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Recommended Citation

Hedman, S. C., W.K. Bates, and D.O. Woodward (1965) "Induction of tyrosinase by B-lactose in *Neurospora crassa*," *Fungal Genetics Reports*: Vol. 8, Article 20. <https://doi.org/10.4148/1941-4765.2123>

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Abstract

Induction of tyrosinase by lactose in *Neurospora crassa*

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During the course of our experiments dealing with the β -galactosidase system in *Neurospora*, we often noted that crude extracts and ammonium sulfate fractions turned dark

upon standing. Assays confirmed that these extracts and fractions had high levels of tyrosinase activity. It soon was evident that growing *Neurospora* with R-lactose as the sole carbon source was responsible for this induction of tyrosinase.

The strain used in these initial experiments was L-5, selected from 74A for its increased ability to use R-lactose as its sole carbon source. Elevated levels of tyrosinase were detected when L-5 was grown in Vogel's minimal medium, pH 5.0, with 1.5% O-lactose as the only carbon source. Adequate growth seemed to be dependent upon aeration, either by shaking or by bubbling, and a growth temperature of about 27°C.

With the above conditions, suitable harvest of the mycelium could be made after approximately a week's growth. Harvesting consisted of collecting the mycelium on a wire screen, followed by several washes with cold water. The mat was then pressed dry with paper towels, fragmented by hand, and placed on the lyophilizer. After lyophilization, the mycelium was ground in a Wiley mill. Extraction at 4°C with 0.0125 M tris-HCl buffer, pH 7.8, with a ratio of 15 ml buffer per gram of powder yielded a crude extract high in tyrosinase activity.

More recently, induction of tyrosinase by R-lactose has been studied by us in other strains of *Neurospora*. These include the strains designated by Horowitz as T^L, T^{Sing-2} and T^{PR}. In these three strains we observed elevated levels of tyrosinase following growth on O-lactose. Following induction by lactose, the tyrosinase levels in these three strains were not equal,

with TL often showing the highest level of activity. No attempts have been made to study the induction of tyrosinase by lactose in low sulfur minimal medium (Horowitz and Shen 1952 J. Biol. Chem. 197:513) or in the presence of aromatic amino acids (Horowitz et al., 1960 J. Mol. Biol. 2:96). We have used this lactose-induced tyrosinase quite satisfactorily in heat lability and disc electrophoresis studies.

This work was supported by an NSF Predoctoral Fellowship to S.C.H. and by NSF and USPHS Grants. ■ ■ ■ Department of Biological Sciences, Stanford University, Stanford, California.