GENETIC MAPPING OF THE pho2, PHO82-pho4 AND pho85 LOCI OF YEAST

AKIO TOH-E*

Laboratory of Biochemical Pharmacology, National Institute of Arthritis, Metabolism, and Digestive Diseases, National Institutes of Health, Bethesda, Maryland 20205

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ABSTRACT

The PHO82-pho4 (PHOO-phoD) locus was mapped on the right arm of chromosome VI, 6.5 cM centromere-distal to met10. A pho85 (phoU) mutant was newly isolated, and its map location was determined on the left arm of chromosome XVI, 15 cM centromere-distal to rad1. A mutant gene that causes temperature-sensitive growth on nutrient medium was found very near (or at) the pho85 locus. The supertriploid method was successfully applied to locate the pho2 (phoB) locus on chromosome IV. The pho2 locus was 40 cM from rna11 on the left arm of chromosome IV.

GENETIC analyses of the production of repressible acid phosphatase of Saccharomyces cerevisiae (SCHURR and YAGIL 1971; TOH-E et al. 1973; TOH-E and OSHIMA 1974; UEDA, TOH-E and OSHIMA 1975; UEDA and OSHIMA 1975) revealed that at least six chromosomal genes, pho2 (phoB)* PHO82-pho4(PHOO-phoD), pho81 (phoS), pho80 (phoR), pho85 (phoU) and pho84(phoT), regulate the synthesis of the repressible acid phosphatase coded for by pho5 (phoE). Among the pho genes described above, pho5 and pho80 have been mapped. HANSCHE, BERES and LANGE (1978) mapped pho5 between tsm134 and lys2 on chromosome II. The pho80 gene was mapped on the right arm of chromosome XV (BERES, personal communication). This report describes the location on the yeast chromosome map of the pho2, PHO82-pho4 and pho85 loci that were mapped by using the supertriploid method (WICKNER 1979), as well as by standard tetrad analysis. Map distances were calculated by PERKINS (1949) equation,

 $x = 100 \frac{T + 6NPD}{2(PD + NPD + T)}$, and shown as centimorgans (cM).

The PHO82 strain OC-6A (apho3 PHO82) was crossed with the multiply labelled strain, AT201. The segregation of markers in cross W221 indicated the linkage between PHO82 and his2 (Table 1). To determine the location of PHO82 with respect to the other markers on the right arm of chromosome VI, two more crosses (W275 and W424) were carried out. Data shown in Table 1 locate the position of PHO82 at 6.5 cM centromere-distal to met10.

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^{*} Present address: Department of Fermentation Technology, Faculty of Engineering, Osaka University, Yamada-Kami, Suita-Shi, Osaka Japan 565.

^{*} The nomenclature system of yeast genetic markers recommended by PLISCHEE et al. (1975) is used throughout this work and the conventional name is shown in parenthesis as appropriate.

TABLE 1

Interval tested	Cross*	PD	No. of asci NPD	Т	сM
his2–PHO82	W221	13	0	6	
	W275	52	0	13	
	W424	37	0	19	
	Total	102	0	38	13.6
cdc14-PHO82	W275	55	0	11	
	W424	43	0	12	
	Total	98	0	23	9.5
met10-PHO82	W424	47	0	7	6.5
cdc14-met10	W424	56	0	6	4.8
his2-met10	W424	48	0	15	11.9
his2–cdc14	W424	55	0	8	7.3
rad1-pho85	W465	29	0	14	
	W480	52	0	18	
	W486	31	0	16	
	Total	112	0	48	15.0
trp1-pho2	W320**	33	6	81	
	W382	1	8	1	
	W425	3	0	6	
	W458	27	3	61	
	W471	10	5	37	
	\mathbf{T} otal	74	15	193	49.8
mak21-pho2	W3 82	2	0	9	
	W425	3	0	6	
	W458	17	5	50	
	W471	12	4	40	
	Total	34	9	105	53.2
rna11-pho2	W425	4	0	5	
	W458	31	3	53	
	Total	35	3	58	39.6
rna11-trp1	W425	7	0	2	
	W458	57	0	31	
	Total	64	0	33	17.0
mak21-trp1	W425	7	0	5	
	W458	45	2	39	
	Total	52	2	44	28.6
rna11-mak21	W425	5	0	6	
	W458	32	3	41	
	Total	37	3	47	37.4
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Mapping of pho genes

 $W275 \frac{a}{a} \frac{his2}{a} \frac{his2}$

	Iı	iterval	tested			C	cross*		PD		No. of asci NPD	т	cM
W424	α	his2	cdc14	PHO	82	+	+	+-	+	+	pho3		
	a	+	+	-+-	m	et10	his4	leu2	ura1	ade2	pho3		
W465	a 		+	+	+	+ ;	$\frac{pno3}{1}$		>				
117/190	α a	his7	cdc4	raai o gal-	cac4	gai4 ; +	enos +	++	pho.	s phe	085		
¥¥40U	α	+	+	+ 4	arg6 i	rad1 d	cyh2	mak3	pho	3			
W486	a	his7	arg6	cyh2	rad1	+	+	pho3	phos	5			
	α	+-	+	+	+	leu1	arg1	pho3	+				
	a	+	+	+-	arg1	phos	pho	o2					
W320	α	cdc1	6 asp5	trp1	+	pho3		- 					
	a	+	+-	+	arg1	pho3	b phe	o2					
W382	α	leu2	mak2	1 +	+-	+	· [KIL-0]	-			
a + trp1 pho3 pho2 [KIL-b]***													
W425	a	arg6	pho3	pho2	+	+		+	[KIL	-b]			
11-125	α	+-	+	+	trp1	rna1.	1 m	ak21	[KIL	-0]			
W 458	a	arg1	pho3	pho2	-+-	+		+	[KIL	-b]			
,, 100	α		+	+	trp1	rna1	1 m	ak21	[KIL	-0]			

TABLE 1—Continued

** Pooled tetrad data from crosses other than listed in this table are included in this line. *** [KIL-b] is a mutant killer plasmid showing superkiller phenotype. This mutant plasmid bypasses some mak mutations, but not mak21 (TOH-E and WICKNER, unpublished).

A new pho85 mutant (B104) was isolated from AT199 (a pho3 his7) after ethyl methanesulfonate mutagenesis (LINDEGREN et al. 1965) and mapped. B104 showed two other mutant phenotypes: temperature-sensitive growth on nutrient medium and respiratory deficiency. Both of these phenotypes segregated 2+:2in the cross of B104 × wild type. The temperature sensitivity co-segregated with the pho85 character, but the respiratory deficiency of B104 segregated independently of the pho85 trait. No recombination could be detected between pho85 and ts among 46 asci tested. A temperature-resistant revertant was isolated from the pho85-ts strain (AT275). The revertant still had the pho85 phenotype. However, this reversion was due to an extragenic suppressor.

Since the pho85 locus was known previously to be centromere-linked, AT275 (a pho3 pho85-ts his7) was crossed with strains having known centromere markers. Linkage between pho85 and rad1 on chromosome XVI was found in cross W465. The pooled tetrad ratio collected from the crosses (W465, W480, and W486) was PD : NPD : T = 112 : 0 : 48, and the distance between these two genes was calculated as 15 cM (Table 1). No close linkage between pho85 and mak3 could be observed. The sequence, pho85-rad1-centromere, was deduced by analyzing individual tetrads of cross W486. This arrangement is consistent with the fact that the frequency of second-division segregation of pho85 (25.5%) is higher than that of rad1 (15%).

The linkage group of *pho2* was analyzed by the supertriploid method described by WICKNER (1979). A *pho2* strain W273-14C (**a** *pho3 pho2 arg6*) was mated with spores of supertriploid 1234 en masse and protrophs were selected. Resulting diploids having random trisomes were dissected, and the segregation patterns of pho2 and that of standard markers were compared. Thus, the possible chromosomal location of *pho2* was narrowed to chromosome *IV*. *VI* or a new chromosome. The tetrad ratio data between pho2 and trp1 were collected from crosses in which both of these two markers segregated 2+ :2-. The cumulative ratio was PD : NPD: T = 33: 6: 81. Since this ratio indicates loose linkage between pho2 and trp1, the linkage between pho2 and other markers on chromosome IV was tested. Cross W366, containing *aro1D* and *pho2*, gave the ratio PD : NPD : T = 0 : 2 : 3with respect to these markers, which showed no linkage between *aro1D* and *pho2*. Then, the linkage between pho2 and rna11 and mak21 was tested. The tetrad distribution shown in Table 1 indicates: (1) pho2 is loosely linked to trp1 and mak21, and (2) the linkage between pho2 and ma11 is closer than that between pho2 and trp1. WICKNER and LEIBOWITZ (1979) mapped mak21 between trp1 and aro1D and explained the inconsistency of map distance between mak21 and rna11 by relocating rna11 on the left arm of chromosome IV. The linkage data shown in Table 1 support this revision.

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