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SCANNING ELECTRON MICROSCOPY OF BASIDIOSPORES OF AGARICUS
BITORQUIS AND OF HEALTHY AND VIRUS-INFECTED AGARICIUS BISPORUS

JOOST A. STALPERS and ANNEMARIE VAN ZAAZEN
Centraalbureau voor Schimmelcultures, Baarn and Mushroom
Experimental Station, Horst, The Netherlands

INTRODUCTION

Virus disease of the cultivated mushroom Agaricus bisporus (Lange) Imbach can still cause considerable crop losses. Early detection is important: as soon as virus infection is confirmed, measures can be taken to restrict damage. A simple, quick but reliable method of testing would be extremely useful. Attempts to develop such test methods have, however, failed as yet (Lemke, 1979).

Schisler et al. (1967) suggested that the wall of basidiospores from virus-infected mushrooms might be slightly thinner than that of spores from healthy mushrooms, since the former usually germinated more quickly and more abundantly than the healthy spores. Nair (1976) examined spores with a scanning electron microscope (SEM). The spores were collected from healthy and virus-infected mushrooms, grown under identical conditions. The spores from healthy mushrooms were found to be intact, whereas spores from virus-infected mushrooms had collapsed. Degree of collapse could perhaps be used to diagnose virus infection of mushrooms.

Although Nair did not mention the technique of preparation used in his study, we assume that his specimens were air-dried and not fixed. Since techniques of preparation are a major factor in the degree of collapse, we compared unfixed air-dried "healthy" and "infected" spores from A. bisporus with spores fixed and critical point-dried (Samson et al., 1979). The four spored Agaricus bitorquis (Quél.) Sacc., immune to mushroom virus disease (van Zaayen, 1976), was also studied.

MATERIALS AND METHODS

Healthy and diseased fruiting bodies were mainly collected from experimental trays or plots in (isolated) growing-rooms of the

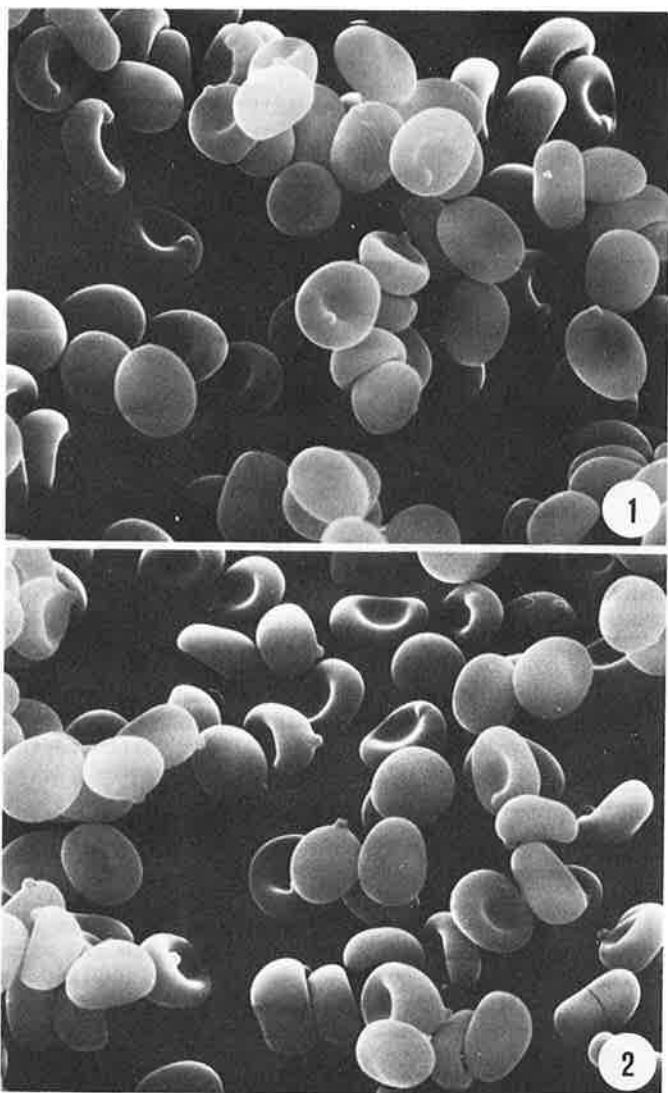


Fig. 1. Spores of healthy *A. bisporus*, air-dried; 2500X

Fig. 2. Spores of virus-infected *A. bisporus*, air-dried; 2500X

Mushroom Experimental Station at Horst (Limburg), the Netherlands.

Small parts of gills containing mature basidia and spores were put into perforated Beem capsules (a Beem Product, size 3) and fixed in unbuffered aqueous 6% glutaraldehyde at 4°C for 24 hours. The material was twice rinsed for 10 min. in water, dehydrated twice in methoxyethanol for 10 min. and washed twice in 100% acetone. The samples were then transferred to a Balzar's critical point-drying apparatus, dried in CO₂, mounted on stubs, coated with gold in a Polaron sputter coater and examined in a Leitz AMR 1000A scanning electron microscope.

Air-dried spores from spore prints on filter paper were examined for comparison.

RESULTS AND DISCUSSION

All specimens, which were fixed and critical point-dried were intact and had not collapsed. No morphological difference could be detected between spores of healthy A. bisporus (Fig. 3, 5), diseased A. bisporus (Fig. 4) and A. bitorquis (Fig. 6) Air-dried spores always showed collapse. However, no significant difference could be found between the degree of collapse of "healthy" (Fig. 1) and "infected" spores (Fig. 2).

There is thus no indication that the spores of diseased Agaricus bisporus collapse on the basidia or after dehiscence. The degree of collapse after air-drying could have been an indication of virus infection, but any differences in collapse between "healthy" and "diseased" spores are too faint to be used as a diagnostic criterion.

SUMMARY

Scanning electron micrographs are shown of basidia and spores of Agaricus bisporus and A. bitorquis. Significant differences in morphology between spores from healthy and from virus-infected mushrooms of A. bisporus could not be detected.

RESUME

Des micrographies au microscope électronique à balayage sont montrées de basides et spores de Agaricus bisporus et A. bitorquis. La morphologie des spores des exemplaires sains et des exemplaires infectés par virus de A. bisporus ne montrait aucune différence significative.

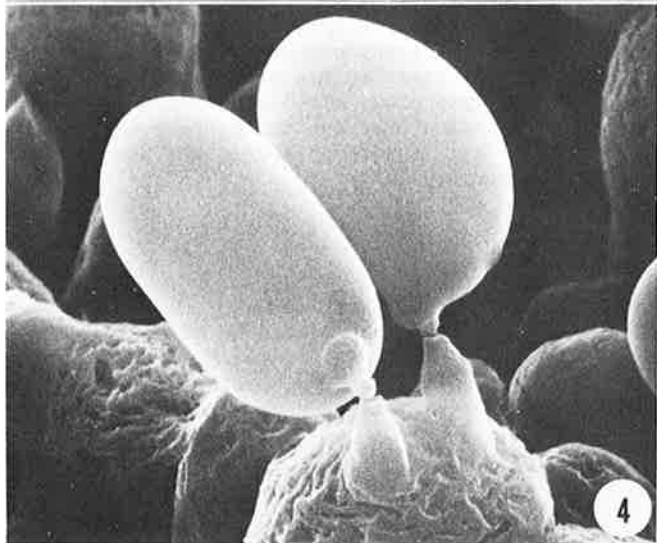
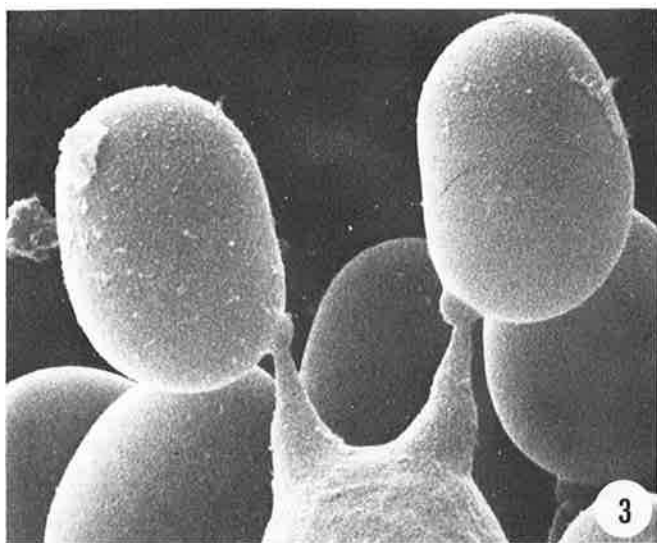


Fig. 3. Basidium and spores of healthy *A. bisporus*. critical point-dried; 10,000X

Fig. 4. Basidium and spores of virus infected *A. bisporus*, critical point-dried; 10,000X

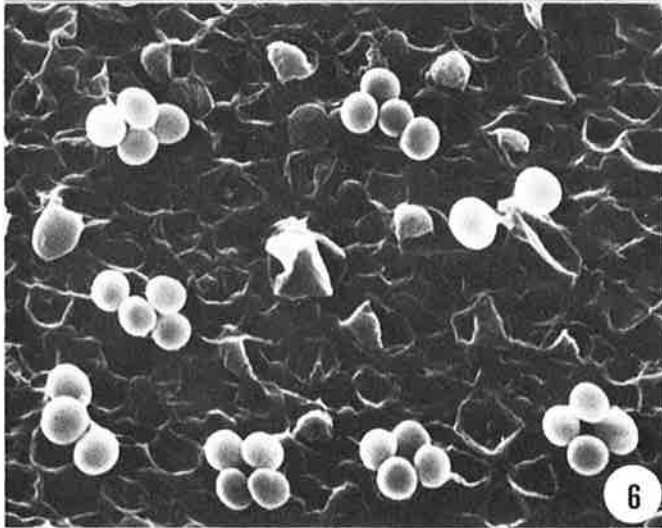
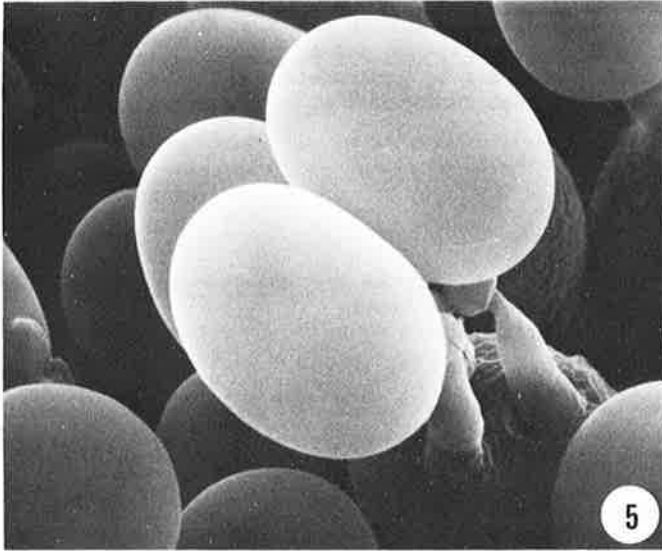


Fig. 5. Basidium and spores of A. bisporus var. tetrasporus, critical point-dried; 10,000X

Fig. 6. Spores of A. bitorquis on basidia; 1500X

ZUSAMMENFASSUNG

Raster elektronenmikroskopische Aufnahmen von Basidien und Sporen von Agaricus bisporus und A. bitorquis werden gezeigt. Gesicherte morphologische Unterschiede zwischen gesunden und von Virus befallenen Sporen von A. bisporus waren nicht zu finden.

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