Photo-reactivation of *Botrytis fabae* Sardiña measured by a Local-lesion Technique

When leaves of broad bean (*Vicia faba* L.) are sprayed with suspensions of spores of *Botrytis fabae* Sardiña they develop chocolate-coloured lesions[1]. By rubbing suitably diluted spore suspensions over the upper surfaces of leaves of the variety Seville Longpod with the forefinger, and then keeping the inoculated plants at about 20° C. in a saturated atmosphere, we find that the discrete lesions are uniformly distributed and, after 24 hr., can be easily counted. The lesions provide an assay method comparable with counting the local lesions that are produced in plants by many viruses, and we have used them to measure how ultra-violet radiation affects the infective capacity of the spores.

Ultra-violet radiation, more than 95 per cent of wave-length 2537 A., was provided by a Hanovia XII lamp. Spore suspensions (0·3 ml. samples contained in cavity slides) were exposed at 40 cm. from the lamp, where the radiation intensity was 173 μW. per sq. cm. After exposure for various times, the irradiated suspensions were inoculated to halves of bean leaves, the opposite halves of which were inoculated with unirradiated spores, and the plants were then placed in a saturated atmosphere and kept in darkness for 24 hr. In these conditions, about 90 per cent of the original infectivity of the spore suspensions was destroyed by exposure for 3½ min. to the radiation. The spores of *B. fabae* are multinucleate and the rate of inactivation did not correspond to a first-order reaction (that is, it is a so-called 'multiple-hit' curve).

In studying the rate of inactivation, inoculated plants were kept in the dark because the effects of ultra-violet radiation on some biological systems can be mitigated if the systems are later exposed to visible light, a phenomenon that has been called photo-reactivation[2]. For example, when irradiated suspensions of bacteria or some fungi are cultured on agar in the light, more grow than if the cultures are kept dark. Similarly, irradiated preparations of some viruses[3,4] can be photo-reactivated; but with these the phenomenon does not occur if the viruses are exposed to light *in vitro*, but only when infected host cells are illuminated. No previous tests seem to have been made to see whether the pathogenicity of irradiated fungi is influenced by visible light, and we have therefore made some with *B. fabae*. Table 1 records the results of two experiments in which spores were irradiated for 3½ min., and then exposed to light for 0, 2, 4 or 8 hr., before all lots were inoculated to plants 8 hr. after irradiation. Plants with four sets of paired leaves were inoculated, and the different treatments were arranged on half-leaves in a 4 × 4 latin square design. Half the inoculated plants were then placed in daylight and half in darkness.

The half-leaves inoculated with spores exposed to visible light produced more lesions than those inoculated with spores kept for 8 hr. in the dark, and the magnitude of the difference usually increased with increases in the length of time the spores were exposed to light. Regardless of the time of exposure to light before inoculation to plants, plants kept in daylight after inoculation always produced significantly more lesions ($P > 0{\cdot}01$) than those kept in darkness (for statistical analyses[5], the numbers of lesions, n, were transformed to log $(n + 10)$). As

Table 1. Effects of Different Lengths of Daylight on the Infectivity of Ultra-violet Irradiated Spore Suspensions of *Botrytis fabae* (Sardina)

Post-inoculation treatment (hr. in daylight after ultra-violet treatment)	Mean lesion count/half-leaf				
	0	2	4	8	Mean
Exp. 25					
Plant in darkness	20	105	204	185	100
Plant in daylight	214	426	407	480	362
Exp. 27					
Plant in darkness	25	46	105	194	93
Plant in daylight	50	145	241	397	208

plants inoculated with unirradiated spore suspensions produce the same numbers of lesions whether they are kept in the light or dark, this increase with irradiated suspensions when inoculated plants were in the light is again attributable to photo-reactivation. The results show that some spores can still be photo-reactivated after 8 hr. in darkness, and they suggest that spores on the leaves of a host respond more readily to light than when they are *in vitro*, for the inoculated plants were in daylight for only about three hours.

As with other biological systems, the amount of photo-reactivation increases with increasing amounts of exposure to ultra-violet radiation. After exposure for 4½ min., the ratio of numbers of lesions on plants in the light to those in dark was 1½ times as great as after exposures for 3½ min.

F. T. Last
E. W. Buxton

Rothamsted Experimental Station,
Harpenden,
Herts.
May 14.

[1] Sardiña, J. R., *Mem. Soc. Esp. Hist. Nat.*, **15**, 291 (1929).
[2] Kelner, A., *J. Bact.*, **58**, 511 (1949).
[3] Dulbecco, R., *J. Bact.*, **59**, 329 (1950).
[4] Bawden, F. C., and Kleczkowski, A., *J. Gen. Microbiol.*, **8**, 145 (1953).
[5] Kleczkowski, A., *Ann. App. Biol.*, **36**, 139 (1949).

Physiological Races of *Puccinia polysora* Underw.

Our early studies on the resistance of maize seedlings to this rust in East Africa led to the hypothesis that a dominant major gene controls the hypersensitive type of resistance[1]. During the past two years, we have bred many lines of maize that are pure for this gene, by selfing resistant plants selected from Central American types and by selfing first crosses between resistant Central American types and East African susceptible types. In seedling-inoculation experiments, variation occurs in the manifestation of the hypersensitive reaction, from necrotic lesions without development of sori (up to eighteen days) to necrotic lesions with well-developed small sori. On a system of recording based on, but modified from, that employed by the wheat breeders (see ref. 2), we have classed the former as 01 (no sori) and the latter as 1 (small sori), the fully susceptible reaction, with large sori and little or no surrounding chlorosis, being class 4.

From the start of this work we had in mind the possibility that more than one physiological race of *P. polysora* might exist in East Africa. Collections of uredospores were made in three separate regions—