

## NOTES

### Virus-Like Particles in *Cephalosporium acremonium*

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*Cephalosporium acremonium* cultures were studied for the presence of virus-like particles. Relatively few particles were found in the preparations, indicating that the number of particles present in these cells may be much lower than in *Penicillium* species.

The report of Ellis and Kleinschmidt (2) demonstrating the presence of virus-like particles in *Penicillium stoloniferum* and that of Banks et al. describing similar particles in strains of *P. chrysogenum* (1) prompted a study of *Cephalosporium acremonium* cultures for such particles. The results of this study are reported here.

A 30-liter stainless steel fermentor containing 13.5 liters of the medium shown in Table 1 was inoculated with 1.5 liters of a 72-hr vegetative culture of a strain of *C. acremonium* ATCC 11550. The fermentor conditions were as follows: incubation temperature, 27 C; agitation, 450

heavy sediment and a clear supernatant layer formed. The supernatant layer was removed, diluted with 0.2 M ammonium acetate buffer, and then differentially centrifuged onto carbon-coated electron microscope grids. These were stained

TABLE 1. *Cephalosporium fermentor medium*

Component	Per cent
Soybean meal.....	2.50
Peanut meal.....	4.50
DL-Methionine.....	0.75
Calcium carbonate.....	0.58
Lard oil.....	5.00
Methyl oleate.....	0.58

rev/min; aeration, 0.36 cfm from 0 to 6 hr and 0.55 cfm from 6 to 56 hr. At 56 hr, approximately 400 ml of the culture was centrifuged at 1,000 × g for 15 min. The supernatant fluid and the upper oily layer of the residue were discarded. The middle portion of the remaining residue was removed and washed with approximately 400 ml of deionized water. The mixture was again centrifuged, the cells were separated from the supernatant fluid, and the wash was repeated once.

Ten grams of the wet cells was suspended in 10 ml of phosphate buffer (pH 6.7) and 0.1% thio-glycolic acid and was passed through a French press at 20,000 psi. After a few days at 4 C, a

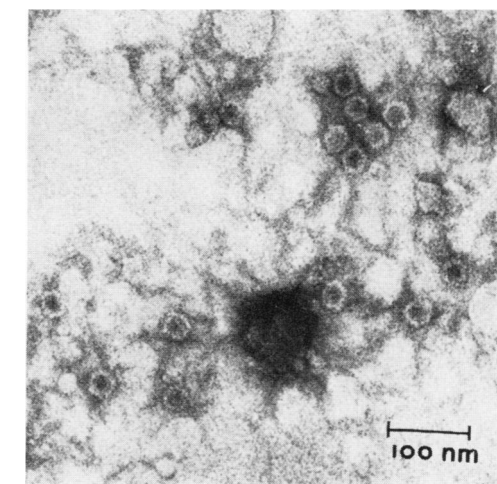


FIG. 1. Electron micrograph of virus-like particles from *Cephalosporium acremonium* cells separated by differential centrifugation directly onto electron microscope grids. Stained with potassium phosphotungstate. × 110,000.

with potassium phosphotungstate and examined in the electron microscope.

The particles (Fig. 1) are of a similar size and shape to those previously reported in *P. stoloniferum* (2). Full and empty particles were observed in this preparation. Observations of a number of particles indicate a possible capsomeric structure.

Relatively few particles were found in the preparations. Breakage of these cells was difficult and might account for fewer particles. Particles aggregated with each other or with cell debris would be effectively removed by the differential centrifugation, which would lower the observed concentration. By comparison, the number of particles present in these cells may be much lower than in *Penicillium* species.

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#### LITERATURE CITED

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