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Gene order in albino region of LGI

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Abstract

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The marker arg-6: arginine-6 is usually shown left of al-2: albino-2 in published maps. This location was bared largely on data with the incompletely albino mutant aur: aurescent and involved the assumption that al-2 and arg-6 are contiguous. The data prerented here suggest that this assumption may be incorrect. arg-6 is almost certainly right of al-2, and is probably located between al-2 and our. The *results* in Table 1 indicate the order ad-9 act-1 al-2 arg-6 aur hr. (The alternative ad-9 act-I al-2 org-6 hs aur is possible but less likely.) act-1: actidione-1 would thus be the best flanking marker left of al-2. ----

Methods: Crosses were made at 25°C. For the first two crosses, ascospores were isolated to agar slants in 75 mm tubes and heat-shocked the tubes. Glycerol complete medium was used in cross 1. Vogel's minimal + L-arginine (OS mg/ml) + DL-homoserine (0.2 mg/ml) was used in cross 2. Scoring was by transfer to slants of minimal medium plus appropriate supplements. Actidione (cycloheximide) was used for testing at 10 µg/ml, with normal autoclaving. Tests were unambiguous.

Although act-I (Hsu 1963 J. Gen. Microbiol. 32:341) is not as close to albino as might be desired, it has the advantages of showing excellent viability and scorability and requiring no supplement. hr: homoserine is also a good marker, though caution must be taken to use supplemented minimal medium rather than complete medium, so as to avoid inhibition by unidentified constituents of the latter. arg-6 presents no difficulties of culture or scoring.

For crosses 3 to 5, ascospores were suspended, heatshocked and plated in minimal medium containing 1% sorbose + 0.05% glucose + 0.05% fructose, and incubated 48 hours at 34°C before colonies were isolated to minimal slants.

Results: The preferred order, al-2 left of arg-6, is clear when the least frequent single-crossover class is compared with the double crossovers, considering three loci *at a time* in crosser 1 and 2, and assuming first one and then the other of the two alternative orders (a) arg-6 right of al-2, and (b) arg-6 left of al-2:

Postulated order	Parentals	Singles Region 1	Singles Region 2	Doubles
Wad-9 al-2 arg-6	66	24	1	0
(b) ad-9 arg-6 al-2	66	24	0	1
(a) act-I al-2 arg-6	359	35	6	1
(b) act-I arg-6 al-2	359	35	1	6

The order al-2 arg-6 hs was indicated by cross 2 on the basis of 2 singles between al-2 and hs versus 0 doubler. This order was supported by cross 3, where the coupling phase is reversed. Ascospores were plated in minimal sorbose medium. Of 67 prototrophic progeny, 62 were albino. The remaining five isolates (from atypical slow colonies) developed orange pigment, but all five produced albino progeny when crossed by al^+. They thus originated as pseudowild types or mixtures, and were excluded from the tabulation. Only prototrophs from fast-growing colonies were isolated in the succeeding crosses.

in 1969 (Genetics 44, second cross on p. 1194) arg-6 was placed left of aur on the basis of a small number of isolates picked visually as prototrophic germinating ascospores. The identical parental strains hove been preserved, and were crossed to obtain the results listed under cross 4. Seventy fort-growing prototrophic colonies were isolated to slants, and only one among them was rejected as an apparent aneuploid that darkened the medium and grew atypically. The 69 bona fide recombinants were aur, consistent either with the order arg-6 aur hs or arg-6 hs aur, but not with the order aur arg-6 hr.

Cross 4 results indicate that aur is much closer to hs than to arg-6. This was borne out in cross 5, where aur separated from hs in only one prototrphic recombinant among a total of 61 between erg-6 and hs. The single aur-hs recombinant favors locating our left rather than right of hr.

There results suggest that al-2 and aur are separate genes located on opposite cider of arg-6, consistent with what is known both as to recombination and complementation between al-2 and aur. Obviously further critical data ore needed. Confirmation of the order al-2 arg-6 aur would support the proposal of Barratt e+ al. (1954) that th e aur locus be designated al-I: albino-1.

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Table 1. Data from random segregants, establishing the order shown. Crosses 1 and 2 involved total isolation, crosses 3-5, selective plating.

Cross No.	Zygote Genotype and Recombination %	Parental combinations	Recombination: Singles Region 1	Singles Region 2	Singles Region 3	Doubles Regions 1 and 2	Total and % germination	Marker isolation numbers
1	+ + al-2 em-6 / ad-9 ace-1 al^{y} + 15.4 13.2 1.1	36 29	6 7	5 6	1 0	1 0	91 (91%)	Y154M37 KH52 15300 ALS4 29997
2	+ + + he / ace-1 al-2 arg-6 + 7.7 1.9 0.6	148 131	8 15	3 2	1 1	1 0	310 (78%)	KH52 15300 29997 51504
3	+ arg-6 + / al-2 + hs	.. ..	... ..	.. 62		0 ..	62 prototrophs (all al)	15300 29997 51504
4	+ + hs / arg-6 aur +	.. ..	69 ..	0 ..		.. ..	69 prototrophs (all aur)	29997 34508 51504
	1959 results:	.. ..	5 ..	3 ..		.. ..	8 prototrophs (5 aur)	
5	+ + aur he / al-2 arg-6 + +	.. ..	.. ..	60 ..	1 ..	.. ..	61 prototrophs (60 wild type, 1 aur)	15300 29997 34508 51504

(The top number in each pair represents the class that has the + allele of the leftmost marker.)

act-1 = resistant, act-1^{+} = sensitive.

order in albino region of linkage group I. Table 1