University of Rajasthan for three years under the system of rotation, two shifts of M Sc classes were being run with only one set of laboratories in the department. This system was not satisfactory and had its own drawbacks. I reduced the admissions by fifty per cent and managed with one normal shift. There was a lot of hue and cry from the teachers of the department and students in general. Though things returned to their original status of two shifts after my term, I was satisfied that I did what little I could to save higher education from being reduced to mass education.

More than sixty per cent women compared to men do postgraduation and research in science subjects. It is also true that eventually most of them do not take up a career in science. We have to find a way so that higher education for women does not suffer and we also get returns for the funds invested. 1. Lakhotia, S. C., Curr. Sci., 2005, 88, 1731–1735.

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## **Apomixis revisited**

The discussions recently published in Current Science, as reviews on apomixis in higher plants, by Kaushal et al.1 and Bhat *et al.*<sup>2</sup>, are valuable contributions for generating new ideas in this agriculturally important area. Besides describing the characteristics of the various forms of apomixis, the authors<sup>1,2</sup> have also attempted to identify techniques that can be used to observe the occurrence of apomixis, and features to be present in model systems, as also approaches that should be employed in these to understand the biology of apomixis in molecular terms. The authors have reached the conclusion that, to unravel the enigma of apomixis (formation of seeds having maternal genotype without fertilization), detailed knowledge of the molecular biology of plant reproductive process is a prerequisite for its apomictic manipulation. They have advised that new experimentation on apomixis be directed in Arabidopsis and rice plants and it be designed to reveal the regulation of meiosis, endosperm development and parthenogenetic or apomictic embryo development. They have opined that knowledge-based deregulation of the key regulatory step(s) of the above-mentioned plant processes might help engineer apomixis in these systems. The conclusion is unquestionable. Indeed, in the long term, the conventional forwardand reverse-genetics strategies should help in the achievement of these objectives.

The articles evoke the question whether the already available information about plant embryogenesis, especially apomixis type and genetics in the species of occurrence, could be used to select apomictic mutants in plants that have normal or wild-type sexual reproduction. Since the purpose of the articles of Kaushal *et al.*<sup>1</sup> and Bhat *et al.*<sup>2</sup> appears to further the evolution of ideas and experimentation on apomixis, in this letter, the rationale and methodology of a possible approach worth trying to select induced apomictic plant mutants is outlined. Availability of induced apomictic mutants may be critical in functional genomics analysis and agricultural exploitation of apomixis.

A variety of plant species belonging to mono- and dicotyledonous families possess apomictic property. Altogether they demonstrate the known forms of gametophytic and sporophytic types of apomictic reproduction<sup>3-7</sup>. A large majority of apomictic species share several features regarding their reproductive functions. (i) Apomictic reproduction occurs concomitantly with sexual reproduction<sup>4,5</sup>. (ii) They are auto- or allo-polyploids of various degrees<sup>8-10</sup>. (iii) Dominant allele(s) at one or more loci are the determinant genetic factors<sup>11–14</sup>. There are several examples of autonomous apomixes, wherein apomicts develop seeds bearing embryo as well as endosperm without the involvement of the fertilization process15,16. There are indications from the recent findings in Arabidopsis thaliana that fis (fertilization-independent seed) mutants are compromised in genomic methylation to different extents and thereby have pleiotropic effects on gene expression<sup>16–20</sup>. These observations allow the following hypothesis: Apomixis is a method to produce progeny; especially to overcome sexual sterility. A gain-of-function mutation brings about a degree of somatic apomictic fertility, over and above sexual fertility. The gain-of-function mutation occurs upstream in pathway(s) for apomictic embryogenesis. Mutation has pleiotropic effects because it turns on genes

that normally remain repressed, including those epigenetically inactivated/silenced via DNA methylation. Apomixis is seen preponderantly in polyploids, among the present-day plants, because apomictic mutations got selected in them for survival in the course of evolution. If apomictic reproduction process had not been superimposed, the concerned polyploids would have become extinct because they suffer from sexual sterility due to problems in meiosis arising from multivalent chromosomal formations instead of bivalents.

The above rationale about the origin of apomictic behaviour dictates the following kind of experimental scheme to recover apomictic mutants among presently nonapomictic, sexually reproducing species. The scheme may be fruitful in some species and not in others, determined by the nature of genetic background on account of the evolutionary history of the species. The plant species to be used in experimentation should be such that it is capable of producing fruits that bear a large number of seeds, as a result of single spontaneous or manual pollination event in its individual flowers. The genotype of the selected species should be such that the plants are able to produce fruits that are largely seedless. It should be possible to produce seeds of such genotypes in large numbers. To isolate the apomictic mutants in such a genetic background, seeds are to be effectively mutagenized with physical or chemical mutagens. Among the resulting M1 population, while bulk of the plants will bear sparsely seeded or seedless fruits, the apomictic mutants will have seed-bearing fruits. The plantlets obtained from densely seed-bearing M1 fruits will be genotyped to identify the products of apomixis. The progeny of

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the identified plants will be retested. It is expected that the mutants so identified will have compounded reproduction comprising apomictic and normal sexual mechanisms. The apomictic mutants will be easier to detect when the starting genotype is characterized by high or complete sexual sterility. The auto- and allo-triploids that are known to be nearly sterile may prove highly useful starting genotypes. To begin with, triploid genotypes of species belonging to Solanaceae, Cucurbitaceae and Brassicaceae appear to offer requisite advantages. In this regard, Arabidopsis (or Brassica), Lycopersicon esculentum (or Solanum melongena), papaya and melons appear promising herbaceous and tree species for experimentation.

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## **Bt**-cotton: Protein expression in leaves is most critical

This has reference to the article by Kranthi *et al.*<sup>1</sup>. Their conclusions ... 'Cry 1Ac expression levels were the lowest in the ovary of flowers and boll rind of green bolls, which constitute the most favoured sites of bollworm attack .... The toxin expression was clearly inadequate to confer full protection to the fruiting parts', have been widely quoted and exploited by certain NGOs to condemn this technology<sup>2</sup>.

No doubt, the fruiting parts, especially squares and bolls, are the most preferred for feeding by *Helicoverpa armigera*, causing direct damage and heavy yield loss. However, what appears to have been less realized is a significant fact that larvae move to these parts after completing their initial feeding on leaves.

*H. armigera* starts its activity when the crop is young and still in its vegetative phase. Its feeding and reproductive activities intensify as the crop enters the reproductive phase and plenty of squares and green bolls, the most cherished food, become available. Its most preferred site for oviposition is the leaf, especially tender leaves at the upper half of the plant<sup>3</sup>, although a small number

is laid on other parts as well. A great majority of the newly hatched larvae initially feed by scraping chlorophyll in the tender leaves and, as they grow, move over to the squares and bolls for further feeding and development<sup>4,5</sup>. The neonates that directly feed on squares and bolls are relatively few. Furthermore, Bt protein is most effective only against the early instar larvae, while the grown-up larvae (3rd instar and beyond), even if they feed on Bt plants, no matter whether it is flowers, squares or bolls, do not die, although they suffer a setback in their overall health. Therefore, it is most critical that in a Bt-cotton plant the expression of Bt protein should be adequate and sustained in the leaves throughout or for most part of the plant life. As a result, a large number of larvae that feed on such leaves, where the protein expression is highest, perish with little chance to advance to the next instar. Of course, if protein expression is adequate in the fruiting parts also, it will add to further mortality of the larvae hatching on them, however small their population may be.

The studies carried out to date<sup>6</sup>, including the recent one by by Kranthi *et al.*<sup>1</sup>, have revealed that *Bt* protein expression is highest in leaves. This matters the most. Hence *Bt*cotton is able to provide satisfactory control of *H. armigera* in India as in China, Australia and other old world countries where the same species occurs, and also related species in USA and other countries<sup>7</sup>.

If the authors had used neonates<sup>8,9</sup> instead of the one-day-old larvae for bioassays, it would have been closer to reality and they would have recorded higher percentage mortality in the tissues of all parts. However, they have brought out certain issues like the importance of parental background for improvement of *Bt*-cotton which should be considered wherever possible. I believe this was their intention.

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