RECESSIVE UAA SUPPRESSORS OF THE YEAST *SACCHAROMYCES CEREVISIAE*

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ABSTRACT

Recessive lysine-independent revertants were isolated from a ψ^+ haploid strain of the yeast *Saccharomyces cerevisiae* containing one of the leucine-inserting UAA suppressors, *SUP29*, and various UAA mutations including *lys1-1*. The majority of the revertants were found to have recessive suppressors in addition to the pre-existing *SUP29* mutation. The recessive suppressors were able to suppress only a very limited number of UAA mutations, and none of the UAG mutations thus far examined. The recessive inefficient UAA suppressors were assigned to three complementation groups, *sup111*, *sup112*, and *sup113*. A high incidence of gene conversion was observed for an allele of *sup111*. An antisuppressor acting on *sup111*, but not detectably on *SUP29*, was inadvertently obtained during the course of the study. Interactions between *SUP29*, *sup111* and the antisuppressor *asu12* were studied.

NONSENSE suppression has proved to be a useful genetic phenomenon to elucidate the protein-synthesizing machinery of the yeast *Saccharomyces* cerevisiae, a eucaryotic organism, as well as of the bacterium *Escherichia coli* and other procaryotic organisms. Of various yeast nonsense suppressors, the best studied are those inserting tyrosine at UAA or UAG codons (GILMORE, STEWART and SHERMAN 1971; HAWTHORNE and LEUPOLD 1974; LIEBMAN *et al.* 1976). The tyrosine-inserting suppressors are obtained in haploid and presumed ψ^- strains. Attempts have been made to expand the range of recoverable suppressors since the detection of suppressors is limited by their efficiencies; *i.e.*, too efficient suppressors may be lethal and too inefficient suppressors may not be manifested. BRANDRISS *et al.* (1975) used ψ^- diploid strains to uncover higher-efficiency suppressors and defined a serine-inserting recessive-lethal UAG suppressor (BRANDRISS *et al.* 1976). ONO, STEWART and SHERMAN (1979 a,b); ONO *et al.* (1981b), on the other hand, used haploid ψ^+ strains in search of lower-effiency suppressors. The ψ^+ cytoplasmic determinant was first defined by COX (1965) on the basis of its ability to enhance the efficiency of a certain UAA suppressor; later, the response of various suppressors to the factor was reported (COX 1971; LIEBMAN, STEWART and SHERMAN 1975; CULBERTSON *et al.* 1977); and recently, the weak suppressor activity of the factor itself was described (LIEBMAN, STEWART and SHERMAN 1975; LIEBMAN and SHERMAN 1979).

By this means, low-efficiency serine-inserting and leucine-inserting UAA suppressors were obtained, in addition to some tyrosine-inserting UAA suppressors.

In addition to the codon-specific nonsense suppressors, there is another class of nonsense suppressors in yeast designated codon-nonspecific or omnipotent suppressors owing to the fact that they suppress UAA, UAG and presumed UGA mutations. Two loci have been identified among codon-nonspecific suppressors obtained in haploid and presumed ψ^- strains (HAWTHORNE and LEUPOLD 1974; SMIRNOV *et al.* 1974; GERLACH 1975; HAWTHORNE 1976). Although it has been thought that recessiveness is a characteristic of the codon-nonspecific suppressors, dominant suppressors acting upon UAA, UAG, and presumed UGA mutations were recently uncovered in a haploid ψ^+ strain (ONO, STEWART and SHERMAN 1981a). These codon-nonspecific suppressors are proved or suggested to be mutations of ribosomal functions (SMIRNOV *et al.* 1978; SURGUCHOV *et al.* 1980; MASUREKAR *et al.* 1981; ISHIGURO *et al.* 1981), whereas the codon-specific suppressors are mutations of tRNAs (CAPECCHI, HUGHES and WHAL 1975; GESTLAND *et al.* 1976; PIPER *et al.* 1977; GOODMAN *et al.* 1977; PIPER 1978; OLSON *et al.* 1977; ONO *et al.* 1981b).

In the present report, we describe another attempt to uncover inefficient suppressors with the aid of *SUP29*, one of the leucine-inserting UAA suppressors described by ONO *et al.* (1979b). The *SUP29* suppressor is unable to suppress the *lys1-1* UAA mutation by itself. Therefore, recessive lysine-independent revertants were selected from a haploid strain containing *SUP29*, *lys1-1* and other markers in a ψ^+ cytoplasm. In this way, we isolated recessive UAA suppressors that were inefficient by themselves.

MATERIALS AND METHODS

Yeast strains and genetic nomenclature: Strains used in the present study are listed in Table 1. Strain VF-1 is a *SUP29*-bearing revertant obtained from strain B0133-3B (**a** ψ^+ *leu2-1 lys1-1 his5-2 ura4-1 can1-100 cycl-72 trp5-48 met8-1*) as described previously (ONO *et al.* 1979b). Since the ψ^+ factor suppresses *trp5-48* and *SUP29* suppresses *leu2-1* and *ura4-1*, the strain VF-1 requires only lysine, histidine and methionine. The following nonsense mutations were used: UAA, *can1-100, his5-2, ilv1-2, leu2-1, lys1-1* and *ura4-1;* UAG, *aro7-1, met8-1* and *trp3-1*.

Yeast UAA and UAG nonsense mutations and their suppressors have been defined using the *cycl* system (SHERMAN, ONO and STEWART 1979). Nonsense mutations in other genes are now well established by their responses to such suppressors.

Codon-nonspecific suppressors are able to suppress at least some defined UAA and some defined UAG mutations. For practical reasons we used mainly *leu2-1* UAA and *met8-1* UAG mutations in order to examine the activity of suppressors, because all of the known UAA and UAG suppressors (respectively) effectively suppress these mutations (HAWTHORNE and LEUPOLD 1974; LIEBMAN, SHERMAN and STEWART 1976; ONO, STEWART and SHERMAN 1979a,b; ONO *et al.* 1981b). All of the codon-nonspecific suppressors also effectively suppress these mutations (HAWTHORNE and LEUPOLD 1974; ONO *et al.* 1981a).

Classification of suppressors as dominant or recessive is convenient and practical even though we do not yet understand the basis of the distinction (HAWTHORNE and LEUPOLD 1974; SMIRNOV *et al.* 1974; ONO *et al.* 1981a). Since dominance/recessiveness is not an intrinsic property of a suppressor, but rather the property of a combination of suppressor and suppressible mutation, we use *leu2-1* and *met8-1* as the indicators for practical purpose only.

An antisuppressor is defined as a mutation that reduces the efficiency of co-existing suppressors (MCCREADY and COX 1973). An allosuppressor is defined as a mutation that has no detectable suppressor activity of its own, but enhances the efficiency of co-existing suppressors (COX 1977).

TABLE 1

Strains and genotypes

Strain	Genotype
VF-1	**a** ψ^+ *SUP29 leu2-1 lys1-1 his5-2 ura4-1 can1-100 cycl-72 trp5-48 met8-1*
MT3-3C	α ψ *SUP29 leu2-1 lys1-1 his5-2 ura4-1 can1-100 met8-1*
B0133-3B	**a** ψ *leu2-1 lys1-1 his5-2 ura4-1 can1-100 cyc1-72 trp5-48 met8-1*
B0122-3A	α *leu2-1 lys1-1 his5-2 ura4-1 trp1-1 aro7-1*
B0393-2B	α *ade2-1 ilv1-2 leu2-1*
No. 1	α *ade6 his2*
D311-3A	**a** *his1 lys2 trp2*

Our definition of allosuppressor is again only operational: they might be reclassified as suppressors if additional mutations were examined.

Media and genetic procedures: Standard media and procedures for yeast genetics (SHERMAN, FINK and LAWRENCE 1974) were used. Canavanine resistance was tested by growth on synthetic medium containing 60 mg/l canavanine sulfate. Diploids were obtained either by marker selection on agar plates or by zygote isolation by micromanipulation. Special care was taken during replica plating, since the expression of weak suppressors and suppressor modifiers occasionally depended on the number of cells transferred. Dilute cell suspensions were inoculated as uniform spots using a 32-point inoculator (LAWRENCE *et al.* 1975). Growth was evaluated after 2, 4 and 7 days of incubation at 28°. Three levels of growth were scored; good (+), intermediate (±) and poor or negative (−). Mating types were tested by complementation with strains No. 1 (α *ade6 his2*) and D311-3A (**a** *his1 lys2 trp2*).

RESULTS

Isolation and classification of revertants: Cells of strain VF-1 were spread on synthetic complete agar medium lacking lysine and were mutagenized by UV irradiation at a dose producing about 50% survival. Colonies appearing after 10 to 14 days of incubation at 28° were subcloned and used for further analysis; the frequency of reversion was approximately 7×10^{-6}. Revertants were then screened for recessive lysine independence. They were first crossed to strain B0122-3A, which is free of suppressor mutations. Diploids were selected and tested for their lysine requirement. Approximately half of the revertants were recessive. Dominant revertants were excluded from further analysis because they probably arose by the intragenic reversion of *lys1-1* or by dominant suppressors acting upon *lys1-1*.

Seventy-three recessive revertants were examined for their pattern of suppression (Table 2). Three classes were distinguished among these revertants. Class 1 revertants were completely independent of lysine and histidine, class 2 revertants were completely independent of lysine but only partially independent of histidine, and class 3 revertants were partially independent of lysine and dependent on histidine. The majority of revertants (93%) belonged to class 1. Only the revertants of this class were further analyzed in the present study. Some of class 2 revertants demonstrated abnormalities in mating and sporulation. Therefore, their genetic analysis has not been straightforward and is not yet completed. Our preliminary analysis indicates that they contain class(es) of inefficient suppressors different from those described here; more description

TABLE 2

Classification of recessive lysine-independent revertants obtained from strain VF-1

Strain	Growth on			No. of revertants
	SC-leucine	SC-lysine	SC-histidine	
B0133-3B	−	−	−	
VF-1	+	−	−	
Revertants				
Class 1	+	+	+	67
Class 2	+	+	±	5
Class 3	+	±	−	1

will be presented elsewhere. Class 3 revertant did not sporulate after crossing, and genetic analysis was therefore impossible.

Segregation of recessive inefficient UAA suppressors from class 1 revertants: Representatives of class 1 revertants were crossed to strain B0122-3A, the diploids were sporulated and the tetrad was analyzed (Table 3). Normal single-factor segregation of *SUP29* was observed by the 2+:2− segregation of uracil dependence. However, leucine dependence segregated 4+:0− and 3+:1− as well as 2+:2−. There were two levels of leucine independence. Only two segregants in each tetrad grew well on medium lacking leucine, and these segregants were always uracil-independent, indicating the presence of *SUP29*. Partial suppression of *leu2-1* indicated the presence of new inefficient suppressors. Such suppressors were recessive since diploids made with partially leucine-independent segregants and B0133-3B or B0122-3A were leucine-dependent. In conclusion, recessive inefficient suppressors segregated independently of *SUP29* in the crosses shown in Table 3.

Segregation of lysine and histidine independence always coincided in tetrads (Table 3). In addition, all segregants independent of lysine and histidine were completely leucine-independent, indicating the presence of allosuppressors interacting with *SUP29*. The identity of the allosuppressors and the recessive inefficient suppressors was suggested from the segregation pattern, *i.e.*, the segregation pattern was compatible with the involvement of two factors, *SUP29* and a new suppressor which together with *SUP29* caused the increased level of suppression. The segregation of the two factors is indicated in Table 3 by PD, T and NPD, representing parental ditype, tetratype and nonparental ditype tetrads, respectively. It should be noted that a few tetrads had segregation patterns incompatible with two-factor segregation; the nature of such tetrads will be discussed later.

A further evidence for the identity of the allosuppressors and the recessive inefficient suppressors was obtained by so-called reconstitution crosses. One segregant (MK18-14D) having the partial suppression of *leu2-1* was crossed to the strain VF-1. Another segregant (MK18-12B) having the suppression of *lys1-1* and *his5-2* was crossed to B0133-3B. Diploids of these crosses were sporulated and tetrad analyzed (Table 4). Segregation of these crosses and that shown in Table 4 were essentially indistinguishable, suggesting the involvement of the

TABLE 3

Segregation of SUP29 *and the weak recessive UAA suppressor*

Segregation pattern[a]				Revertant crossed to B0122-3A[c]				
Uracil	Leucine	Lysine histidine	Tetrad-type[b]	LC5-15 *sup111*	LC5-7 *sup111*	LC3-16 *sup111*	LC3-15 *sup112*	LC1-4 *sup113*
+:−	+:±:−	+:−						
2:2	2:0:2	2:2	PD	3	1	2	5	4
		1:3		2	0	0	0	0
		0:4		0	0	0	0	0
2:2	2:1:2	2:2		0	0	0	0	1
		1:3	T	5	5	4	5	2
		0:4		0	0	0	0	0
2:2	2:2:0	2:2		0	0	0	0	0
		1:3		0	0	0	0	0
		0:4	NPD	2	1	1	0	2
				12	7	7	10	9

[a] All of the uracil-independent segregants were leucine-independent, and all of the lysine-independent segregants were leucine-dependent.

[b] PD, T and NPD represent parental ditype, tetratype and nonparental ditype tetrads, respectively, according to the two-factor segregation of SUP29 and a new weak suppressor (see text).

[c] The newly arisen recessive suppressor locus defined in each revertant is indicated.

same genes in these crosses. The newly identified recessive inefficient suppressor in revertant LC5-15 was designated as *sup111*. Characteristics of *sup111* are shown in Table 5. It was recessive as a suppressor and caused increased efficiency of *SUP29*. This interaction with *SUP29* was also recessive. Action of *sup111* was limited to the *leu2-1* and *ilv1-2* UAA mutations among the nonsense mutations listed in MATERIALS AND METHODS.

Complementation analysis of class 1 revertants: Thirty-two class 1 revertants were subjected to complementation analysis. They were crossed with MK18-1C (containing *sup111* but not *SUP29*) and the resulting diploids were examined for leucine independence. Eight revertants produced lysine-independent diploids, indicating that they contained alleles of *sup111*, whereas 24 revertants produced lysine-dependent diploids. One of the latter group, LC3-15, was crossed to B0122-3A in order to construct a strain containing the second recessive inefficient suppressor, *sup112*. Segregant MT34-1A was such a strain and was used for the second round of complementation analysis. This time, 18 revertants did not complement and 14 complemented. Those that did not complement with the *sup111* strain complemented this time. The third recessive inefficient suppressor, *sup113*, was obtained from LC1-4, which complemented with both the *sup111* and the *sup112* strains. The *sup113* strain, MT33-2A, did not complement with the revertants that complemented with both the *sup111* and the *sup112* strains, but complemented with the remaining revertants. In summary, we could distinguish three complementation groups among 32 re-

TABLE 4

Segregation of SUP29 *and* sup111 *in different crosses*

Segregation[a]		Cross					
Leucine	Lysine	LC5-15 × B0122-3A		MK18-14D × VF-1		MK18-12B × B0133-3B	
		SUP29 sup111[b]		+ *sup111*		*SUP29 sup111*	
+:±:−	+:−	+	+	*SUP29*	+	+	+
2:0:2	2:2[c]	3		5		3	
	1:3	2		1		0	
	0:4	0		0		0	
2:1:1	2:2	0		1		0	
	1:3[c]	5		13		8	
	0:4	0		1		1	
2:2:0	2:2	0		0		0	
	1:3	0		0		2	
	0:4[c]	2		4		5	

[a] Uracil dependence segregated 2+:2−, and all of the uracil independent segregants were leucine-dependent. All of the lysine-independent segregants were leucine-independent.

[b] Cross shown in Table 4.

[c] Tetrads expected from normal two-factor segregation of *SUP29* and *sup111*.

vertants containing the new recessive UAA suppressors. The distribution of revertants among each group strongly indicated that there were only three loci in the yeast genome giving rise to the suppressors in this category. No significant linkage was detected among these loci (data not shown).

Extremely high frequency of gene conversion for sup111-1: A few exceptional tetrads that were not explicable by two-factor segregation were noted above. Table 6 is a summary of the exceptional tetrads found in different crosses containing *sup111-1* (derived from revertant LC5-15) and *SUP29*. Of 56 tetrads, 8 (14%) were exceptional. At first these tetrads were thought to be the result of more than two segregating loci. For example, the lysine and histidine independence of segregant 1A (Table 6) was an indication that it contained an allosuppressor different from *sup111*, because segregants 1C and 1D contained *sup111*. In another example, the absence of suppression of *leu2-1* on segregant 3D was an indication that it contained an antisuppressor acting upon *sup111*, because segregants 3A and 3B did not contain *sup111*; and so on. Each of the segregants in exceptional tetrads was test-crossed to strains with or without *SUP29* in order to verify the genotype. The revealed genotypes are indicated in Table 6. In every case, the apparent abnormality was attributable to gene conversion of *sup111-1*. The frequency of 14% was rather high compared to the frequency of gene conversion of an average yeast marker (MORTIMER and HAWTHORNE 1975). The gene conversion frequency of sup111-1 was also examined in diploids of *SUP29/SUP29* and $sup29^{+}/sup29^{+}$, where the segregation of *sup111-1* was observed directly (Table 7). It was apparent that these gene conversion frequencies were about the same in different crosses. There was no preferential direction of conversion.

TABLE 5

Interaction between SUP29 *and* sup111

Genotype	Suppression of		
	leu2-1	*lys1-1*	*his5-2*
SUP29 +	+	−	−
+ *sup111*	±	−	−
SUP29 sup111	+	+	+
$\frac{+\ sup111}{+\ +}$	−	−	−
$\frac{+\ sup111}{+\ sup111}$	±	−	−
$\frac{SUP29\ sup111}{SUP29\ +}$	+	−	−
$\frac{SUP29\ sup111}{+\ sup111}$	+	+	±, +
$\frac{SUP29\ sup111}{SUP29\ sup111}$	+	+	±, +

TABLE 6

Analysis of exceptional tetrads

Segregation pattern	Spore	Growth on			Genotype	No. of tetrads[a]
		SC-leu	SC-lys	SC-his		
1	A	+	+	+	*SUP29 sup111*	
	B	+	−	−	*SUP29* +	
	C	±	−	−	+ *sup111*	
	D	±	−	−	+ *sup111*	2
2	A	+	+	+	*SUP29 sup111*	
	B	+	+	+	*SUP29 sup111*	
	C	±	−	−	+ *sup111*	
	D	−	−	−	+ +	1
3	A	+	−	−	*SUP29* +	
	B	+	−	−	*SUP29* +	
	C	±	−	−	+ *sup111*	
	D	−	−	−	+ +	2
4	A	+	+	+	*SUP29 sup111*	
	B	+	−	−	*SUP29* +	
	C	−	−	−	+ +	
	D	−	−	−	+ +	3

[a] Tetrads of the three crosses shown in Table 4 were pooled.

It should be mentioned that a high frequency of gene conversion is a unique feature of the *sup111-1* allele, and not of the *sup111* locus. The alleles of *sup111* present in revertants LC5-7 and LC3-16 did not demonstrate this abnormal segregation.

Detection of an antisuppressor acting on sup111: In the cross between MT3-3C and MK18-5B, which was heterozygous for both *SUP29* and *sup111*, a tetrad was found to contain segregants independent of leucine and lysine but depend-

TABLE 7

Gene conversion of sup111-1 *in different crosses*

Configuration of *SUP29*	Segregation of *sup111-1*				
	4:0	3:1	2:2	1:3	0:4
SUP29/+[a]	0	3	48	5	0
SUP29/SUP29	0	2	22	1	0
+/+	0	1	18	1	0

[a] Result shown in Table 6.

ent on histidine. All other segregants of the cross (and of other crosses with the similar genetic constitution) were histidine-independent if independent of leucine and lysine (Table 8). These unusual segregants were crossed to LC5-15. The resulting diploids were histidine-independent, indicating the presence of recessive antisuppressors in these segregants. The diploids were then analyzed for tetrad composition. In each case, single-factor segregation of the antisuppressor was revealed because histidine independence segregated 2+:2−. These antisuppressors were separated from *SUP29* and *sup111* after consecutive crosses. Antisuppressors in the two unusual segregants were allelic by complementation tests. The antisuppressor was designated *asu12*.

DISCUSSION

In the previous study (ONO *et al.* 1979a,b), we demonstrated the additivity of low-efficiency UAA suppressors. This property led us to utilize the low-efficiency suppressors to uncover highly inefficient suppressors. In the present study, we obtained recessive inefficient UAA suppressors which, together with *SUP29* (one of the leucine-inserting UAA suppressors), resulted in increased levels of suppression. Whether the interaction of these suppressors with *SUP29* is additive or synergistic is not known.

An examination of suppression patterns established that there is a gradient of suppressibility among the UAA mutations we used: (*leu2-1, ilv1-2*) > *ura4-1* > *lys1-1* > *can1-100* > *his5-2* (Table 9). Apparently, *asu12* did not detectably reduce *SUP29* activity. The action of *asu12* may be specific to *sup111*, or it may be too small to be detected by the interaction with *SUP29* (*i.e.*, the suppression of *leu2-1* and *ilv1-2* by *sup111* is marginal, and a small reduction of efficiency due to *asu12* results in loss of suppression, whereas a small reduction of efficiency of *SUP29* caused by *asu12* does not result in loss of suppression of *leu2-1* and *ura4-1*). Similarly, the suppression of *his5-2* by the combination of *SUP29* and *sup111* is marginal and is readily lost by a small reduction of efficiency caused by *asu12*.

The new suppressors we uncovered are UAA suppressors by our operational definition because they suppress only *leu2-1* and *ilv1-2* among the various nonsense mutations examined. Another characteristic of these suppressors was their recessiveness; this, however, may result from their inefficiency. If the efficiency of their suppression of *leu2-1* is marginal in haploid cells, their

TABLE 8

Occurrence of asu12 *in a cross heterozygous for both* SUP29 *and* sup111

Tetrad type	Growth on SC-leu	SC-lys	SC-his	Genotype	No. of tetrads[a]
Parental ditype	+	−	−	*SUP29* +	
	+	−	−	*SUP29* +	
	±	−	−	+ *sup111*	
	±	−	−	+ *sup111*	4
Tetratype	+	+	+	*SUP29 sup111*	
	+	−	−	*SUP29* +	
	±	−	−	+ *sup111*	
	−	−	−	+ +	18
Nonparental ditype	+	+	+	*SUP29 sup111*	
	+	+	+	*SUP29 sup111*	
	−	−	−	+ +	
	−	−	−	+ +	3
Unusual	+	+	−	*SUP29 sup111 asu12*	
	+	+	−	*SUP29 sup111 asu12*	
	−	−	−	+ + +	
	−	−	−	+ + +	1

[a] Tetrads containing gene conversion of *sup111-1* were excluded.

reduced efficiency in heterozygous diploid cells may result in the failure of suppression. It has been reported that a tyrosine-inserting UAA suppressor, *SUP11*, is dominant for the suppression of *leu2-1*, *lys1-1*, *his5-2* and other UAA mutations, but recessive for the suppression of the UAA mutation *ade2-1* (HAWTHORNE and LEUPOLD 1974). A codon-nonspecific suppressor, *SUP46* is dominant for the suppression of *leu2-1* and *met8-1* mutations, but its suppression of *lys1-1* and *his5-2* is reduced or lost in heterozygous diploids (ONO, STEWART and SHERMAN 1981a; ISHIGURO *et al.* 1981).

Previously described UAA suppressors have varying efficiencies of suppression as estimated by suppression spectra and action on defined *cyc1* mutations (HAWTHORNE and LEUPOLD 1974; ONO, STEWART and SHERMAN 1979a,b; ONO *et al.* 1981b). There is a striking correlation between classifications based on suppressor efficiency and those based on amino acid insertion; the order of efficiency is tyrosine-insertors > serine-insertors > leucine-insertors (SHERMAN, ONO and STEWART 1979). All of these suppressors suppress *leu2-1* effectively in both haploid and diploid cells. By this criterion, the new suppressors constitute a unique and least efficient class of UAA suppressors. If the new suppressors are mutations of tRNAs as are other UAA suppressors, they are probably mutations of $tRNA^{Gln}$, $tRNA^{Lys}$, and/or $tRNA^{Glu}$. Because, these tRNAs are expected to generate UAA suppressors by single-base substitutions in their anticodons, but no such suppressor has been identified as yet. However, we consider the presence of complementation groups among the new suppressors

TABLE 9

Interactions among SUP29, sup111, *and* asu12

Genotype	Suppression of					
	leu2-1	*ilv1-2*	*ura4-1*	*lys1-1*	*can1-100*	*his5-2*
+ + *asu12*	−	−	−	−	−	−
+ *sup111 asu12*	−	−	−	−	−	−
+ *sup111* +	+	+	−	−	−	−
SUP29 + +	+	+	+	−	−	−
SUP29 + *asu12*	+	+	+	−	−	−
SUP29 sup111 asu12	+	+	+	+	+	−
SUP29 sup111 +	+	+	+	+	+	+

as a strong argument against this possibility. It is very unlikely that tRNA suppressors compose complementation groups. Our speculation is that these new suppressors are mutations of cellular components other than tRNAs involved in protein synthesis.

It has long been believed that codon-nonspecific suppressors are distinct from codon-specific suppressors, not only in their patterns of codon recognition, but also in their recessiveness (HAWTHORNE and LEUPOLD 1974; SMIRNOV *et al.* 1974; GERLACH 1975). However, dominant codon-nonspecific suppressors have recently been described (ONO *et al.* 1981a). The difference between dominant and recessive codon-nonspecific suppressors may simply be caused by their different efficiencies. This concept may be extended to the distinction between codon-nonspecific and codon-specific suppressors. Suppose the suppressibilities of *leu2-1* and *met8-1* differ because some inefficient suppressors are unable to suppress one of them: such suppressors would then be operationally designated UAA or UAG codon-specific suppressors. Our hypothesis is that our newly described recessive suppressors are highly inefficient members of codon-nonspecific suppressors. This hypothesis predicts that these suppressors will act better upon *leu2-1* than upon *met8-1*. If this is the case, then these suppressors may well be mutations of ribosomal functions, as are codon-nonspecific suppressors.

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