

Presumptive new alleles of het-c

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Abstract

Presumptive new alleles of *het-c*

Perkins, D. D. Presumptive new alleles of het-c,

detected by the use of partial diploids.

The genetic basis of heterokaryon incompatibility is difficult to study because differences at numerous incompatibility (het) loci are widespread in commonly used laboratory stocks (Holloway 1955 Genetics 40: 117; Wilson and Garnjobst 1965 Genetics 53:621). It may thus be a formidable

task to obtain appropriately marked strains that differ in the allele at only a single het locus. In spite of the difficulty, Wilson and Garnjobst succeeded in devising tester stocks with defined genotypes at het-c, -d, and -e, and with no incompatibility differences at other loci. Two alleles were identified at each locus. These testers have been useful references for strains having similar genetic backgrounds. However, identifying multiple alleles or identifying and mapping additional het loci remains exceedingly laborious so long as tests depend on the ability of strains to form stable heterokaryons. The basic difficulty with heterokaryon tests of vegetative incompatibility is that they reflect incompatibility anywhere in the genome, with incompatibility at one locus masking compatibility at all others. What is needed is a practical technique for studying the effect of one locus at a time.

The difficulties inherent in heterokaryon testing can be largely avoided by using appropriate duplications that cover only a single het locus, leaving the rest of the genome haploid. Heterozygosity for incompatibility genes in such a duplication is known to result in abnormal vegetative growth. Duplications were first used in this way for the heterokaryon-incompatibility associated with the mating-type locus (Newmeyer and Taylor 1967 Genetics 56: 771; Newmeyer 1970 Can. J. Genet. Cytol. 12:914). Duplications that cover the region of III including het-c have also been used extensively. With them, five different genotypes have been distinguished that behave as though they were multiple alleles of het-c. (The possibility has not been excluded that these may involve incompatibility differences at another linked locus in the duplication. However, there is no evidence for such differences among our commonly used laboratory strains. Until another locus has been demonstrated the genotypes will be represented simply as het-c allele.)

It is hoped that the use of heterozygous duplications will be extended to other incompatibility loci, and that a molecular analysis of the incompatibility phenotype will be encouraged by the availability of these strains, from which heterozygous duplication can be made up in any combination desired for analysis.

When translocation T(III→VR)NM149 is crossed by Normal sequence, one third of the viable progeny contain viable duplications for a portion of III that includes het-c. If the two parents have identical het alleles, the duplications are homozygous and phenotypically wild-type. If the two parents carry different het alleles, the duplications are heterozygous and phenotypically abnormal, initial growth being flat, conidial, and inhibited to various degrees depending on the particular alleles involved (Perkins 1968 Proc. 12th Intern. Congr. Genet. 1:67; Genetics 61:47). Crosses with T(III→VR)NM149 testers can thus be used for defining the het-c genotype, irrespective of what genes may be present at het-d, het-e, or other loci outside the duplicated segment.

Table 1. Presumed alleles of het-c, detected in duplication from T(III→V)NM149 X Normal.

Allele	Source of standard reference allele	Heterozygote*	Phenotype of heterozygote**
het-C	OR23-1 A (FGSC#987)	C/c	Brown-flat, without conidia or aerial hyphae, spreading to cover slant.
		C/PA	Nonspreading highly inhibited wispy; also a few spreading brown-flat.
		C/AD	Fern-like brown flats, distinguishable from C/c. Include some highly inhibited.
		C/GR	Slow inhibited sparse; also some spreading brown-flats.
het-c	Em a (FGSC#692)	c/PA	Similar to C/PA.
		c/AD	Similar to C/AD.
		c/GR	Similar to C/GR.
het-c ^{PA}	Panama a (FGSC#1131)	PA/AD	Inhibited, not covering slant. Resemble "Dark Agar" A/o duplications from In (II-IR) H4250.
		PA/G R	Strikingly abnormal.
het-c ^{AD}	Adiopodoume A (FGSC#430)	AD/G R	Very inhibited.
het-c ^{GR}	Groveland-1c a (FGSC#1945)		

* Homozygous duplications C/C, c/c, PA/PA, AD/AD and GR/GR are all wild-type in morphology, but are Barren in crosses.

** Brown pigment is produced only on a complete medium (GCP is used) or on minimal containing tyrosine and phenylalanine.

The descriptions of morphology are based on casual observations and are by no means precise or systematic.

Laboratory strains of *N. crassa* have fallen into two classes, corresponding to he+-c or he+-C of Wilson and Garnjobst. This is true also of a number of *N. crassa* strains collected from nature in America and Africa. However, three distinct new genotypes have been discovered, with the origins and characteristics shown in Table 1. Their behavior is that expected of multiple alleles of he+-c. Tester strains have been prepared that contain each of the five Presumptive alleles, both in Translocation sequence and in Normal sequence (Table 2).

Each of the following *N. crassa* strains has a he+-c allele similar to one of those described in Table 1, judged according to whether duplications from intercrosses are phenotypically normal or abnormal. Most of these strains have been tested only against C and c testers, but Costa Rice A (FGSC 851) has been tested with all five. FGSC stock numbers are in parentheses.

he+-C: OR8-1 a (988). Lein 7A (847). Lein 8a (1693), Chilton a (1691), Em A (691), SY4f8a (621), ro-3 a (43).

he+-c: Abbott A (1228), Abbott 12a (351), fl^PA (295), fl(P346) a (297), fl^PA (1838), fl^Pa (1690), Liberia a (967), Puerto Rico 18a (429).

het-c^{PA}: Panama a (1130, 1132, 1133, 1165). Costa Rica A (851, 852).

Table 2. Stocks for testing he+-c compatibility via duplications

Allele	Chromosome sequence*	Genotype of stock	FGSC#
<u>he+-C</u>	T	T(IIL→VR)NM149 m-3 <u>he+-C</u> A	2011
	T	T(IIL→VR)NM149 m-3 <u>he+-C</u> a	2012
	N	<u>he+-C</u> A; use OR23-1 A	987
	N	<u>he+-C</u> a; use ORB-1 a	988
<u>he+-c</u>	T	T(IIL→VR)NM149 <u>het-c</u> A	1483
	T	T(IIL→VR)NM149 <u>he+-c</u> a	1482
	N	<u>he+-c</u> A; (in preparation)	
	N	<u>he+-c</u> a; use Em a	692
<u>het-c</u> ^{PA}	T	T(II→V)NM149 <u>het-c</u> ^{PA} A	2187
	T	T(II→V)NM149 <u>het-c</u> ^{PA} z	2188
	N	<u>het-c</u> ^{PA} A	2189
	N	<u>het-c</u> ^{PA} a	2190
<u>het-c</u> ^{AD}	T	T(II→V)NM149 <u>het-c</u> ^{AD} A	2191
	T	T(II→V)NM149 <u>het-c</u> ^{AD} a	2192
	N	<u>het-c</u> ^{AD} A; use Adiopodoume A	430
	N	<u>het-c</u> ^{AD} a; (in preparation)	
<u>het-c</u> ^{GR}	T	T(II→V)NM149 <u>het-c</u> ^{GR} A	2193
	T	T(II→V)NM149 <u>het-c</u> ^{GR} a	2194
	N	<u>het-c</u> ^{GR} A	2195
	N	<u>het-c</u> ^{GR} a; use Groveland-1 a	1945

* T = translocation sequence; N = normal wild-type sequence.

The search for additional he+-c alleles is continuing with newly collected *N. crassa* wild strains, and with he+-c alleles introgressed from non-*crassa* material by repeated backcrosses to *N. crassa* strains containing a marker (ro-3) that is closely linked to het-c. ■ ■ ■ Department of Biological Sciences, Stanford University, Stanford, California 94305.