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

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., 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

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 war found that medium mode in this way acted in the same way as medium in which the PA hod been autoclaved.

This experiment show, therefore, a correlation between PA level, spore color and spore germination in the pan-2 mutants, and lends further support to the suspicion that spare color mutants in Neurospora, in contrast to those in Ascobolus and Sordaria, hove a low frequency of spore germination because of the changer 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.