

Selitrennikoff, C. P. and M. Bailey. A simple classroom complementation experiment.

The utility of *Neurospora* in an undergraduate genetics laboratory course for demonstrating basic genetic principles has been previously described (*Neurospora* in teaching 1966 NN#10: 15). A reliable, visual method for demonstrating intergenic complementation is presented here. This procedure exploits the observation that *csp-1* and *csp-2* strains produce conjoined conidia so that when agar slant cultures are inverted and sharply tapped no freed conidia are observed falling - the "tap test" (Selitrennikoff and Nelson 1973 NN#20: 34). However, when either wild-type cultures or forced heterokaryons of *csp-1/csp-2* are topped, they release an easily observed copious cloud of freed conidia.

The one allele of *csp-1* (UCLA37) and the three alleles of *csp-2* (FS591, FS590, UCLA 101) were each crossed to *nic-2* (43002) and *nit-3* (Y31881) and heterokaryon compatible double mutant cultures isolated (Selitrennikoff; in preparation). These compatible strains can be used in a classical "cis-trans" test requiring only the tap test to detect the presence or absence of complementation. To illustrate, we present an experiment which was performed by the Fall 1973 Genetics Laboratory students at California State University at Fullerton. Each group of 3 students was given agar slant cultures (Vogel's medium N + 1.5% sucrose + 50 µg/ml nicotinamide) of the following strains (each tube was labeled with only the allele number and the appropriate auxotrophic locus designation) and 20 sterile slants of Vogel's N + 1.5% sucrose minimal medium.

1. *nic-2* A
2. *nit-3* A
3. *csp-1* (UCLA37), *nit-2* A
4. *csp-1* (UCLA37); *nic-3* A
5. *csp-2* (FS590); *nic-2* A
6. *csp-2* (FS590); *nit-3* A
7. *csp-2* (FS591); *nic-2* A
8. *csp-2* (FS591); *nit-3* A
9. *csp-2* (UCLA 101); *nic-2* A
10. *csp-2* (UCLA 101); *nit-3* A.

Table 1. Heterokaryotic cultures were formed by co-inoculation of aerial hyphae onto minimal medium. After 7 days' growth at 25°C, tubes were scored by the tap test.  
+ = free conidia  
- = conjoined conidia.

STRAIN	<i>nic-2</i>	UCLA37 <i>nic-2</i>	FS590 <i>nic-2</i>	FS591 <i>nic-2</i>	UCLA101 <i>nic-2</i>
<i>nic-3</i>	+	+	+	+	+
UCLA37 <i>nic-3</i>		-	+	+	+
FS590 <i>nic-3</i>			-	-	-
FS591 <i>nic-3</i>				-	-
UCLA101 <i>nic-3</i>					-

The students were asked to design their own experimental protocol in order to obtain data which would allow them to answer these questions: 1) Is each morphological mutant gene recessive to its wild-type allele? 2) How many loci do the 4 mutant genes i.e., UCLA37, FS591, FS590 and UCLA 101, represent? All groups independently performed the heterokaryon analysis with the results indicated in Table 1 and concluded that FS590, FS591 and UCLA 101 were functional alleles, UCLA37 was a separate locus, and all were recessive. The above 10 strains are available from the Fungal Genetics Stock Center.

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