news and views

Transferring foreign genes to plants

It is well established that purified DNA can be taken up by bacteria and that the recipient cells are capable of incorporating DNA into their chromosomes and expressing the genes thus acquired. Attempts to perform similar experiments with eukaryotes, however, are often viewed with scepticism although an increasing number of workers are investigating this problem.

In their simplest form these experiments involve the use of two closely related organisms which differ with respect to an easily identifiable phenotypic characteristic. Attempts are then made to transfer this character to individuals that lack it by applying purified DNA from the appropriate donor. The experiments of Hess involving white and red flowered plants of *Petunia* fall into this category (Hess, *Z. Pflanzenphysiol.*, **60,** 348-358; 1969; **61,** 286-298; 1969; **63,** 461-467; 1970; **68,** 432-440; 1973). In some instances the test organism has a deficiency in an essential metabolic pathway and corrected individuals are selected by their ability to grow without the appropriate nutritional supplement. In many experiments the donor DNA is derived from organisms totally unrelated to the recipient and evidence for uptake and possible incorporation into host chromosomes has been obtained. Occasionally bacteriophages have been used as an enriched source of specific genes and there is evidence for the synthesis of enzymes coded for by the phage DNA (Carlson, *Proc. natn. Acad. Sci. U.S.A.*, **70,** 598-602; 1973).

To establish that true transformation is possible in eukaryotes, it is necessary to study the fate of applied DNA by biochemical techniques, to show that changes in the recipient organism are directed by the donor DNA and to demonstrate that these changes are inherited. Ledoux and his colleagues have been carrying out some of these experiments for more than a decade and have obtained some remarkable results (Ledoux and Huart, *Eur. J. Biochem.*, **23,** 96-108; 1971). In this issue they present data which suggest that thiamine deficiency in *Arabidopsis thaliana* may be corrected by bacterial DNA and furthermore that this correction can be inherited.

Arabidopsis is particularly suitable for such experiments. It is comparatively small and easy to manipulate and grow aseptically and it flowers within a few weeks of germination. Ledoux and his team used a number of lines with lesions in the branched pathway for thiamine biosynthesis. These plants normally require either exogenous thiamine, pyrimidine or thiazole for continued growth and reproduction but they found that a small number of the plants grew and produced seed in the absence of supplements after they had been treated with the appropriate bacterial DNA.

Ledoux states that DNA with a molecular weight in excess of 10^7 is necessary and the DNA solution should be applied to the ungerminated seeds during imbibition. DNA-corrected plants grow more slowly than supplemented mutants and only about one third of those that grow actually produce seed. The observed frequency of correction can be as high as 10^{-2} compared with a spontaneous reversion rate of the *Py* locus of less than 5×10^{-6}, and $4\times^{-5}$ for reversion induced by ethyl methane sulphonate. DNA from phage T7 and 2C is ineffective and DNA from *Escherichia coli* unable to produce the thiazole moeity of thiamine does not correct the corresponding *Arabidopsis* mutant although it is effective with plants deficient in the pyrimidine pathway.

Seeds from corrected plants were germinated and selfed to produce F_1 to F_3 progeny. Although these plants showed some variegation in chlorophyll content no typical thiamine-deficient segregants were observed. This suggests that the parental lines are homozygous which is a rather surprising result although it should be noted that Hess observed a similar situation with *Petunia*. Ledoux suggests that in the case of *Arabidopsis*, during meiosis and afterwards, the gametes containing the information for thiamine production are favoured over mutant gametes. In support of this proposal he cites the finding that if during DNA correction plants showing signs of abortive flowering are supplemented with thiamine then lethal mutants appear in the F_1. Although this result would also be expected in the absence of applied DNA, in this case no corrected plants would be anticipated. In crosses between progeny of DNA-corrected plants and either wild type or mutant individuals, lethal mutants and a variety of leaky mutants are obtained. There is no evidence for cytoplasmic inheritance of the corrected character.

Obviously the mechanism involved in these DNA-mediated corrections is of major interest, but it would be premature to attempt a deailed interpretation. From the figures quoted by Ledoux the frequency with which the corrections are obtained is too high to be explained by spontaneous reversion of the mutation. On the other hand direct evidence for the synthesis of a bacterial enzyme in these plants is not available at present. Other experiments by the Mol group suggest that foreign DNA can become integrated in some way into plant DNA although the significance of these findings has been questioned (Hotta and Stern, in *Informative Molecules in Biological Systems* (edit. by Ledoux) 176–178, North Holland; 1972). One thing is certain; experiments of this nature hold out intriguing possibilities and they will continue to generate interest and occasionally heated discussion.

From our Plant Cell Physiology Correspondent

Number of cells at the time of X activation

ALTHOUGH female mammals possess two X chromosomes, in any particular adult cell only one of these homologues is active, providing a dosage compensation which ensures that females and males both display the same number of active-X-linked genes. The fertilised egg and very early embryo of eutherian mammals, including man and mouse, possess two active X chromosomes; that one of these two chromosomes is randomly inactivated in each cell at an early stage of genetic development was first suggested by Lyon (*Nature*, **190,** 372; 1961).

The random nature of the process is suggested by the expression of sex linked genes in adult females. In heterozygotes in which one X chromosome carries a wild type and the other carries a mutant allele, those cells in which the wild type X chromosome is active must show wild phenotype whereas cells instead possessing the active mutant X chromosome display the mutant phenotype. One visible