

Ahmad, Majeed. A quick method of obtaining double mutants and heterocaryon compatible isolates from mutants derived from foreign stocks.

the single spore cultures are classified and then the mating-type of the relevant ones is determined by crossing with A and a wild type stocks. This process was found to be laborious and time consuming.

In order to obtain double mutants quickly, spores from crosses were plated on sorbose minimal medium (S.M.) with single supplements and non-growing germinated spores, which showed normal germ tube tips at both ends, were isolated. As shown in the table, by following this procedure, 4 to 40% of the spores isolated proved to be double mutants whereas the percentage of double mutants obtained by following the older procedure ranged from 0 to 4% only.

Mating-type was next determined by putting conidia from the double mutants on V.M. along with A and a heterocaryon compatible mutant strains. This way the mating-type could be determined within 24 to 48 hours of testing.

The same method was adopted for obtaining heterocaryon compatible isolates from foreign mutant stocks by plating their crosses to the wild type on S.M. and isolating non-growing germinated spores. Thus in the case of asco, which has a highly reduced spore viability, about 63% of the lightly coloured non-growing germinated spores isolated, proved to be asco, as against 5% when isolations were made by the usual procedure of adding supplement and isolating growing spores.

Cross from which spores plated.	Supplement added to SM Plating Medium	No. of growing spores isolated	No. of non-growing spores isolated	No. of double mutants	No. of mutant spores	% of double mutant or mutant spores from crosses to wild type.
10575 (<u>tryp-1</u>)x <u>leu-1</u>	leu + <u>tryp</u>	30	-	0		0
	leu	-	19	6		32
A106 (<u>tryp-1</u>) x <u>leu-1</u>	leu + <u>tryp</u>	100	-	4		4
	leu		39	5		13
A67 (<u>tryp-1</u>) x <u>tyr.</u>	AA + <u>tyr</u>	51		1		2
	AA		51	2		4
A65 (<u>tryp-1</u>)x <u>hist</u>	AA		42	17		40
A106 (<u>tryp-1</u>) x <u>hist</u>	AA		39	14		36
<u>asco</u> x <u>EmA</u>	lys	40			2	5
			30		19	63

leu = leucine; tryp = tryptophan; AA = Anthranilic acid; tyr = tyrosine; lys = lysine.

It is customary to obtain the double mutants or heterocaryon compatible isolates from foreign mutant stocks by spreading spores from relevant crosses on supplemented sorbose minimal medium and isolating a number of growing spores. Next,