

Fungal Genetics Reports

Volume 11

Article 23

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F. Cooke

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Recommended Citation

Cooke, F. (1967) "Ascospore color mutants and low germination in *Neurospora*," *Fungal Genetics Reports*: Vol. 11, Article 23. <https://doi.org/10.4148/1941-4765.1988>

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Abstract

Ascospore color mutants and low germination

Cooke, F. Ascospore color mutants and low germination in *Neurospora*.

Tokyo 74: 110), have proved to have inviable ascospores and therefore to be of limited use for recombination studies. A pale-spored, pantothenic-acid-requiring mutant described by Threlkeld (1965 *Can. J. Genet. Cytol.* 7: 171) has proved to be more promising since the pale ascospores will germinate, but with a lower frequency than the wild type spores. This mutant has been used to investigate the relationship between paleness and germination frequency. It was found that the color of the spores could be altered by varying the amounts of pantothenic acid in the crossing medium.

Isoallelic crosses were set up on media with different concentrations of pantothenic acid (PA). Apart from a wild type control cross, all crosses were *pan-2* (B3) A x *pan-2* (B3) a. With no PA added, no growth was possible. With 0.5 mg of PA per liter, vegetative growth was normal. The table shows the effect of the varying concentration of PA on perithecial and spore production and on spore viability. A wild type cross was carried out at each concentration of PA and without PA. In none of these cases were there noticeable differences in growth rates, perithecial production or spore color. Percentage germination of the wild type cross was 97%. In this cross, therefore, PA has no effect on growth rate, fertility or germination.

| Concentration of PA in mg/l | Growth and perithecial production | Spores | % Germination |
|-----------------------------|-----------------------------------|-----------|---------------|
| 0.05 | weak growth = no perithecia | | |
| 0.1 | weak growth = few perithecia | none | |
| 0.2 | moderate growth = few perithecia | very pale | 33 |
| 0.5 | normal growth = many perithecia | pale | 80 |
| 1.0 | as above | pale | 80 |
| 5.0 | as above | dark | 97 |
| 20.0 | as above | dark | 95 |

Whereas ascospore color mutants have been much used in *Sordaria* and *Ascobolus* to select for aberrant tetrads, such mutants have proved difficult to find in *Neurospora*. Those which are described, *asco* (Stadler 1956 *Genetics* 41: 528) and *ts* (Nakamura 1961 *Bot. Mag.*,

In the *pan-2* x *pan-2* cross the results show a progressive increase in spore darkness with increasing PA. The spore color at 5 mg/l and 20 mg/l of PA was indistinguishable from the wild type spore color. The results also show an increasing ability of the spores to germinate with increasing PA. The spore germination at 5 mg/l and 20 mg/l of PA was not significantly different from wild type spore germination. Both germination frequency and spore darkness decrease progressively with reduction in PA concentrations.

Since it was possible that some PA would break down during the autoclaving of the medium, the media were made up by

adding solutions of PA through a Millipore filter subsequent to autoclaving. It was found that medium made in this way acted in the same way as medium in which the PA had been autoclaved.

This experiment shows, therefore, a correlation between PA level, spore color and spore germination in the *pan-2* mutants, and lends further support to the suspicion that spore color mutants in *Neurospora*, in contrast to those in *Ascobolus* and *Sordaria*, have a low frequency of spore germination because of the change in the spore color due to the presence of the mutation. If the evidence from *asco*, *ts* and *pan-2* proves to be of general application, then all spore color mutants in *Neurospora* will have reduced ascospore germination and will be of limited value as markers for the selection of aberrant tetrads for intragenic recombination studies. ■ ■ ■ Department of Biology, Queen's University, Kingston, Ontario, Canada.