Response of *Fusarium solani* to Fluctuating Temperatures

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AS PROGRAMMED incubators have become available, several researchers have attempted to evaluate the effect of temperature and temperature change on the growth of fungi (*1, 4, 7*). Conflicting conclusions have been reported about whether temperature fluctuation itself has an effect on fungus growth. This point must be clarified before the question of whether growth of an organism in a fluctuating temperature regime can be predicted from growth data collected in constant temperature studies.

The purpose of our study was to measure growth under a range of constant temperatures and under a series of fluctuating temperature regimes, and to determine if growth in the fluctuating temperature regimes could be predicted satisfactorily from the growth data collected in the constant temperature experiments. Growth was measured on both agar and liquid culture to determine if the reported differences were due to substrate instead of temperature treatments.

MATERIALS AND METHODS

Fusarium solani (Mart.) App. & Wr. emend Synd. & Hans. was grown in an incubator controlled by a cam-type temperature programmer for both the constant and fluctuating temperature treatments. For the fluctuating temperature experiments, the programmer was equipped with a cam that caused the temperature in the incubator to change in a linear manner from one temperature extreme to the other and back again during a 24-hour period. The constant temperature treatments were 15, 20, 25, 30, and 35°C. The fluctuating temperature treatments were 25 ± 3, 25 ± 6, 25 ± 9, 30 ± 3, 30 ± 6, and 30 ± 9°C.

Six isolates of *F. solani* were grown at each temperature treatment. Five of the isolates, F1M, F2P, F3Y, F5P2, and F6P3, were collected from cankers on hardwood trees; the sixth, F4S, was isolated from soil. F4S may also be parasitic on hardwood trees, even though it was isolated from soil.

Liquid and agar cultures were used in all experiments. The liquid medium contained: sucrose, 10 g.; $NH_4H_2PO_4$, 2 g.; KH_2PO_4, 0.6 g.; K_2HPO_4, 0.4 g.; $MgSO_4 \cdot 7H_2O$, 0.5 g.; $FeC_6H_5O_7 \cdot 5H_2O$, 10.0 mg. (6); $ZnSO_4 \cdot 7H_2O$, 4.4 mg.; $MnSO_4 \cdot 4H_2O$, 5.0 mg.; $CaCl_2$, 55.0 mg.; $CoCl_2 \cdot 6H_2O$, 1.0 mg.; and distilled water to 1 liter. Fifty ml. of medium was placed in each 250-ml. Erlenmeyer flask, sterilized at 121°C. for 20 minutes and inoculated with 0.5 ml. of a spore suspension that contained approximately 2 million spores per ml. After 7 days in the incubator, the mycelium was collected on tared filter paper, washed with 200 ml. of distilled water, and dried at 100°C. for 24 hours; and dry weight was determined.

Potato dextrose agar was used as the growth medium in the agar culture experiments. These cultures were inoculated by inverting a plug, cut with a No. 3 cork borer from an actively growing culture, in the center of the petri plate. After 5 days in the incubator, the mean diameter of each culture was determined by measuring the culture diameter at two locations at right angles to each other.

All treatment combinations were repeated twice. Four replications were included each time for both the liquid and agar cultures.

RESULTS

The optimum constant temperature for growth of all the isolates in liquid culture — except F5P2 — was 25°C. or lower; the optimum constant temperature for all isolates in agar culture was 25°C. or higher (fig. 1). Larger dry-weight differences were found among the isolates in liquid culture than in agar culture.

In liquid culture, a 6°C. fluctuation around a mean of 25°C. caused a reduction in growth of three isolates of *F. solani* and an increase of two isolates (fig. 2), but fluctuations of 12 and 18°C. caused a reduction in growth of all isolates. Fluctuation ranges of 6 and 12°C. around a mean of 25°C. had very little effect on growth of any of the isolates in agar culture, but an 18°C. fluctuation caused a slight reduction in growth of all the isolates.

Growth of only one isolate, F2P, was reduced with a 6°C.

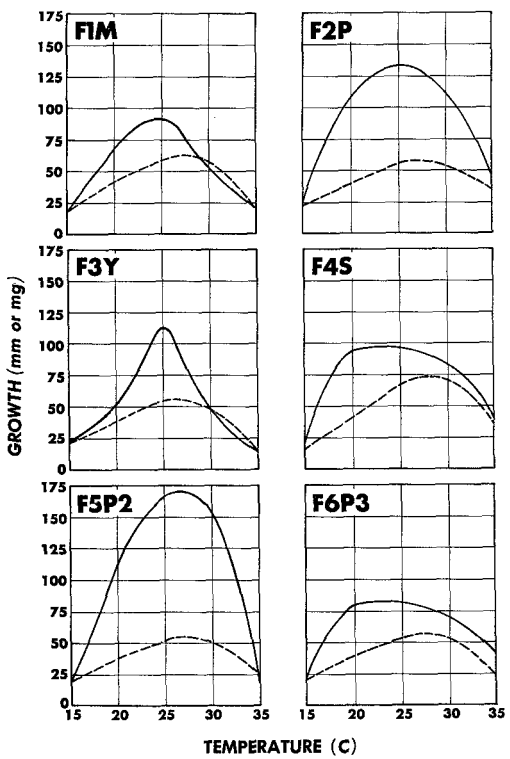

Figure 1. — Effect of constant temperature on growth of *F. solani* isolates in liquid culture (—) and agar culture (– –). Measurement of growth in liquid culture is in mg., and measurement of growth in agar culture is in mm.

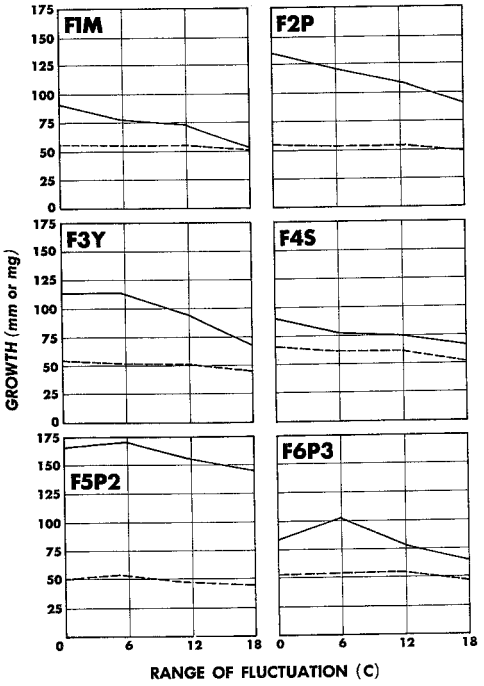

Figure 2.—Effect of fluctuating temperature around a mean of 25°C. on growth of isolates of *F. solani* in liquid culture (—) and agar culture (– –). Measurement of growth in liquid culture is in mg., and measurement of growth in agar culture is in mm.

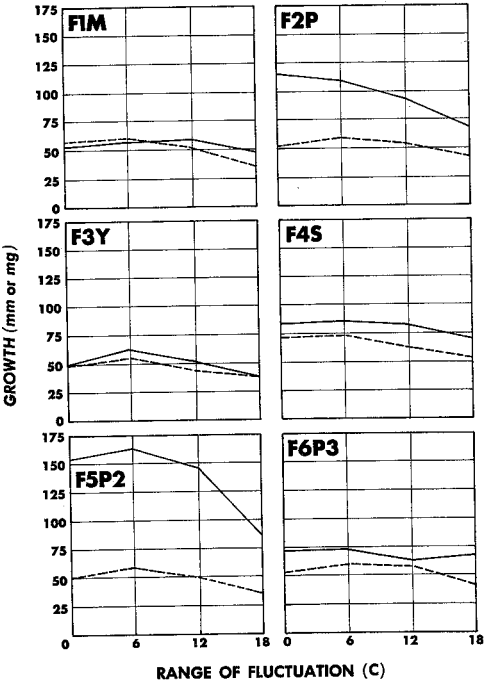

Figure 3.—Effect of fluctuating temperature around a mean of 30°C. on growth of isolates of *F. solani* in liquid culture (—) and agar culture (– –). Measurement of growth in liquid culture is in mg., and measurement of growth in agar culture is in mm.

fluctuation around a mean of 30°C. (fig. 3) in liquid culture, but no reduction was found in any of the isolates in agar culture. A 12°C. fluctuation caused a large reduction in growth of F2P in liquid culture and a small reduction for all isolates in agar culture — except F2P and F6P3 — compared to growth at 30°C. An 18°C. fluctuation caused a reduction in growth of all isolates in agar culture and of all isolates, except F6P3, in liquid culture.

DISCUSSION

To determine if fluctuating temperature itself affects growth of the fungus, growth under fluctuating temperature regimes was compared with growth under a constant temperature regime at the midpoint of the fluctuation range and with the shape of the constant temperature growth curve over the fluctuation range.

If fluctuating temperature does not affect growth, growth under the fluctuating temperature regime will be equal to growth under the constant temperature regime when the constant temperature growth curve over the range of fluctuation is linear; growth under the fluctuating temperature regime will be less than growth under the constant temperature regime when the constant temperature growth curve over the range of fluctuation curves downward; growth under the fluctuating temperature regime will be greater than growth under the constant temperature regime when the constant temperature growth curve over the range of fluctuation curves upward.

From an inspection of the constant temperature growth curves of *F. solani* in liquid culture around 25°C., growth under all the fluctuating temperature regimes with a midpoint of 25°C. should be less than growth at a constant temperature of 25°C. However, growth of three isolates either remained the same or increased with a 6°C. fluctuation range. From an examination of the constant temperature growth curve for liquid culture around 30°C., only two isolates, F1M and F3Y, should increase in growth in a fluctuating temperature regime. However, growth of all but one isolate, F2P, increased or remained the same with a small range of fluctuation.

In agar culture, growth in all the fluctuating temperature regimes with a midpoint of 25°C. was very close to growth in a 25°C. constant temperature regime. Growth in agar culture in fluctuating temperature regimes with a midpoint of 30°C. was less than growth in a 30°C. constant temperature regime only at the wide ranges of fluctuation. From an inspection of the constant temperature growth curves for agar cultures around 25 and 30°C., growth in all fluctuating temperature regimes would have been expected to be less than growth at the midpoint of the fluctuation range. This is especially true for the 18°C. fluctuation range.

These increases in growth of *F. solani* in fluctuating temperature regime that are not predicted from constant temperature growth suggest that temperature fluctuation itself affects the growth of some fungi. This conclusion is not in agreement with the conclusion reached by Burgess and Griffin (*1*) who proposed a model for predicting growth of fungi under fluctuating temperature regimes from constant temperature growth data, based on the assumption that temperature change itself does not affect the growth rate of fungi. These growth data were collected from agar cultures, but an analysis of our data suggests that growth, both in agar and liquid culture, may be affected by fluctuating temperatures.

The increase in growth of *F. solani* and decay fungi (*4*) with temperature fluctuations may be due to a carryover effect from more suitable to less suitable temperature or a direct effect of temperature on the organism. A positive carryover effect would occur if some temperature-sensitive system within the fungi would continue to function briefly at the high level obtained at a suitable temperature for a short period after the suitable temperature had passed. However, if this can occur, a negative carryover effect from less suitable to more suitable temperature may also occur. Burgess and Griffin (*1*) stated that detrimental effects of high temperature may have persisted and affected later growth at lower temperature, and that this may be the reason for the failure of their model at extreme temperatures.

A direct effect of temperature fluctuation on higher plants has

been reported. Kramer (*5*) and Hellmers (*3*) found that certain tree species made the best growth under the largest day-night differentials. Therefore, some fungi may also grow more if they are exposed to fluctuating temperatures as physiological processes within fungi each have a temperature optimum (*2*).

In this study we also compared growth in agar cultures and in liquid cultures. Two situations are evident: (1) the optimum temperature depends on the growth substrate, and (2) dry-weight measurements in liquid cultures give a larger range of differences than do diameter measurements in agar culture. Because of this larger range of differences, growth in liquid cultures is more sensitive than growth in agar cultures for measuring the effect of temperature treatments on growth of fungi. Also, in liquid culture all the growth is measured, whereas in agar culture only the horizontal spread is measured.

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