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The pattern of the parasexual cycle in *Aspergillus amstelodami*

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INTRODUCTION

The parasexual cycle has been adequately demonstrated in a number of fungi. Of these, however, linkage has been shown in only a few instances. In the more recent work on *Aspergillus nidulans*, Käfer (1958) gave evidence for eight linkage groups. Linkage has also been demonstrated in *Penicillium expansum* by Barron (1962) and indicated by Pontecorvo *et al.* (1953) in *Aspergillus niger*, and Hastie (1962) in *Verticillium albo-atrum*.

Recently Strømnaes & Garber (1963), in a detailed study of parasexuality in *Aspergillus fumigatus*, analysed 1184 haploid segregants from 17 heterozygous diploids. Although 23 markers were used in the various crosses, linkage was found in only one instance. This linkage in the so-called *al-lys leu bu* group, confirmed a previous finding by Berg & Garber (1962). Strømnaes and Garber also reported a most unusual distribution of haploid progeny which could not be related either to linkage or to random assortment of chromosomes at the time of haploidization.

Experimental

In a strain of *Aspergillus amstelodami*, originally isolated from stored corn, we established a heterocaryon between a brown strain requiring methionine (*br meth*) and a white strain requiring para-aminobenzoic acid and leucine (*w paba leu*). A second heterocaryon was established between the brown methionine-requiring strain and a pale-blue strain requiring para-aminobenzoic acid, adenine and arginine (*pb paba ad arg*). Heterozygous diploid strains were recovered without difficulty from both crosses.

Segregants from those diploids were obtained by selecting coloured heads or sectors from small colonies derived from single diploid spores. Only one segregant of any one colour was recovered from each colony thus ensuring independent origin for all segregants analysed. The results of the classification of these segregants are summarized in Table 1.

An analysis of the results from diploid I showed that all 58 white haploids required para-aminobenzoic acid, indicating linkage between the *w* and *paba* loci. The lack of *paba*-requiring diploids suggested that the two loci concerned are either on opposite sides of the centromere or on the same side with the *paba* locus very close to the centromere. Free recombination has taken place between the *w* locus and all other loci concerned in the cross.

In diploid II, the 17 pale-blue haploids were adenine-requiring, indicating linkage between these two markers. Of the pale-blue diploids, four required adenine and the remainder were prototrophs. The four adenine-requiring diploids confirmed the linkage relationship suggested by the haploids and showed that the adenine locus was proximal to the centromere on the same arm of the chromosome. The low frequency of adenine-requiring diploids suggests that the *ad* and *pb* loci are not closely linked.

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Table 1. *Classification of* 196 *independent segregants from two heterozygous diploids in* Aspergillus amstelodami

Diploid I: $\frac{br}{+}\ \frac{meth}{+}\ \frac{+}{w}\ \frac{+}{paba}\ \frac{+}{leu}$			Diploid II: $\frac{br}{+}\ \frac{meth}{+}\ \frac{+}{pb}\ \frac{+}{paba}\ \frac{+}{ad}\ \frac{+}{arg}$		
	White segregants			*Pale-blue segregants*	
Ploidy	*Nutritional Requirements*	*Number*	*Ploidy*	*Nutritional Requirements*	*Number*
2*n*	wild-type	81	2*n*	wild-type	36
n	*paba*	23	2*n*	*ad*	4
n	*paba meth*	10	*n*	*ad*	12
n	*paba meth leu*	10	*n*	*ad arg*	4
n	*paba leu*	15	*n*	*ad meth*	1

DISCUSSION

The recovery of both diploid and haploid recombinants from the segregants from these crosses reveals that the parasexual cycle is active in *Aspergillus amstelodami*. The high proportion of haploids recovered, especially from diploid I, is a point of special interest, in that haploids of this order of frequency are not usually recovered without special chemical treatments or double selection techniques. The establishment of two linkage relationships in crosses involving a total of nine markers indicates that the pattern of the parasexual cycle in *A. amstelodami* is similar to that in *A. nidulans* rather than to *A. fumigatus*. In *A. amstelodami*, the segregation of non-linked loci does not conform exactly with the classical thesis of random chromosome assortment however, as proposed for *A. nidulans*, the ratio seems to be biased against the double auxotrophic segregants in diploid II.

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