Influence of ultraviolet radiation on the colony formation of Fomes annosus (Fr.) Cooke diaspores suspended in water

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Abstract. — *F. annosus* diaspores suspended in water were exposed to ultraviolet radiation, wavelength 365 nm. Colony formation of the diaspores was reduced in exponential response to the ultraviolet radiation. The colony formation of basidiospores declined more than that of asexual diaspores.

Introduction

The wavelength, dose and dose-rate of radiation affect fungal growth and sporulation (LEACH 1962a, b, 1963). The effect of radiation on fungi may be a result of the substances it induces in the fungi (LEACH 1965) or the mutations it produces (NORMAN 1951, CASTRO F. et al. 1971). The same part of the spectrum has been found to affect both sexual and asexual reproduction (LEACH & TRIONE 1966).

Irradiation of *Glomerella cingulata* Stoneman spores with ultraviolet wavelengths below 313 nm showed that an exposure time of 5 sec. inhibited the germination, and an exposure time of 15 sec. killed all the spores. Ultraviolet irradiation was also found to inhibit the mycelial growth of *G. cingulata* (STEVENS 1928). Radiation of a wavelength of 265 nm has been seen to inhibit the germination of *Rhizopus suinus* Niels. conidia (DIMOND & DUGGAR 1940). Inactivation of micro- and macroconidia of *Neurospora crassa* Shear & Dodge by ultraviolet radiation has also been demostrated. The effect was most intense in the wavelength range of 240—280 nm. The inactivating effect on microconidia exceeded that on macroconidia (NORMAN 1951). The conidia of *Pyricularia oryzae* Cav. died in 180 sec. under ultraviolet radiation (MANIBHUSHANRAO & SURYANARAYANAN 1971).

Information about the influence of visible and near visible radiation on wood-decaying fungi is relatively scant. Light has been found to stimulate the sporulation of *Trichoderma viride* Pers. ex Fries (GUTTER 1957). Violet and blue light were very effective in stimulating the sporulation of *Trichoderma lignorum* (Tode) Harz. (MILLER & REID 1961). Infrared radiation of long duration is most potent in retarding the mycelial growth of *Fomes annosus,* whereas infrared radiation lasting less than 10 min. stimulates mycelial growith. Solar, ultraviolet and X-ravs radiation, if of long duration, inhibits the mycelial growth of this fungus (NEGRUCKIJ 1962). In the laboratory, the fungus has produced conidia both in light and darkness (RISHBETH 1951).

As day advances and the sun rises higher, the short-wave portion of the spectrum extends into a range of even shorter wavelength radiations, while in the long-wave the radiation is of approximately the same wavelength all day long (SÜRING et al. 1934, LUNELUND 1945). Clouds in front of the sun considerably reduce the amount of ultraviolet radiation reaching the ground. Even a thin shroud of cloud reduces the ultraviolet irradiance by

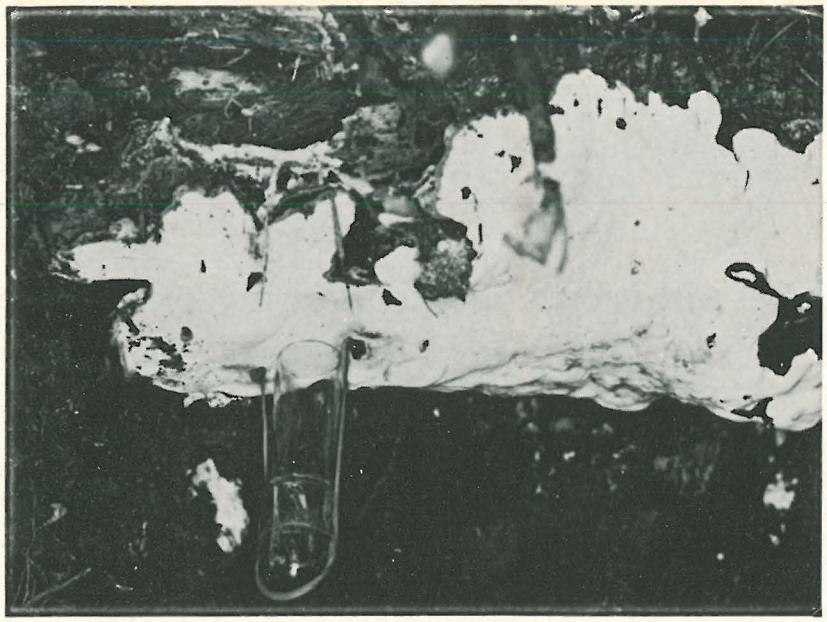

Fig. 1. Collection of basidiospores.

20 per cent (LUNELUND 1945). The increase of short-wave radiation in the middle of the day may contribute towards the sharp fall in the deposition of airborne *F. annosus* diaspores around noon (KALLIO 1970). The purpose of the present study was to investigate the effect of ultraviolet radiation of 365 nm on the colony formation of both sexual and asexual diaspores of *F. annosus*.

Material and methods

1. Effect of ultraviolet radiation on colony formation by asexual diaspores

The *F. annosus* isolate used in the study was derived from a spruce tree *(Picea abies* (L.) Karst.) in Helsinki. Isolations were grown on 9 cm Petri dishes with 25 ml malt agar. After 10—15 days on this substrate, the agar was transferred to a flask containing 1000 ml water. The agar remaining unbroken in the flask rotated gently for 10 min. Ten ml suspension was then pipetted from this flask into 50 ml water. The suspension was kept in glass bottles under ultraviolet radiation, 50 cm from the source of light (lamp Airam HgMu 125 W). Thermal radiation of the bottles was inhibited by means of a water filter. According to the measurements of Oy Airam Ab, Lamp Manufacturers (Research Engineer M. Saarinen), the water filter and the bottles used allowed 90—92 per cent of the emitted 365 nm radiation to pass through. Having passed through the water filter and the top surface of the bottle the dose-rate was 113—115 μ W/cm². The irradiation took place in a dark room with an air temperature of approx. 26°C. The exposure times were of 2, 4 and 6 hours duration. There was one control bottle per exposure time. The diaspores were suspended in the water for the prescribed time, but their bottles were kept in darkness and were not exposed. On completion of irradiation, 0.1 ml suspension was cultured on malt agar in a Petri dish, diameter 14 cm, the suspension being spread as evenly as possible over the agar surface. The number of *F. annosus* colonies was counted from the substrate 10 days later. The results were calculated as mean values of four parallel cultures. The experiment was repeated 20 times.

2. *Effect of ultraviolet radiation on basidiospores*

Basidiospores were caught during the period from August 2 to September 10, 1971, from sporophores on the root undersurfaces of old stumps in a hundred year old, *F.*

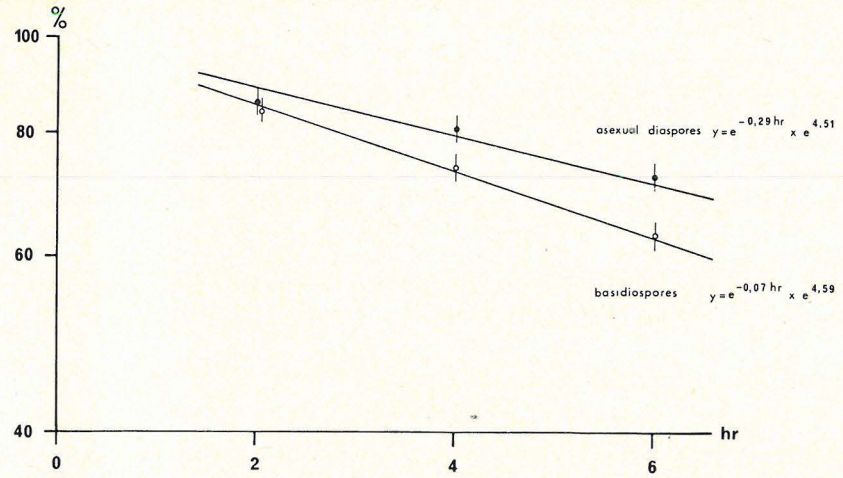

Fig. 2. Influence of ultraviolet radiation on colony formation by diaspores. Triple standard deviation is shown on either side of the percentages.

annosus contaminated spruce stand in Helsinki. With the utmost care, a little earth was removed from beneath the sporophores so that it was possible to attach a test tube against the white, active-looking sporophores (Fig. 1). The length of the tubes used was 60 mm, the diameter 12.7—13.9 mm, and the cross-section area 127—152 mm². Attached by an elastic band, the tube remained in position, with its opening against the sporophore surface, for 18 hours (from 14.00 to 08.00). The sporophore had been exposed 2—4 hours before the tube was fastened to its surface. The tube was never fastened twice to the same point on the sporophore surface, and a tube was applied to the same sporophore on a maximum of two days. The hole dug in the ground to expose the sporophore was covered by plastic film during this time. Hence the active surface covered by the tube was fresh every time and was able to continue undisturbed spore production. The production during the period of the study ranged from 48,000 to 419,000 spores/cm²/hr. After the observations were completed, the sporophore was removed and studied in the laboratory. Not once were conidiophores seen on its surface.

Basidiospores were irradiated in the same way as asexual diaspores. 3 ml water was added to the test tube that had been covering the sporophore. 1 ml of this suspension was added to 1000 ml water. 10 ml of this suspension again was added to 50 ml water, and the resulting suspension was exposed to ultraviolet radiation in the same way as the suspension of asexual diaspores. Cultures were carried out and results calculated in the same way as for asexual diaspores.

Results and discussion

The results are shown in the form of graphs in Fig. 2. The effect of ultraviolet radiation on the colony formation by *F. annosus* diaspores can be seen from the exponential response. A similar result, by a closely corresponding method, was obtained with the conidia of *Pyricularia oryzae* Cav. (MANIBHUSHANRAO & SURYANARAYANAN 1971) and *Neurospora crassa* Shear & Dodge (NORMAN 1951).

The effect of ultraviolet radiation on the *F. annosus* diaspore colony formation, according to the t test, was highly significant for all three exposure times (2, 4 and 6 hours). The differences between 2 and 4, and 4 and 6 hours were also highly significant. After an exposure time of 2 hours the difference in colony formation between asexual and sexual diaspores was significant with a probability of 2 per cent, and after exposure times of 4 and 6 hours with a probability of 0.1 per cent. The difference may be due to the larger number of nuclei in the conidia

(cf. ROLL-HANSEN 1940, NORMAN 1951), or other structural differences between sexual and asexual diaspores (DIMOND & DUGGAR 1941, SUSSMAN & HALVORSON 1966). The diaspore density in the suspension subjected to irradiation varied in the present study, averaging 54/0.1 ml among the asexual diaspores able to form colonies, calculated from the control suspension, and the basidiospore density, accordingly, 29/0.1 ml. This may also have contributed to a smaller fall in the colony formation of asexual diaspores than in that of basidiospores (cf. NORMAN 1951). According to regression analysis, the time of ultraviolet radiation explained 18 per cent of the variation in the colony formation of asexual diaspores, while for the sexual diaspores the percentage was 56. This finding would also suggest that some factor other than ultraviolet radiation exerted a greater effect among the asexual than the sexual diaspores. This view is supported by the result of the two-factor variance analysis.

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