IMPROVED BACKCROSSED STRAINS GIVING CONSISTENT MAP DISTANCES

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Backcrossed mutant strains which produce consistent map distances and negligible interference.

To obtain strains for mapping of uvs, nuh and mus mutants which would also serve for detection of effects on crossing over, a large number of commonly used mutant strains having normal growth rate under permissive conditions have been backcrossed 5-10 times to the St. Lawrence wild type (SL 74-OR23-1VA. FGSC #2489A). The most useful and basic of these strains (single and multiply marked strains) are now deposited at FGSC (Fig. 1, Table I). Hopefully, various special purpose strains, and statistical evaluation of the mapping data, will be available in a year or two. So far all these strains were found to be heterokaryon compatible (presumably het C d e).

1) Wild type 74-ORS-6a (deposited as FGSC #4200) is a progeny from the 6th backcross to 74-OR23-1VA of ORS a (FGSC #2490 a), which shows invariant neat round colonies when grown on DNA test agar plus sorbose. It was used for backcrossing when only A mutant strains were available.

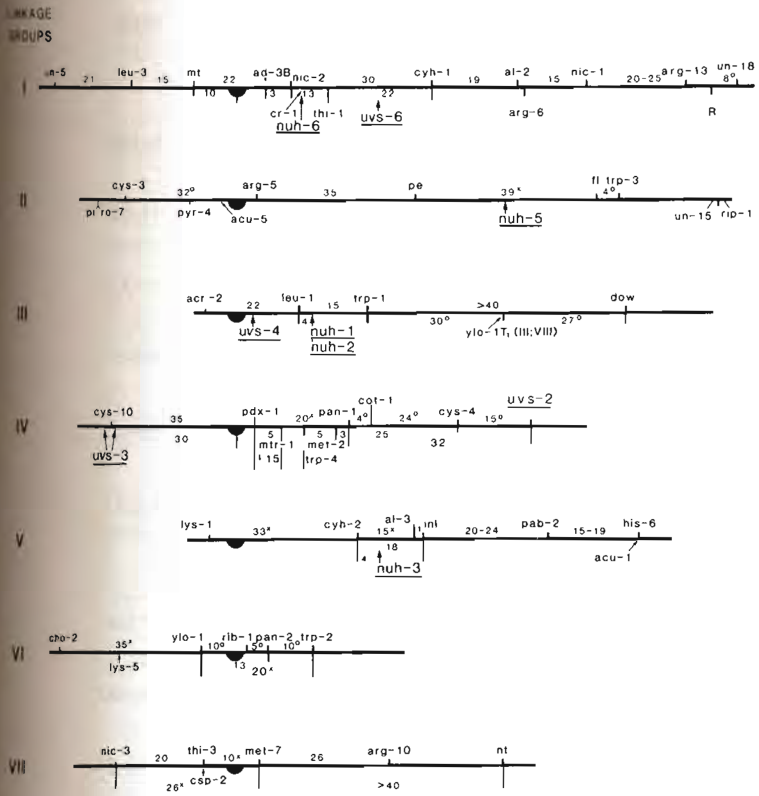

Fig. 1: Map distances of mutants in strains back-crossed to St. Lawrence wild type:
ˣ = several crosses, > 1000 progeny tested;
no superscript, 2 or more crosses, 300-1000 progeny tested;
° = single crosses, totals < 300.

2) Morphological mutants in wild type and mutant backcrosses: Since morphological mutants interfere with DNase halo tests, we have recorded any morphologically abnormal variants from all back crosses. Even in small samples (30-100) and also in backcrosses of wild type, these are found not infrequently. Many show a similar phenotype (resembling so) on IR in the so 1:1 in individual asci. One of these mutants has been mapped on IR in the so region. It seemed that the A wild type (74-OR23-1VA, #2489) would be heterokaryotic for so, since it was the common parent. However, when we intercrossed recently obtained A and a progeny from the 7th wild type back cross, we also obtained some asci segregating 1:1 for a morphological mutant (of a slightly different type). The current hypothesis is, therefore, that spontaneous mutants affecting growth rate and morphology are common enough to be produced with low frequency in any strain, and to be found after "purification" in the occasional ascospore progeny of most crosses.

3) Single marker strains: Fig. 1 shows all the markers, as well as uvs and nuh mutants, deposited with FGSC at this time. Markers below the line are deposited only as single mutant strains, except for the case of uvs mutants which also are available with nutritional markers for forced heterokaryons (FGSC #4190-4199). In some cases two different alleles are available when a less leaky or more fertile mutant became available belatedly.

Single marker strains in various linkage groups have the following FGSC #:

LG. I	4000 - 4023	LG. IV	4053 - 4068	LG. VI	4093 - 4110
II	4024 - 4042	V	4069 - 4080	VII	4081 - 4092
III	4053 - 4068				

upr and uvs mutants: FGSC #4171-4179 (some backcrossed by A. Schroeder). nuh mutants: 4180-4189.

4) Multiple marker strains for approximate localization of mutants inside each linkage group: Mutants indicated above the line in Fig. 1 have been combined into strains with widely-spaced markers of one linkage group (those for linkage group II are provisional). These strains can be used after crosses to alcoy when linkage group has been found. In such cases, we routinely set up crosses to strains with markers for both possible groups and isolate a sample from the cross that discharges ascospores first. Testing in this sample for the unmapped mutant and the marker closest to the alcoy translocation break reveals which of the two crosses should be analysed in detail.

TABLE I

Backcrossed strains deposited with F.G.S.C.*

FGSC	mt	Genotype	Allele No.
Linkage Group I			
4000	A	un-5	
4001	a	un-5	b39(t)
4002	A	leu-3	
4003	a	leu-3	R156
4004	A	ad-3B	
4005	a	ad-3B	2-017-0034
4006	A	nic-2	
4007	a	nic-2	43 002
4008	A	cr-1	
4009	a	cr-1	B123
4010	A	thi-1	
4011	a	thi-1	56501
4012	A	cyh-1	
4013	a	cyh-1	KH52
4014	A	al-2	15300
4015	a	arg-6	29997
4016	A	nic-1	
4017	a	nic-1	S1413
4018	A	arg-13	
4019	a	arg-13	RU12
4020	A	un-18	
4021	a	un-18	T 54 M49(t)
4022	A	R	
4023	a	R	35408
Linkage Group II			
4024	A	ro-7	
4025	a	ro-7	R2470
4026	A	pi	
4027	a	pi	B101
4028	A	cys-3	
4029	a	cys-3	P22
4030	A	pyr-4	
4031	a	pyr-4	36601
4032	A	acu-5	
4033	a	acu-5	JI18
4034	A	arg-5	
4035	a	arg-5	27947
4036	A	pe	
4037	a	pe	Y8743m
4038	A	trp-3	
4039	a	trp-3	td37
4040	A	un-15	T54 M50(t)
4041	A	rip-1	
4042	a	rip-1	4M(t)
Linkage Group III			
4043	A	acr-2	
4044	a	acr-2	KH5
4045	A	ad-4	
4046	a	ad-4	44 206,t(F15)
4047	A	leu-1	
4048	a	leu-1	33757
4049	A	trp-1	
4050	a	trp-1	10575
4051	A	dow	
4052	a	dow	P616
Linkage Group IV			
4053	A	cys-10	(50t)
4054	a	cys-10	39816
4055	A	pdx-1	
4056	a	pdx-1	37803
4057	A	mtr	
4058	a	mtr	(DRS)15
4059	A	trp-4	
4060	a	trp-4	Y2198
4061	A	met-2	K43
4062	A	met-2	P159
4063	A	pan-1	
4064	a	pan-1	5531
4065	A	cot-1	
4066	a	cot-1	C102(t)
4067	A	cys-4	
4068	a	cys-4	K-7
Linkage Group V			
4069	A	lys-1	
4070	a	lys-1	33933
4071	A	cyh-2	
4072	a	cyh-2	KH53(r)
4073	a	al-3	RP-100
4074	A	inl	
4075	a	inl	37401
4076	A	pab-2	
4077	a	pab-2	H193
4078	A	his-6	
4079	a	his-6	Y152 M105
4080	a	acu-1	JI48
Linkage Group VII			
4081	A	nic-3	
4082	a	nic-3	Y31881
4083	A	thi-3	
4084	a	thi-3	18558
4085	A	csp-2	
4086	a	csp-2	UCLA101
4087	A	met-7	
4088	a	met-7	4894
4089	A	nt	
4090	a	nt	65001
4091	A	arg-10	
4092	a	arg-10	B317
Linkage Group VI			
4093	A	cho-2	
4094	a	cho-2	47904
4095	A	lys-5	37402
4096	a	lys-5	(asco)
4097	A	lys-5	
4098	a	lys-5	DS6-85
4099	A	ylo-1	
4100	a	ylo-1	Y30539y
4101	A	rib-1	
4102	a	rib-1	51602(t)
4103	A	pan-2	
4104	a	pan-2	Y154-M64(B5)
4105	A	pan-2	
4106	a	pan-2	B36(M.Case)
4107	A	trp-2	
4108	a	trp-2	75001
4109	A	trp-2	
4110	a	trp-2	(DRS)41

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*Not all of the strains were included in the Eleventh Revision of the Stock List (NN #29) at the time we went to press. However, all will be available shortly. Interested person should write F.G.S.C. for any strain listed in the Table but not in the Eleventh Revision.

FGSC	mt	Genotype	Allele No.
4111	A	un-5 leu-3 A nic-2 cyh-1	
			b39(t) R156 43002 KH52
4112	a	cyh-1 al-2 nic-1 arg-13	
			KH52 15300 1413 RU12
4113	A	al-2 nic-1 arg-13 un-18	
4114	a	al-2 nic-1 arg-13 un-18	
			15300S 1413 RU12 T54M49(t)
4115	A	arg-5 pe fl trp-3	
4116	a	arg-5 pe fl trp-3	
			27947 Y8743m L td37
4117	A	acr-2 leu-1 trp-1 dow	
4118	a	acr-2 leu-1 trp-1 dow	
			KH5 33757 10575 P616
4119	A	acr-2 trp-1 dow	
4120	a	acr-2 trp-1 dow	
			KH5 10575 P616
4121	A	cys-10 pdx-1 pan-1	
4122	a	cys-10 pdx-1 pan-1	
			39816 37803 5531
4123	A	cys-10 pdx-1 pan-1 uvs-2	
4124	a	cys-10 pdx-1 pan-1 uvs-2	
			39816 37803 5531 (no#)
4125	A	pdx-1 pan-1 cys-4	
4126	a	pdx-1 pan-1 cys-4	
			37803 5531 K7
4127	A	pdx-1 pan-1 cys-4 uvs-2	
4128	a	pdx-1 pan-1 cys-4 uvs-2	
			37803 5531 K7 (no#)
4129	A	lys-1 cyh-2 al-3 inl^t pab-2 his-6	
4130	a	lys-1 cyh-2 al-3 83201^t + his-6	
			33933 KH53,r RP100 H193 Y152M105
4131	A	lys-1 al-3 inl^t pab-2	
			33933 RP100 83201(t) H193
4132	a	lys-1 cyh-2 al-3 inl^t pab-2	
			33933 KH53,r RP100 83201(t) H193
4133	A	cyh-2 al-3 inl^t pab-2	
4134	a	cyh-2 al-3 inl^t pab-2	
			KH53r RP100 83201(t) H193
4135	A	lys-1 cyh-2 al-3 inl^t his-6	
			33933 KH53,r RP100 83201(t) Y152M105
4136	a	lys-1 inl his-6	
			33933 37401 Y152M105

FGSC	mt	Genotype	Allele No.
4137	A	cho-2 ylo-1 trp-2	
4138	a	cho-2 ylo-1 trp-2	
			47904 Y30539y 75001
4139	A	rib-1 pan-2(B2) trp-2	
4140	a	rib-1 pan-2(B2) trp-2	
			51602(t) Y153M66 (DRS)41
4141	A	nic-3 met-7 arg-10	
4142	a	nic-3 met-7 arg-10	
			Y31881 4894 B317
4143	A	thi-3 met-7 nt	
4144	a	thi-3 met-7 nt	
			18558 4894 65001
4145	a	acr-2 leu-1 ylo-1 T1 (III;VI) dow	
			KH5 33757 Y30539y 1 P616
4146	A	leu-3 ad-3B thi-1 cyh-1	
			R156 2-017-0034 56501 KH52
4147	A	leu-3 ad-3B cyh-1	
			R156 2-017-0034 KH52
4148	a	cyh-1 al-2 nic-1	
			KH52 15300 1413
4149	a	cys-3 arg-5	
			P22 27947
4150	A	arg-5 pe fl	
			27947 Y8743m L
4151	A	pdx-1 cot-1 cys-4	
4152	a	pdx-1 cot-1 cys-4	
			37803 C102(t) K7
4153	A	acr-2 dow; cho-2 ylo-1 trp-2	
4154	a	acr-2 dow; cho-2 trp-2	
			KH5 P616 47904 Y30539y 75001
4155	A	cot-1; al-3	
4156	a	cot-1; al-3	
			C102(t) RP100
4157	A	sn cr-1; csp-2	
			C136 B123 UCLA101
4158	A	sn cr-1	
4159	a	sn cr-1	
			C136 B123
4160	A	sn cr-1; al-3 inl^t	
4161	a	sn cr-1; al-3 inl^t	
			C136 B123 RP100 83201(t)

FGSC	mt	Genotype	Allele No.
4162	A	sn cr-1; pe fl	
4163	a	sn cr-1; pe fl	
		C136 B123 Y8743m L	
4164	A	sn cr-1; pe fl; al-3 inl^{t}	
4165	a	sn cr-1; pe fl; al-3 inl^{t}	
		C136 B123 Y8743m L RP100 83201 (t)	
4166	A	cr-1; pe fl; al-3 inl^{t}	
		B123 Y8743m L RP100 83201(t)	
4167	A	cr-1; pe fl	
4168	a	cr-1; pe fl	
		B123 Y8743m L	
4169	A	pe fl	
4170	a	pe fl	
		Y8743m L	
4171	a	upr-1	(no#) (RWT)
4172	A	uvs-2	
4173	a	uvs-2	(no#) (DRS)
4174	A	uvs-3	
4175	a	uvs-3	ALS11
4176	A	uvs-4	
4177	a	uvs-4	ALS12
4178	A	uvs-6	
4179	a	uvs-6	ALS35
4180	A	nuh-1	
4181	a	nuh-1	FK001
4182	A	nuh-2	
4183	a	nuh-2	FK027

FGSC	mt	Genotype	Allele No.
4184	A	nuh-3	
4185	a	nuh-3	FK003
4186	A	nuh-5	
4187	a	nuh-5	FK005
4188	A	nuh-6	
4189	a	nuh-6	FK006
4190	A	acr-2; uvs-2; his-6	
		KH5 (no#) Y152M105	
4191	a	pan-1 uvs-2	
		5531 (no#)	
4192	A	uvs-3; ylo-1 pan-2 (B5)	
4193	a	uvs-3; ylo-1 pan-2 (B5)	
		ALS11 Y30539y Y154M64	
4194	A	uvs-3 trp-4 pan-1	
4195	a	uvs-3 trp-4 pan-1	
		ALS11 Y2198 5531	
4196	A	acr-2 uvs-4 leu-1	
		KH5 ALS12 33757	
4197	a	uvs-4 leu-1	
		ALS12 33757	
4198	A	uvs-6; mtr-1 met-2 pan-1	
		ALS35 (DRS)15 P159 5531	
4199	a	uvs-6; nic-3	
		ALS35 Y31881	
4200	a	wild type (SL)	
		(BCVI of ORSa) 74-ORS-6a	

Strains covering each linkage group have the following numbers:

LG.	
I	4111 - 4114 and 4146 - 4148
II	4115 - 4116 and 4169 - 4150
III	4117 - 4120 and 4145
IV	4121 - 4128 and 4151 - 4152
V	4129 - 4136
VI	4137 - 4140
VII	4141 - 4144

Similar strains carrying sn cr-1 for replica testing are now being tested and will be deposited if standard procedures can be devised (those for groups III - VII are ready and available from Montreal directly for anyone willing to try them out).

5) Strains for alcoy follow up and mutagenesis:

Alcoy follow ups: III; VI, several markers: FGSC #4153 and 4154
IV; V: cot-1 al-3: FGSC #4155 and 4156

Strains for velvet replication: sn cr-1; csp-2: FGSC #4157
sn cr-1 FGSC #4158 and 4159

Various other strains carrying sn, cr-1, pe fl, and al-3 inl^{t} in several combinations: FGSC #4160 - 4170

6) Linkage data: To increase efficiency and effectiveness of back crossing, up to three markers of the same linkage group have been back crossed together. Single mutant strains were separated, generally in the 5th back cross, and checked singly in a 6th back cross. The final multiply-marked strains were then built up from the latter strains. Extensive coupling and repulsion linkage data are therefore available for various intervals. In general, recombination values did not change significantly during backcrossing and are shown in Fig. 1 as single average values (with a rough indication of sample size as a measure of reliability). For the few cases with inconsistent values a range of averages is indicated. Surprisingly, practically no interference was observed (possibly expected for the large intervals involved) and in some cases ordering of markers was difficult when two mutants were very close and the third at considerable distance (so that double and single cross overs were almost equally rare). In short, patterns of recombination seem to be identical to those of *Aspergillus nidulans*, but thorough statistical analysis will be needed before definite conclusions can be reached. - - - Department of Biology, McGill University, 1205, Ave. Docteur Penfield, Montreal, H3A 1B1 Canada.