Fungal Genetics Reports

Volume 19 Article 19

Reference strains of Neurospora sitophila

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Recommended Citation
Perkins, D. D. (1972) "Reference strains of Neurospora sitophila," *Fungal Genetics Reports*: Vol. 19, Article 19. https://doi.org/10.4148/1941-4765.1877

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Abstract

Reference strains of *Neurospora sitophila*

Perkins, D. D. Reference strains of Neurospora sitophila.

When new heterothallic strains of Neurospora are collected from nature, the easiest and most reliable way to determine their taxonomic status is probably to cross them to reference strains representing the already established species. A fully fertile test cross should indicate not only species status and mating type, but also in favorable cases the absence or presence of chromosome rearrangements.

For this purpose, tester strains of each species should include both mating types and should ideally be highly fertile and isosequential, not only with each other but with the original type material on which the species description was based, or authentic derivatives of the type material.

There is no real problem in choice of references for N. crassa, where all the commonly used wild-type strains (Lindegren, Emerson, St. Lawrence) are essentially similar in fertility, chromosome sequence and morphology. The situation is much less clear for N. sitophila and N. intermedia.

Numerous cultures labelled as belonging to these species are maintained in culture collections (Centraalbureau voor Schimmelcultures, American Type Culture Collection, Fungal Genetics Stock Center). In 1969, I intercrossed several of these strains that supposedly belonged to the same species. Unexpectedly, a number of the crosses were infertile, producing rudimentary or barren perithecia, or none at all. Infertility of this type could result from the accumulation of mutations or chromosome rearrangements during stock maintenance. Alternatively it could reflect original misidentification, contamination, or mislabelling of stocks, so that the species designations were incorrect. Fertile, authenticated reference stocks were clearly needed.

By returning to original type cultures, or to the most reliable cultures of known pedigree, it has been possible to derive fertile pure cultures of sitophila that in all likelihood correspond to the original type material. I hove not yet succeeded in establishing authentic intermedia references, however.

Derivation of the sitophila references is shown in the pedigree. Starting material consisted of two pairs of stocks. One pair, CBS 178.27A and CBS 179.27a, was deposited by Shear in the Centraalbureau voor Schimmelcultures, Boom, in 1927, the year the species was described. We obtained these in 1971 from the Fungal Genetics Stock Center shortly after the FGSC had received them from the CBS. The second pair, 56.8A and 56.4a, was derived by Dodge from a cross of Arlington 6A X Arlington

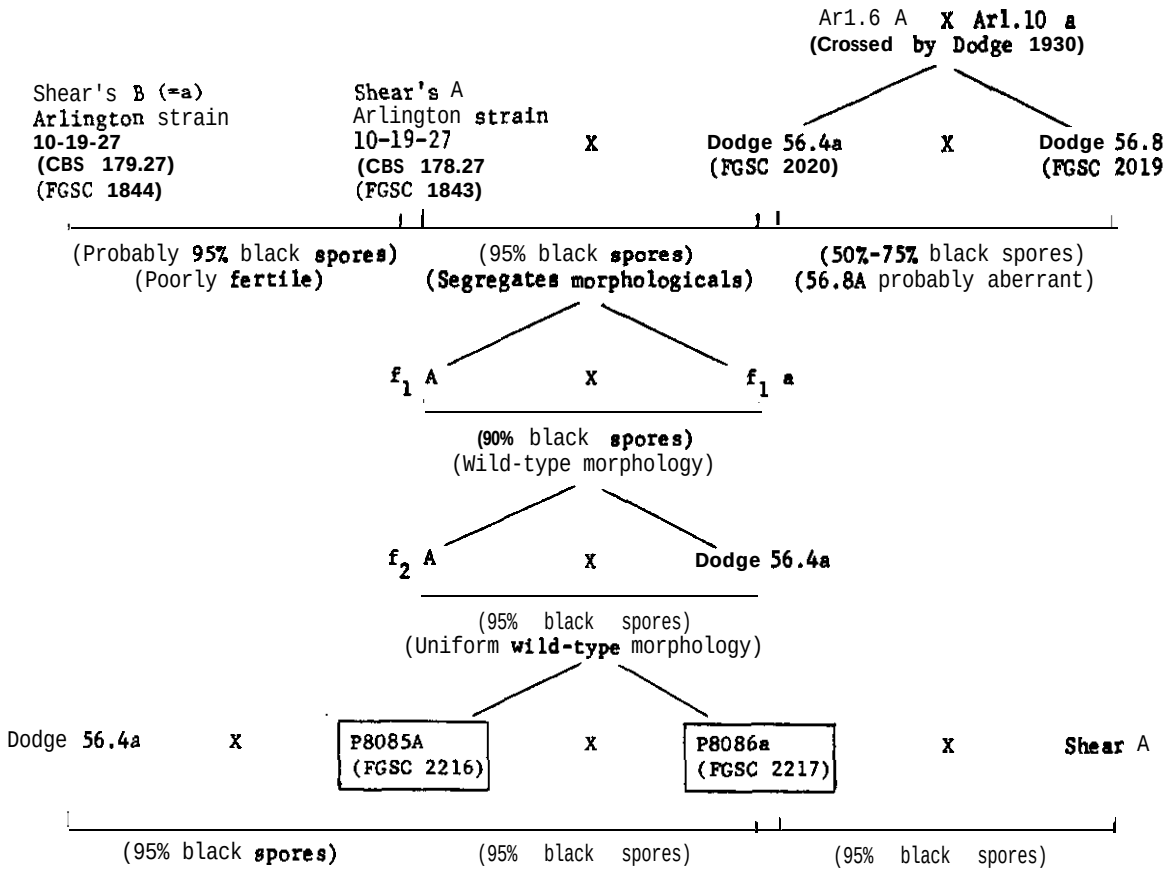
108 (Dodge 1930 Mycologia 22:9). (8 is the old way of designating mating-type a.) M. P. Backus obtained cultures 56.8 and 56.4 from Dodge, and they were maintained at Wisconsin since about 1930. Transfers were kindly sent to us by D. P. Mahoney II in 1969.

CBS 179.278 is now aconidiate and very poorly fertile. CBS 178.27A corresponds more nearly to the described wild-type phenotype. Enough ascospores were produced in a cross between 179.270 and 178.27A to indicate that they probably do not differ by any gross rearrangement such as a translocation.

The two Dodge stocks are morphologically wild-type and interfertile, but about 50% of the ascospores from the intercross were white and inviable, indicating that the two strains probably differ in chromosome structure or in a gene affecting spore pigmentation. Patterns of defective spores in asci from 56.8 x 56.4 suggest that a reciprocal translocation either was present in Dodge's cross, or originated in the 56.8 culture. (56.8A also gives 20-50% white spores when crossed by P8086a - - see later.)

In contrast, the cross CBS 178.27A x 56.4a is both highly fertile and apparently isosequential, with 95% of ascospores viable and fertile. Progeny from this cross were highly variable in morphology, and included classes of segregants resembling the N. crassa mutants ropy and peach. Apparently one or both parents is heterokaryotic - not surprising for strains that have been carried for 40 yews in stock. As shown in the pedigree, two intercrosses and a backcross were used to obtain the strains P8085A and P8086a. These and their siblings were morphologically wild-type and homogeneous, and tests showed them to be fertile and isosequential both with each other and with the original Shear and Dodge strains.

Derivation of Neurospora sitophila reference stocks P8085A and P8086a.



It is proposed to use P8085 A and P8086 a as reference N. sitophila stocks, to which strains of unknown species-status will be crossed and the fertility determined as an indication of relationship. The chromosome sequence of P8085 A and P8086 a will be designated as the standard for N. sitophila. The recommended reference stocks hove been deposited in the FGSC and given the FGSC numbers shown in the pedigree.

Shear and Dodge bored the species description of N. sitophila on material deposited in CBS by Herter (1919 Z. Ges. Getreidewesen 1 1:1), from Berlin. The Arlington strains came from contaminated media in a greenhouse at the USDA Experimental Farm, Rosslyn (Arlington), Virginia. Crosser of Herter X Arlington strains, and of Arlington X Arlington, were shown to produce normal perithecia (Shear and Dodge 1927 J. Agr. Rer.34: 1019, Table 6), and the Arlington strains were classed as N. sitophila on this basis. The shear strain CBS 178.27 and the Dodge strain 56.4 ore fully fertile with one another and appear to be isosequential after being maintained separately for over four decodes. This strengthens confidence in their authenticity. As expected, each is barren in crosses with N. crassa. - - - Department of Biological Sciences, Stanford, University, Stanford, California 94305.