

Effect of ridge gourd pollen on zoospore germination of *Pseudoperonospora cubensis* and its significance in epidemiology

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Abstract. Ridge gourd pollen has a stimulatory effect on the germination of *Pseudoperonospora cubensis*. The rate and percentage germination of zoospores increased in the presence of pollen leachates. Spraying of leaves with a mixture of pollen and sporangial suspension enhanced the development of lesions. Early germination of zoospores in the presence of pollen proved advantageous for infection as it provided a prolonged favourable infection period. The results are discussed in relation to the epiphytotics of the disease during flowering period.

Keywords. *Pseudoperonospora cubensis*; ridge gourd; pollen effect; zoospore germination; epidemiology.

1. Introduction

Pseudoperonospora cubensis (Berk. and Curt.) Rostow, the incitant of cucurbit downy mildew is one of the serious and production limiting diseases of *Luffa acutangula* Roxb. in India. The disease attains serious proportions when the plants start flowering and as a result the susceptible varieties of plants fail to produce fruits. Bains and Jhooty (1975) reported that in Cantaloupes downy mildew appeared during 1972-74, under field conditions only during the flowering and fruiting stage. The effect of host pollen on stimulation of spore germination of fungal pathogens has been worked out in different crops (Chou and Preece 1968; Fokkema 1976; Preece 1976; Meenakshi and Ramalingam 1979; Suryanarayana and Ramalingam 1979). So far no report on the effect of host pollen on germination of downy mildew pathogens has been made.

2. Materials and methods

2.1. *In vitro* effect of pollen on zoospore germination

One of the susceptible varieties of ridge gourd (Pusa Nasdar) was grown in the downy mildew experimental plots to obtain the sporangial inoculum and the host

pollen. Sporangial suspension was prepared by the following method: downy mildew infected leaves were collected at 6 p.m. and the remnants of the downy growth was washed off with moist cotton under running tap water. The leaves were air dried and small bits of leaves with lesions were cut and placed inside petri plates containing a wet blotter with the adaxial surface of the leaf in contact with the blotter. A good crop of sporangia was obtained after incubating the leaf bits for 12 hrs at 22° C in dark. The sporangia were scraped with a blade into a dish containing distilled water. The concentration of sporangial suspension was measured using a haemocytometer and was adjusted to about 10,000/ml. Host pollen was collected and stored at 5° C. Five mg of the pollen was mixed in 10 ml of the sporangial suspension. A suspension of the mixture was placed on glass slides and incubated in moist chambers at room temperature (22–26° C). In controls no pollen was added. Observations for zoospore germination were made under binocular microscope and results were recorded at hourly intervals after fourth hour.

2.2. *Effect of pollen on infection of host leaves and lesion development*

Pollen plus sporangial suspension was sprayed on the lower surface of the leaf of 20–30 day old plants and retained inside a glass house. The leaves were covered with moist polythene bags for about 24 hrs and observations for number and size of lesions were made. The effect of pollen in reducing the infection threshold was tested for moisture requirement by covering the leaves with polythene bags for 3, 4, 5 and 6 hrs. After removing the polythene bags the leaf was air dried and left inside the glass house.

2.3. *Disease development in the field in relation to age of the crop*

Two varieties of ridge gourd viz. Pusa Nasdar, a highly susceptible variety and long variety which is moderately resistant were sown in the month of August 1980 in plots. Disease rating was made at weekly intervals using a 0 to 5 scale as described by Thomas (1977). Fertiliser (NPK 17 : 17 : 17) was applied twice at the age of 20 days and 45 days.

3. Observations

3.1. *In vitro effect of pollen on zoospore germination*

Sporangial suspension when incubated at room temperature (22–26° C) released zoospores within 1½–2 hrs. Maximum number of zoospores were observed after 2 hrs of incubation. Zoospores remained active in water for 90 to 120 min and then encysted. The data with regard to percentage of zoospore germination and germ tube length with and without pollen are recorded in figures 1 and 2 respectively. Zoospore germination started an hour earlier in the presence of pollen. The percentage of zoospore germination and vigour of the germ tubes in the presence of pollen was greater compared to the zoospore which germinated in the absence of pollen.

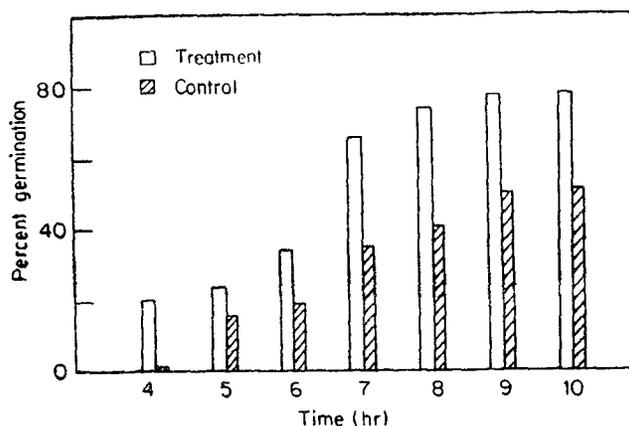


Figure 1. Effect of host pollen on the germination of zoospores of *P. cubensis*.

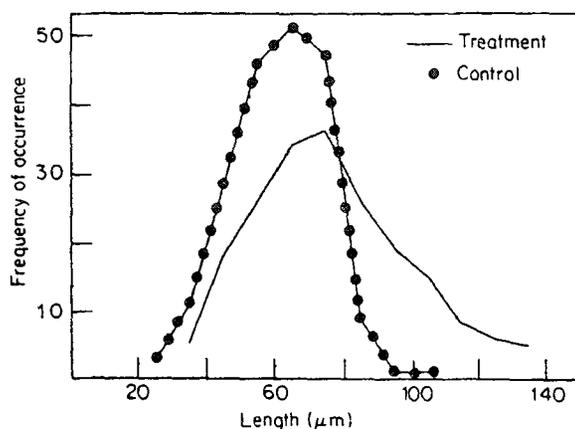


Figure 2. Comparison of the germ tube length of zoospores in presence of pollen and in distilled water and their relative frequency of occurrence.

3.2. Effect of pollen on infection of host leaves and lesion development

Difference in disease reaction was apparent in plants sprayed with the mixture and the sporangial suspension alone. The number and size of lesions were more on leaves sprayed with the pollen mixture. Leaves inoculated with sporangial suspension required a minimum of four hrs of leaf wetness for successful infection under glass house conditions. Only 3 hrs of leaf wetness was needed for infection in the presence of pollen under the same conditions of temperature and inoculum concentration.

3.3. Disease development in the field in relation to age of the crop

Downy mildew of ridge gourd makes its appearance at the seedling stage. The young true leaves are resistant to downy mildew. When the seedlings attain the

age of 20 days (3–5 true leaf stage) symptoms appear on the true leaves as greenish to yellow lesions. Disease appearance in ‘Long’ is delayed by a week in comparison with Pusa Nasdar. Severity of disease in relation to age is plotted in figure 3. Flowering in both the varieties start 35–40 days after planting. Till then disease severity is less than stage 3 of the 0–5 scale. Soon after flowering the disease reaches severe proportions and it ultimately results in the death of vines in ‘Pusa Nasdar’ whereas in ‘Long’ it is not very severe and the vines continue to grow but the yield is significantly reduced.

4. Discussion

In saprophytic fungi and facultative pathogens the stimulating effect of pollen on germination of conidia is attributed to carbohydrates (Suryanarayana and Ramalingam 1979 ; Fokkema 1976) but the aggressiveness of such fungi depends on the pollen leachates, rather than the nutrients (Chou and Preece 1968). In *P. cubensis* the zoospores germinate in distilled water thereby showing that it is not nutrient dependent. Hence it is quite probable that pollen leachates provide a stimulatory effect on zoospore germination.

Preece (1976) stated that the effect of pollen on leaf infection may be due to (a) increase in the speed and rate of spore germination, (b) restoring the germinability and infectivity of old spores and (c) reducing the infection threshold by enhancing the speed and virulence of the pathogen. From the present study, it is evident that there is an increase in the number of zoospores germinating and the vigour of germination is enhanced. Under field conditions, sporangial liberation occurs during morning hours (Cohen and Rotem 1971 ; Thomas 1977 ; Bains and Jhooty 1978). It starts at around 6 a.m. and reaches a peak at 8 a.m. For the successful infection of a fresh host leaf a minimum of 4 hrs of leaf wetness is needed. But when the zoospore germinates in presence of host pollen it needs only a period of 3 hrs for infection.

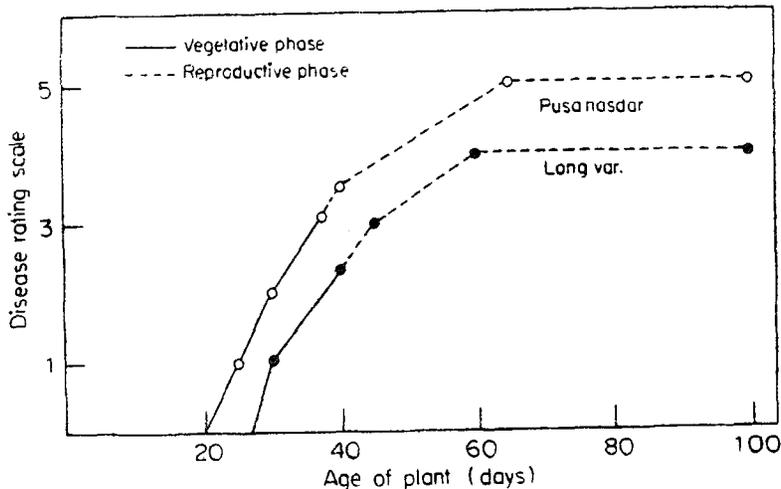


Figure 3. Disease progress in two varieties of ridge gourd in relation to age of the plant.

The sporangia liberated during morning hrs are subjected to a period of unfavourable conditions which last until the next dew fall and infection of host plant occurs during night hrs (Cohen and Rotem 1971 ; Cohen and Eyal 1980). From our experiments under Mysore conditions, it is quite probable that those sporangia liberated early in the morning get deposited on the leaves and they start germinating by the production of zoospores. As dew persists at Mysore condition till 9·30 to 10 a.m., infection of the leaf tissue in presence of host pollen can occur successfully during the daytime. In addition, those sporangia which are deposited in later hours of the day, *i.e.*, those sporangia which fail to infect during the morning hours due to the advent of unfavourable period for infection, survive till the next dew fall with a considerable loss in viability of sufficient number of sporangia. Those viable sporangia germinate and cause infection during night hours. As a result, severity of the disease increases significantly during flowering period. This is supported by the results of studies of lesion development and zoospore infection under different leaf wetness periods.

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