

Pall, hl. L. Blasticidin-S: on inhibitor

of protein synthesis in *Neurospora*.

tein synthesis. This difficulty makes it desirable to have alternative antibiotics which effectively inhibit protein synthesis in *Neurospora* for comparison.

This note concerns a study of the antibiotic, **blasticidin-S**, on two-day-old mycelial pads of wild type strain ST74A, grown as described previously (Pall 1970 *Biochim. Biophys. Acta* 203:139). Pods were shaken with **blasticidin-S** for various periods of time before being shaken with $1 \mu\text{C L-}^3\text{H}$ lysine for 2 minutes. The mycelial pods were then washed, extracted with 5% TCA and the uptake and incorporation into protein (hot TCA insoluble, NaOH soluble fraction) were measured. As shown in Table 1, even a half-minute preincubation with **blasticidin-S** gives significant inhibition of incorporation into protein. A ten minute preincubation with **50 $\mu\text{g/ml}$ blasticidin-S monohydrochloride** gives almost complete inhibition of incorporation. Thus **blasticidin-S** is rapidly effective in inhibiting lysine incorporation into protein. Cycloheximide inhibits incorporation even more rapidly than does **blasticidin-S**, a half-minute preincubation with **10 $\mu\text{g/ml}$ cycloheximide** inhibiting incorporation by **98%**.

Blasticidin-S, under the above condition*, shows little effect (<20%) on lysine uptake. The amino acid pool, as measured by cold TCA extractable ninhydrin positive material, shows little (0-30%) increase in the presence of **blasticidin-S**. Consequently the inhibition of incorporation into protein would be expected to be a good measure of the inhibition of protein synthesis. Other experiment* showed that a 10-minute preincubation of pads with **50 $\mu\text{g/ml}$ blasticidin-S monohydrochloride** had little or no (00%) effect on the rate of uridine uptake or its incorporation into nucleic acid. The results support the conclusion that **blasticidin-S** is a rapid, relatively specific inhibitor of protein synthesis in *Neurospora*.

The **blasticidin-S monohydrochloride** was manufactured by the **Kaken Chemical Co., Ltd., Japan** and was a gift of **Marubeni-Lida (America), Inc., San Francisco**. The author would be happy to supply samples of **blasticidin-S monohydrochloride** to interested investigators.

Cycloheximide has been used widely as an inhibitor of protein synthesis in *Neurospora*. It is difficult to eliminate the possibility that results obtained with cycloheximide or other antibiotics may be due to secondary effect* independent of the inhibition of pro-

tein synthesis in *Neurospora*.

Table 1. Inhibition of lysine incorporation into protein.

Time of preincubation	Antibiotic used	% Inhibition of incorporation
1/2 min.	1 $\mu\text{g/ml}$ blasticidin-S . HCl	18
1/2 min.	10 $\mu\text{g/ml}$ blasticidin-S . HCl	72
1/2 min.	50 $\mu\text{g/ml}$ blasticidin-S . HCl	86
1/2 min.	10 $\mu\text{g/ml}$ cycloheximide	98.2
10 min.	1 $\mu\text{g/ml}$ blasticidin-S . HCl	68
10 min.	10 $\mu\text{g/ml}$ blasticidin-S . HCl	93.9
10 min.	50 $\mu\text{g/ml}$ blasticidin-S . HCl	99.0