prototrophic. Tetrad analysis in conjunction with Southern blotting revealed that transformation had mainly occurred through the integration of the entire hybrid plasmid at homologous chromosomal sites (principally at the *leu*2 locus, although also at other sites which contain a repeat sequence present on the plasmid). Furthermore, the integrated hybrid plasmid could be recovered intact by excision with a restriction endonuclease followed by transformation of colicin E sensitive *E. coli*. Thus, this system could, in theory, be exploited as a primitive cloning vehicle. In fact, its present value as such is limited not only by the low frequency of transformation obtained but also because the introduced DNA might be altered in the process of integration. Such is the pace of research in this field that both of these problems have already been elegantly solved, with the result that at least two types of versatile vector may very shortly be on the market.

The first of these stems from the observation that the frequency of transformation of different loci by similar *E. coli* plasmids containing different inserts varies considerably. For instance, J. Carbon (University of California) has found that while plasmids containing *leu*2 or *his*3 gave frequencies in the order of 10^{-6} to 10^{-7}, those containing *arg*4 or *trp*1 yielded transformants in the order of 10^{-4}. These differences have been more thoroughly investigated and exploited by Davis's group. They found that the plasmid pBR322 containing *his*3 gave a low level of transformants, in all of which the plasmid had been integrated at the *his*3 locus. However, if a fragment containing *trp*1 was added, then the frequency of *his* transformants was increased by a factor of 10^2 or 10^3 (all of them were also transformed for *trp*). Surprisingly, the introduced plasmid was not integrated but present as a closed circular molecule, at a concentration of approximately one per cell. The tentative explanation for this phenomenon is that the *trp*1, but not the *his*3, fragment possesses a DNA replication origin. It would seem that the replication of the *his*3 fragment (and the bacterial plasmid sequence joined to it) is conditional on integration, whereas any hybrid containing the *trp*1 fragment can replicate autonomously. However, there is at present no independent evidence that specific replication origins exist in yeast or indeed in other eukaryotic organisms.

The initial interest in *trp*1 stemmed from its proximity to the centromere of chromosome IV. It is not yet known whether a centromere is present on the plasmid nor, if it is, whether it is important for the plasmid's autonomy and stability. If it is active, then the plasmid may be behaving as a minichromosome. Irrespective of the mechanism, this phenomenon can be usefully exploited. A similar *trp*1 fragment has been inserted into λ to form a hybrid that will also replicate as an autonomous closed circular molecule once introduced into yeast. Thus, a new breed of hybrid vector has been obtained that will not only transform *trp* yeast strains at a high frequency but will also form viable plaques (even from crude lysates of yeast) in *E. coli*. It is Davis's intention to replace the repressor region of λ by the *trp*1 fragment, thereby freeing its inessential central region for cloning, with all its advantages for insert selection.

Yeast 2 μm plasmid

The second type of vector has been developed using the yeast 2 μm plasmid. This molecule is found in nearly all strains, at 50 to 100 copies per cell. Although there is continuing debate as to whether it is located in the cytoplasm or in the nucleus, it is known that its replication is subject to the same controls as those for nuclear DNA (Livingston *Genetics* **86**, 73; 1977) and is confined to the beginning of S phase (V. Zakian & W. Fangman, University of Washington). Several transcription products from the plasmid have been identified (J. Broach, C. McGill & J. Atkins, Cold Spring Harbor Laboratory) but there is no evidence for its function. For instance, no genes have been conclusively linked to it. Despite this ignorance, several groups (Davis; G. R. Fink, Cornell University; J. D. Beggs, Plant Breeding Institute, Cambridge) (*Nature* in the press)) have constructed, in *E. coli*, hybrid molecules composed of the yeast plasmid, an *E. coli* plasmid, and a wild type yeast gene such as *his*3 or *leu*2. Such mongrels will transform appropriate yeast auxotrophs at a very high frequency; in one case as high as 3×10^{-3}. The transformants possess multiple free copies of the introduced plasmid and, in some cases at least, also a single integrated copy. The state of prototrophy of all transformants is inherited in a non-Mendelian manner (as is the resident plasmid) although with different degrees of both mitotic and meiotic stability depending on the type of composite plasmid. Here too, the transforming plasmid can be easily recovered in the appropriate strains of *E. coli* by selecting for either *E. coli* plasmid-specified antibiotic resistance or yeast DNA-specified prototrophy after transformation with yeast extracts. In its present form, the vector is still in its infancy, but may soon be streamlined by removing all sequences other than those strictly necessary as a marker for transformation of both *E. coli* and yeast (for example the yeast *leu*2 gene) and the replication origins necessary for growth in both organisms.

The outstanding features of both the *trp*1 and the 2 μm plasmid based vectors are that they can just as readily be grown in *E. coli* as in yeast, thereby retaining the different opportunities that each offers; principally, the use of *E. coli* to construct gene banks and to amplify sequences, and yeast to study gene function. Possibly, both types of hybrid vector reside in the nucleus whilst in yeast, thus enjoying an appropriate environment for the study of gene control. One can envisage one possible drawback to the use of these vectors. This is that due to their wider host range, which may indeed extend to other eukaryotes, they may be considered as a greater biohazard than present vectors.

The immediate implications of this work will of course mainly concern yeast geneticists. Given the high rates of transformation, one may now confidently select for any yeast gene for which there is a mutant to be complemented. There are many interesting genetic loci, such as the mating type locus and those involved in the cell cycle and meiosis, for which no gene products, let alone a hybridisation probe, are presently assigned or available. Their DNA sequence may now be directly obtained. For such loci, it is going to become, ironically, much easier to isolate their genes and study the control of their expression than to understand the function of their gene products.

In the longer term lies the potential for using yeast vectors for studying the expression of genes from higher eukaryotes. It remains to be seen whether such genes will indeed function in the yeast environment. There is hope as far as transcription is concerned, for yeast possesses a typically eukaryotic spectrum of RNA polymerases. On the other hand, it is less certain whether

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Erratum

In the article 'Lunar atmosphere past and present' (*News and Views* **273**, 489; 1978) the last sentence in paragraph 2 of column 3 should read "A crater *1280 m* across and 180 m deep", and not ". . . 128 m across . . .". In paragraph 3, column 3, line 16 should read . . . "It is highly unlikely that the mass distribution of the cosmic dust cloud has changed considerably in the past 10^8 yr," and not . . . "10 yr . . .".