}) indicates that at 2.5 meq L^{-1}, the harmful effect of Na on the plant may have outweighed any harmful effect on the zoospores. This conclusion is substantiated by the observation that uninoculated plants had reddish brown rather than white roots at high Na:Ca ratios and that this browning was more pronounced when the overall ion concentration was low. This observation is in agreement with previous reports that Na is more harmful to the plant when the overall ionic concentration (including the Ca concentration) is relatively low (12,32,33).

Many physiological processes are altered in plants grown under

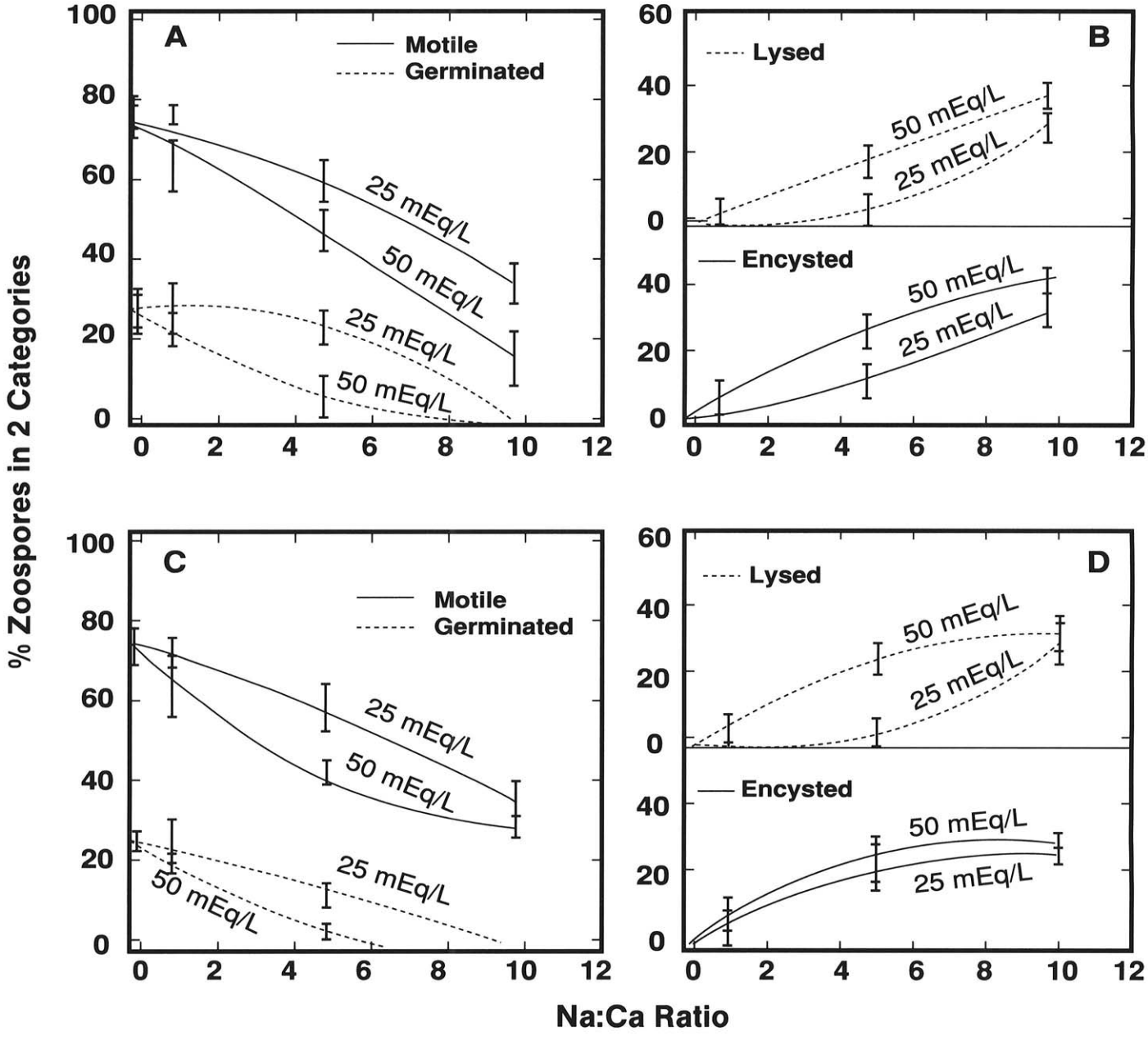

Fig. 4. Effect of sodium:calcium ratio of a modified Hoagland's solution at two ionic concentrations (25 and 50 meq L^{-1}) on percentages of zoospores of *Phytophthora parasitica* isolate S−, **A and B,** from nonsaline soil, and isolate S+, **C and D,** recently obtained from saline soil in the categories **A and C,** motile or germinated and **B and D,** encysted or lysed. Vertical bars denote standard deviations.

saline conditions, which may lead to a reduction in growth and yield (6). The effects of salinity have been attributed mainly to osmotic stress by some authors (6). However, from an extensive literature review, Läuchli and Epstein (20) concluded that specific ion effects (ion imbalances and specific ion toxicities) are at least as important as osmotic effects of salinity. Excess Na in the root environment can replace Ca in the pectic complexes of the middle lamella and cell wall (23). Reduction of Ca in pectic substances has been implicated in increased susceptibility to various soilborne pathogens that produce pectolytic enzymes (13,23). In addition, excess Na can impair selective permeability of plant membranes (12,19,24) because of the replacement of Ca by Na on the membranes (12), thereby increasing leakage of ions (and presumably organic carbon compounds) from salt-stressed roots (12). This physiological effect probably was responsible for the increased attachment of encysted zoospores of *P. cryptogea* to chrysanthemum roots exposed to salt stress before inoculation (25) and may have been responsible for the increased root rot by *P. parasitica* by preinoculation salt stress in our study.

Besides the effects on cell wall and plasma membrane, Na can disrupt the structural and functional integrity of intracellular membranes, enzymes, and the cytoskeleton (18). These effects can be moderated by Ca (18). Ca also affects isoprenoid metabolism, specifically sesquiterpenoid phytoalexin synthesis, in plants (36). Postinoculation salt stress (NaCl at 20 dS m^{-1} for 24 hr) of chrysanthemum roots delayed the cytological defense responses and enhanced colonization by *P. cryptogea* (35). The reduced resistance might have been associated with a reduction in phytoalexin production (26).

Based on our results with Phytophthora root rot of tomato and published reports on other host pathogen systems (32,33), we suggest that the effect of salt stress on disease development depends not only on the intensity of the stress but also on the ionic composition and the sensitivity of both host and pathogen to the resultant osmotic potential and specific ion activities.

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