Thus the nucleoprotein core of Sindbis virus seems to be surrounded by a layer of lipid, which may project the core protein from proteolytic enzymes. The finding that the glycoprotein can be completely removed without destroying the integrity of the viral membrane indicates that the glycoprotein is located outside of, rather than within, the lipid layer. The membrane in particles devoid of spikes is sufficiently stable to withstand centrifugation in a potassium tartrate gradient; in the intact virion, the glycoprotein may add to the stability of the membrane. The marked aggregation of particles devoid of glycoprotein suggests that the carbohydrate provides a hydrophilic surface layer which reduces clumping of virus particles in aqueous suspension.

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[1] Strauss, J. H., Burge, B. W., Pfefferkorn, E. R., and Darnell, J. E., *Proc. US Nat. Acad. Sci.*, **59**, 533 (1968).
[2] Strauss, J. H., Burge, B. W., and Darnell, J. E., *J. Mol. Biol.*, **47**, 437 (1970).
[3] Acheson, N. H., and Tamm, I., *Virology*, **32**, 128 (1967).
[4] Simpson, R. W., and Hauser, R. E., *Virology*, **34**, 568 (1968).
[5] Strauss, J. H., Burge, B. W., and Darnell, J. E., *Virology*, **37**, 367 (1969).
[6] Caliguiri, L. A., Klenk, H.-D., and Choppin, P. W., *Virology*, **39**, 460 (1969).
[7] Acheson, N. H., and Tamm, I., *Virology*, **41**, 306 (1970).

Prototrophic Regulatory Mutants of Adenylosuccinate Synthetase in Yeast

MUTANTS of the genes *ade*12 and *ade*13 in *Saccharomyces cerevisiae* require adenine, which suggests that they are defective in the conversion of inosine monophosphate (IMP) to adenosine monophosphate (AMP). Two enzymes are required for this conversion: adenylosuccinate lyase and adenylosuccinate synthetase. The former has been shown to be absent in mutants of *ade*13 (ref. 1) so that one can infer that *ade*12 specifies the latter. Mutants of *ade*12 have been isolated on the basis of an adenine insensitive pigment accumulation by strains carrying *ade*1 or *ade*2 (refs. 1 and 2). They seem to synthesize purines constitutively and excrete hypoxanthine[1]. These observations suggest that in addition to its catalytic function adenylosuccinate synthetase has an important function in the regulation of purine synthesis.

Prototrophic mutants of yeast defective in the control of purine synthesis have been isolated and characterized in this laboratory[3,4]. These mutants have been assigned to six unlinked loci, *pur*1 to *pur*6. They excrete a mixture of hypoxanthine, inosine and xanthine (our unpublished work) and like *ade*12 mutants can consequently crossfeed to purine-requiring auxotrophs. Mutant alleles of the loci *pur*1, *pur*3, *pur*4 and *pur*6 also cause adenine insensitive pigment accumulation in strains of the genotype *ade*2 *pur*. Because these mutants and *ade*12 have a number of properties in common we thought it important to test them for allelism.

The techniques and media that we used have been described previously[4]. A representative mutant of each *pur* gene was crossed with the mutant *ade*12-*M*8, donated by Dr Susan Armitt, and the resulting diploids and ascospore progeny were tested for purine excretion and adenine requirement (Table 1). The genetic analysis was hindered by poor ascospore germination in some of the crosses so we have summed data from complete and incomplete tetrads in the table. The presence of prototrophic non-crossfeeding recombinants in any *ade*12 × *pur* cross indicates that the loci are not allelic.

Table 1 Functional and Genetic Tests for Allelism between *ade*12 and the *pur* Genes

Cross	Purine excretion by diploid	No. asci	Segregants analysed	Non-crossfeeding prototrophic recombinants
ade12 × pur1	+	40	144	0
ade12 × pur2	—	21	70	16
ade12 × pur3	—	24	85	24
ade12 × pur4	—	8	23	9
ade12 × pur5	—	17	65	13
ade12 × pur6	+	28	107	26
ade12 × PUR6	—	15	60	12

Although all of the diploids are prototrophic those involving *ade*12 in combination with *pur*1, *pur*3 and *pur*6 are phenotypically mutant in that they excrete purine. The genetic analysis, however, indicates free recombination between *ade*12 and all of the *pur* loci except *pur*1. The results of the functional tests with *pur*3 and *pur*6 are thus anomalous and indicate some kind of interaction between gene products. Similar anomalous results are obtained in diploids containing *pur*1 in combination with *pur*3 and *pur*6 (refs. 3 and 4).

Mutants of *ade*12 and *pur*1 are closely linked, are non-complementary with respect to purine excretion and have other phenotypic characteristics in common and we feel justified in concluding that they are lesions of the same gene. The *pur*1 phenotype would thus represent an alteration of adenylosuccinate synthetase in which the enzyme has lost its regulatory function but retained catalytic activity, whereas in *ade*12 mutants both the catalytic and regulatory functions of the enzyme are impaired. If this is true then one would predict the possibility of obtaining prototrophic revertants of *ade*12 which have regained the catalytic function but still synthesize and excrete excess purine. This prediction has been confirmed in this laboratory and by Dorfman[5].

Our results with the *ade*12 *pur*1 system add weight to the suggestion that adenylosuccinate synthetase acts as a regulatory protein. Analogous properties have been proposed for several other enzymes, for example, anthranilate synthetase in *Escherichia coli*[6], phosphoribosyl-ATP-pyrophosphorylase in *Salmonella typhimurium*[7] and nitrate reductase in *Aspergillus nidulans*[8]. The anomalous interactions in diploids shown by both *ade*12 and *pur*1 in combination with other *pur* mutants suggest that the regulatory function of adenylosuccinate synthetase may be exerted through a complex involving the products of at least three genes. If this is so, the genetic control of purine biosynthesis in yeast seems to be extremely complex and its study may yield new insight into the regulatory systems of eukaryotes.

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[1] Dorfman, B., *Genetics*, **61**, 377 (1969).
[2] Woods, R. A., and Armitt, S., *Heredity*, **24**, 467 (1968).
[3] Armitt, S., and Woods, R. A., *Heredity*, **24**, 468 (1968).
[4] Armitt, S., and Woods, R. A., *Genet. Res., Camb.*, **15**, 7 (1970).
[5] Dorfman, B., Goldfinger, B. A., Berger, M., and Goldstein, S., *Science*, **168**, 1482 (1970).
[6] Somerville, R. L., and Yanofsky, C., *J. Mol. Biol.*, **11**, 747 (1965).
[7] Kovach, J. S., Phang, J. M., Ference, M., and Goldberger, R. F., *Proc. US Nat. Acad. Sci.*, **63**, 481 (1969).
[8] Cove, D. J., and Pateman, J. A., *J. Bact.*, **97**, 1374 (1969).