RECOVERY FROM THE BLUE-LIGHT INHIBITION OF SPORULATION IN *BOTRYTIS CINEREA*

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(With 4 Text-figures)

Sporulation in *Botrytis cinerea* Pers. ex Fr. is potentiated by black light. This potentiation is reversed by blue light, and the reversal can be nullified by 15–30 min near-ultraviolet (70 μW/cm²), or more than 6 h continuous far-red (1800 μW/cm²) irradiation. The time course of sporulation for such treated cultures was no different from that of cultures kept in darkness after photo-induction by black light, but an intervening dark period between blue irradiation and subsequent irradiation reduced the ‘re-promotive power’ of near-ultraviolet or far-red light.

Potentiation of sporulation by near-ultraviolet is reversed by subsequent irradiation with blue light in *Helminthosporium oryzae* V. Breda de Haan (Honda & Sakamoto, 1968), *Alternaria tomato* (Cke.) Weber (Kumagai & Oda, 1969), and *Botrytis cinerea* Pers. ex Fr. (Tan & Epton, 1974; Tan, 1974a, b). This reversal of potentiated sporulation in turn can be nullified by subsequent near-ultraviolet irradiation (Honda, Sakamoto & Oda, 1968; Kumagai & Oda, 1969; Tan & Epton, 1974; Tan, 1974a, b). But in *Alternaria solani* (Ell. & G. Martin) L. R. Jones & Grout red light was found to reverse this inhibition of sporulation (Lukens, 1965). It was thus of interest to study the effects of various types of irradiation on the re-promotion of sporulation in *B. cinerea*.

MATERIALS AND METHODS

The same isolate of *Botrytis cinerea* Pers. ex Fr. as in Tan & Epton (1973) was used.

The general cultural procedures have already been described (Tan & Epton, 1973, 1974). The spore suspension for inoculation was prepared with sterile distilled H₂O instead of diluted Tween 80. Cultures were incubated in the dark for 4½–5 days at 20 ± 1° before irradiation at 21 ± 1°.

The light sources and filters used for irradiation experiments were as described previously (Tan & Epton, 1973, 1974). Near ultraviolet light was obtained using Ilford No. 828 under black light fluorescent tubes, blue light obtained using Ilford No. 622 under blue fluorescent tubes, yellow and red light obtained using Ilford No. 626 and No. 608, respectively.

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under white fluorescent tubes, and far-red light obtained using Ilford No. 207 under incandescent lamps.

Irradiances at culture level, measured as described by Tan & Epton (1973), were: near ultraviolet, 70 μW/cm²; blue, 250 μW/cm²; yellow, 143 μW/cm²; red, 150 μW/cm²; far-red, 1800 μW/cm²; black light, 151 μW/cm².

The experimental schedule outlined below was employed in all the experiments described unless otherwise stated.

![Diagram showing the schedule of irradiation and dark periods.](image)

It was previously found that blue light was most effective when given at the 12th hour after the end of photoinduction and that 4 h exposure resulted in about 35-40% inhibition (Tan, 1974b). Throughout this paper the term 'blue-light interruption' refers to this 4 h exposure in the schedule given above. Cultures were returned to the dark after the irradiation programme.

Spore counts were made 72 h after the start of black light irradiation except in studies on the kinetics of sporulation where spore counts were made at various times during the dark period after irradiation. The procedure has already been described (Tan & Epton, 1973) and spore numbers were expressed as a percentage of sporulation of cultures kept continuously in the dark after 12 h black light irradiation (Tan, 1974b).

RESULTS

Light quality effective in re-promotion of sporulation

After the blue-light interruption, cultures were exposed for 4 h to either black light, yellow light, red light, or far-red light, before being replaced in the dark. Spore counts showed that black light and far-red light re-promoted sporulation to the control level, while in yellow or red light sporulation was at the blue-light-inhibited level. That red light was found to be ineffective is consistent with the results of Kumagai & Oda (1969) for *A. tomato*.

Re-promotion of sporulation by near ultraviolet light

Cultures were subjected to black light or near ultraviolet light for 0–60 min after the blue-light interruption. The results showed that even a brief exposure of black or near ultraviolet light could re-promote sporulation (Fig. 1). Complete recovery from the blue-light inhibition was obtained after 30 min black light, and almost complete recovery was obtained with more than 15 min near-ultraviolet irradiation. The difference in the two curves may be accounted for by the difference in the irradiances employed and also the slight difference in light quality.

The time course of sporulation of cultures subjected to the blue-light interruption followed by 30 min near-ultraviolet irradiation was compared
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Fig. 1. Re-promotion of sporulation in B. cinerea by (a) black light, BLB, and (b) near ultraviolet light, NUV. ○, Expt 1; △, Expt 2. Each symbol is mean of two cultures, ten counts per culture. Vertical bars show confidence limits at $P = 0.05$.

Fig. 2. Kinetics of sporulation of cultures subjected to blue-light interruption followed by 30 min near-ultraviolet light (○, △), and of cultures kept in the dark after photoinduction (●, ▲). Circles, Expt 1; triangles, Expt 2. Each symbol is mean of two cultures, ten counts per culture. Confidence limits at $P = 0.05$. Treatment for ○, △ shown by the horizontal rectangle (B, blue-light; NUV, near-ultraviolet light).

with that of cultures in darkness after photoinduction. The results showed that the time course of sporulation of cultures subjected to the above irradiation treatment was no different from that of cultures kept in darkness after photoinduction (Fig. 2).

The effect of a dark period between the blue-light interruption and the subsequent near-ultraviolet irradiation was studied to find out whether there is any loss of effectiveness of re-promotion by near ultraviolet.

After the 4 h blue-light irradiation, cultures were placed in the dark
for 0–20 h before being subjected to 30 min near-ultraviolet irradiation. The results (Fig. 3) showed that even a half hour intervening dark period could reduce the 're-promotive' effect of near ultraviolet light. When the intervening dark period was about 20 h, the cultures appeared to have lost the ability to recover from the blue-light inhibition of sporulation. A 'point of no return' has thus been reached.

Re-promotion of sporulation by far-red light

After the blue-light interruption, cultures were subjected to far-red irradiation for 0–60 min in one series of experiments, and 120–360 min in another. The results (Fig. 4) showed that far-red light is not as effective
as near-ultraviolet in the re-promotion of sporulation. Although about 15 min irradiation resulted in some re-promotion, complete recovery from the blue-light inhibition required more than 360 min of continuous far-red irradiation.

The time course of sporulation of cultures subjected to blue-light interruption followed by 4 h far-red light was compared with that of cultures in darkness after photoinduction. The results showed there to be no significant difference between the two sporulation curves.

Essentially similar experimental procedures as in the study of the effect of intervening darkness between blue light and near-ultraviolet irradiation were employed to study the situation with far-red irradiation. After 4 h blue light, cultures were placed in the dark for 0–28 h before being subjected to 4 h far-red irradiation. It was found that 1 h of intervening darkness reduced the ‘re-promotive’ effect of far-red light, and when this intervening dark period was as much as 28 h, the cultures were almost unresponsive to the far-red irradiation. This finding is similar to that for the near-ultraviolet studies, which may suggest that far-red light has the same action as ultraviolet, although a higher dose is required.

DISCUSSION

The blue-light inhibition of sporulation of B. cinerea was found to be reversed by near ultraviolet and far-red light. Red light which was reported to be effective in A. solani (Lukens, 1965), was found to be ineffective. Red-light irradiation immediately following the blue interruption has, in fact, been found to result in greater suppression of sporulation in B. cinerea (Tan, 1975). In another fungal response, Ingold (1968, 1969) found that the suppression of spore discharge by blue light in Sphaerobolus stellatus Todes ex Pers. could be reversed by subsequent yellow light, and Ingold & Oso (1969) found that in Ascobolus crenulatus P. Karst., blue-light-promoted ‘puffing’ could be prevented if the apothecium was simultaneously illuminated with yellow light.

It is still not clear why full recovery of the blue-light inhibition of sporulation by far-red light required some 360 min of continuous irradiation whereas with near ultraviolet light only 30 min or so sufficed. The results suggest that the photoreceptor may have a lower peak of action in the far-red region of the spectrum, thus indicating that its absorption in the far-red is less. Full recovery from the blue-light inhibition of sporulation in H. oryzae (Honda et al. 1968) and A. tomato (Kumagai & Oda, 1969) were also obtained with near-ultraviolet irradiation of a short duration, of 30–45 min.

The findings that sporulation of cultures was the same whether subjected to blue-light interruption followed by near-ultraviolet or far-red light, or kept in darkness after photoinduction, confirmed the view that sporulation in B. cinerea is controlled by an interconvertible photoreceptor. Sporulation which is reversed by blue light could be immediately re-promoted by subsequent near-ultraviolet or far-red irradiation.

It may be argued that the loss of the ‘re-promotive’ effect of near
ultraviolet or far-red light could be attributed to the reduced sensitivity due to age (Tan & Epton, 1973), especially in view of the fact that cultures with 20 h intervening darkness were almost 7 days old when they were subjected to near ultraviolet. Cultures with intervening darkness of 0.5–8 h, however, were still within the sensitive phase, so the age factor can be disregarded. This loss of re-promotive power will be explained in Tan (1975), where models for the mechanism of photoreception will be described.

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REFERENCES


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