Environmental Effects on Inoculum Quality of Dormant Rust Uredospores

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ABSTRACT

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Uredospores of *Puccinia graminis* f. sp. *tritici* and *P. recondita* f. sp. *tritici*, fungi which cause stem rust and leaf rust of wheat, respectively, were produced in the greenhouse on wheat seedlings (*Triticum aestivum* 'Little Club'). Newly harvested, mature, dormant uredospores immediately were subjected to specific light, temperature, and relative humidity (RH) treatments for 24 and 48 hr, then tested for germinability and infectivity. Compared with untreated spores, which consistently germinated >91% and which produced 60 ± 20 infections per leaf in standardized tests, germination and infectivity were reduced or eliminated by increased treatment temperatures, especially above 25 C. Germinability and especially infectivity further were reduced by exposure to 100% RH. Supplemental light reduced or eliminated the detrimental effects of high RH, but otherwise caused no measurable effect. Reductions in infections per leaf and in infection efficiency always were proportionally greater than were losses in germinability. The possible epidemiological significance of these findings is discussed.

Additional key words: spore germination, epidemiology.

Uredospores of wheat rust fungi are the principal inoculum that contributes to rust epidemics. Their production, dispersal, and germination, along with host penetration and infection comprise an important cycle of events in the development of rust diseases (Fig. 1). Each of these events is affected by environmental variables that can limit the rate and extent of disease development and, therefore, each is an important quantitative parameter in conceptual models of the initiation and progress of disease.

In nature, the rate of completion of the uredospore cycle is regulated by physical and biological variables; principally temperature, moisture, light, and the host-parasite relationship. The environmental variables that accompany and influence the various cycle events have been studied extensively (1,3, 5,7,9–14,17). Attention also has been given to environments that reduce uredospore viability or regulate their subsequent performance (5,7,9,10,15–17). For example, hydration (exposure of dormant spores to humid atmospheres) promotes the subsequent germination of old spores (7,9,14–17) but induces a light-sensitive inhibition of the germination of newly produced spores (15–17). Evidence also is accumulating which suggessts that the performance of spores as inoculum may reflect the specific host-parasite relationship that regulated their production (M. V. Wiese, *unpublished* and W. A. Shipton, James Cook Univ., North Queensland, Australia, *personal communication*). Such studies demonstrate environmental variables that regulate subsequent uredospore performance and, perhaps, the resultant epidemic.

Disease cannot occur unless some portion of the inoculum population is infectious. However, uredospore inoculum, a parameter of prime importance to disease development, normally is described only in quantitative terms (2,4,6,8). The inoculum quality of the spores is either assumed to be constant or unknown.

In earlier studies, the germinability of mature dormant uredospores exposed to various temperature, light, and RH regimes has been described (15–17). The objective of the present study was to measure the influence of the same or similar postproduction-pregermination environments on the performance of uredospores as inoculum (Fig. 1). Such assessments of inoculum quality could be used to improve conceptual models of disease development that presently only account for inoculum quantity (2,4,6,8).

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MATERIALS AND METHODS

Procedures previously used for studies of germinability (15–17) were extended to incorporate measurements of infectivity. Uredospores of stem rust (*Puccinia graminis* f. sp. *tritici*) race 56 and leaf rust (*P. recondita* f. sp. *tritici*) race 2 were produced on seedlings of wheat (*Triticum aestivum* L. 'Little Club'). Newly produced spores were harvested and immediately separated into 3 to 5 ± 0.01-mg lots. The conditions for spore treatment were as described (15,16) except that the weighed spore lots were dispersed on 10 × 10-mm glass planchets and incandescent light was unfiltered and fixed at 22,630 lx.

Measurements of spore performance were made at the time of spore collection and after treatment. In all cases, the germinability and infectivity of treated spores was compared with that of untreated spores from the same collection held at ambient RH at 4 C in the dark for the duration of the treatment period (15–17).

After environmental treatment, each spore lot was added to Mobilsol 100 (13), and the suspension was adjusted to 1 mg spores per milliliter. One-tenth milliliter aliquots of this suspension were quantitatively atomized onto groups of 10 first leaves of wheat seedlings and onto duplicate strips of newly-prepared 1% water agar.

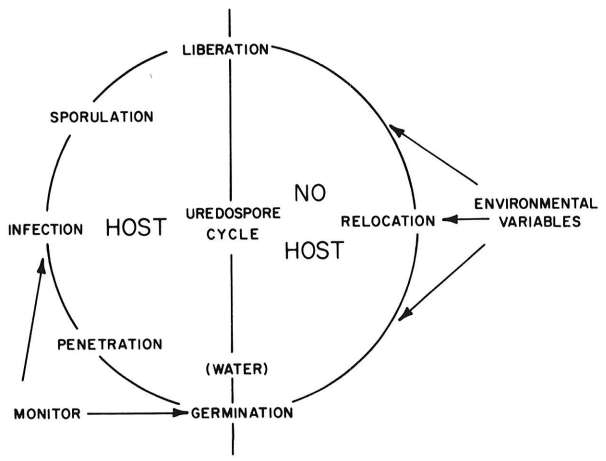

Fig. 1. Repeating cycle of uredospore inoculum in a cereal rust epidemic.

Inoculation procedures and conditions for incubating inoculated plants were as described by Rowell and Olein (13). The techniques ensured the consistent production of 60 ± 20 infections per leaf by untreated control spores. Infectivity of all spore lots was measured as infections per first leaf based on counts for a minimum of 20 leaves.

To increase the prospect of detecting changes in infectivity due to environmental treatment, seedlings of two susceptible (but genetically different) assay hosts were employed. Seedlings of the wheat cultivars Ionia and Little Club selected for uniform size and maturity of first leaves, were inoculated with each spore suspension. Resultant infections were counted 12–14 days after inoculation.

For germination measurements, each spore suspension was atomized onto 1% water agar on 20 × 50-mm and 5 × 50-mm glass strips. By placing the 5-mm strips upright between seedling leaves during inoculation, estimates of the density of the spore deposit on leaves were obtained for use in calculations of infection efficiency. Percent germination was calculated from counts of a minimum of 100 spores per glass strip. Only spores with germ tubes equal to or exceeding the lengths of the uredospore after 20-hr of incubation in the dark at 21 C were considered germinated. Under these standard conditions, all lots of control spores germinated >91%. All data presented herein (Fig. 2, 3, and Table 1) are grand means from three separate experiments using spores treated for 24 hr and assayed on water agar and on Little Club seedlings.

RESULTS

As with earlier studies (15,16), all changes in uredospore performance were related directly to the duration of treatment. Treatment for 48 hr, for example, merely accentuated the behavioral changes induced by 24-hr treatment (15). Although plants of cultivar Little Club often supported more infections per leaf than those of Ionia, the two cultivars were approximately equivalent assay hosts for measuring the performance of treated and untreated uredospores of both rust species. In many instances, the infectivity of treated and untreated spore lots differed markedly (Fig. 2, 3), but total infections per leaf on Little Club and Ionia seedlings within each test did not differ significantly ($P = 0.05$).

All reductions in germinability were accompanied by reductions in infectivity. No treatment significantly enhanced germinability, infectivity, or infection efficiency relative to untreated spores (Fig. 2, 3 and Table 1). In all cases, infectivity was reduced more than could be explained by reductions in germinability alone. Dark-hydrated stem rust uredospores, for example, were not infectious although up to 40% were germinable (Fig. 2).

Temperature. In general, the germinability and infectivity of *P. graminis* and *P. recondita* uredospores were reduced as treatment temperature increased above 20 C (Fig. 2, 3). In this regard,

TABLE 1. Infection efficiency[a] of uredospores of *Puccinia graminis* f. sp. *tritici* subjected to moisture and light treatments for 24 hr at 25 C

		Dark		Light[e]	
	Ck[b]	Hydrated[c]	Nonhydrated[d]	Hydrated	Nonhydrated
All spores	19	<0.1	18	6	16
Germinated spores only	25	<0.1	28	8	20

[a] Infection efficiency = (infections per cm^2 of leaf)/(spores per cm^2 of leaf) × 100.
[b] Untreated spores were held in the dark at 4 C for 24 hr.
[c] Water-saturated atmosphere (100% RH).
[d] Ambient atmosphere (25 ± 5% RH).
[e] Incandescent illumination at 22,630 lx.

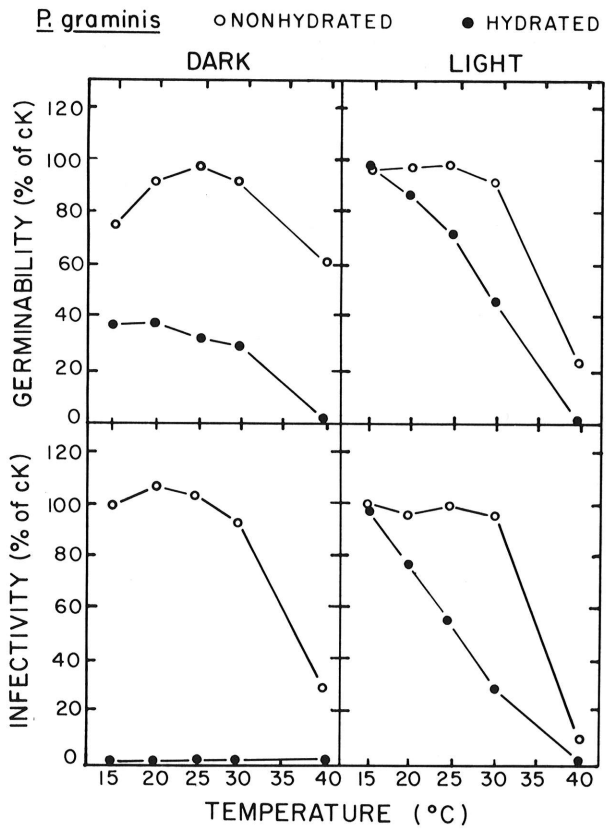

Fig. 2. The germinability and infectivity of uredospores of *Puccinia graminis* f. sp. *tritici* following 24-hr treatment at different temperatures in atmospheres of 25 ± 5 (nonhydrated) and 100% relative humidity (hydrated) in the dark and light (22,630 lx). Untreated spores (ck) germinated >91% and produced 60 ± 20 infections per leaf.

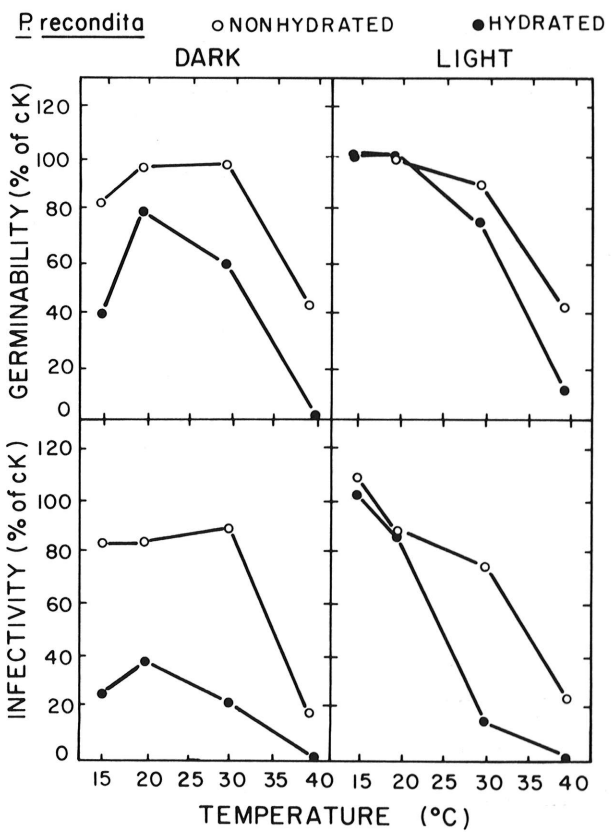

Fig. 3. The germinability and infectivity of uredospores of *Puccinia recondita* f. sp. *tritici* following 24-hr treatment at different temperatures in atmospheres of 25 ± 5 (nonhydrated) and 100% relative humidity (hydrated) in the dark and light (22,630 lx). Untreated spores (ck) germinated >91% and produced 60 ± 20 infections per leaf.

temperatures >30 C were especially detrimental. Treatment at 40 C for 24 hr, for example, routinely reduced germination to less than 50% and prevented infection.

Moisture. Adding moisture to the treatment regimes consistently was detrimental (15–17). The germinability, and especially the infectivity, of spores held in water-saturated atmospheres (hydrated) was markedly reduced relative to that of spores held at the same temperatures in ambient atmospheres (nonhydrated). At all treatment temperatures, dark-hydrated spores normally had less than half the germination and infection potential of comparable nonhydrated spores. Dark-hydration was more detrimental to stem rust uredospores than to leaf rust uredospores, rendering them noninfectious at all treatment temperatures (Fig.2).

Light. Light added to the treatment regimes was influential only in water-saturated atmospheres. By reducing the detrimental effects of hydration, it maintained the germinability and infectivity of uredospores of both rust fungi near the level of nonhydrated spores (Fig. 2,3).

DISCUSSION

Except for the greater loss of infectivity among dark-hydrated stem rust spores, uredospores of *P. graminis* and *P. recondita* responded similarly to environmental treatment (Fig. 2,3). Their performance was inversely related to the levels of temperature and RH to which they previously were exposed. Light, already known to maintain spore germinability by counteracting the detrimental effects of hydration (15–17), also acts to maintain infectivity. The mechanisms for these light reactions appear to be metabolic, but are largely unexplored. Light does not appear to act by influencing water uptake since dark- and light-treated hydrating spores increase in weight at nearly equivalent rates (15).

All environments detrimental to germination percentage (especially 100% RH) were preferentially damaging to infectivity (see data for *P. graminis* in Fig. 2). Thus, the phenomena of germ tube elongation and host infection, which are affected differentially by environmental treatment, differ physiologically as well as morphologically and chronologically. Since a significant proportion of the spores with capacity to germinate were noninfectious (Fig. 2,3 and Table 1), routine assessments of uredospore germinability cannot be assumed to reflect their infectivity especially when light is limiting.

Micrometrological conditions to which uredospores are exposed in the field are similar to those evaluated herein and may have similar profound effects on subsequent spore performance as inoculum. Knowledge only of numbers of spores in the field could be misleading or unnecessarily alarming since local environmental conditions may render all or significant portions of the inoculum noninfectious. The progress of rust epidemics perhaps should be examined for possible inverse relationships to temperature and RH and direct relationships to light intensity.

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