RESEARCH PAPER ANALYSIS

CORN: FROM PROTOPLASTS TO FERTILE PLANTS

Plant biotechnologists have taken another important step towards the genetic engineering of maize. Two papers in this issue of *Bio/Technology* report the regeneration of fertile corn plants from protoplasts. Although our group had previously demonstrated the regeneration of mature plants from the same source—protoplasts (Rhodes et al. *Bio/Technology* **6**:56, Jan. '88)—none of the plants were fertile. We attributed this to the condition of the donor cell culture used to derive the protoplasts. Now Laudenir Prioli et al. and Raymond Shillito et al., using different cultures, have succeeded in obtaining fertile plants. In theory their results mean that new traits introduced by *in vitro* genetic manipulations, which are essential for the development of improved maize varieties, can now be recovered in progeny.

Prioli and her colleagues at DNA Plant Technology (Cinnaminson, NJ) used, as a source of protoplasts, an embryogenic suspension culture of a Cateto inbred genotype adapted to tropical regions. Shillito and his coworkers at CIBA-Geigy (Research Triangle Park, NC) achieved their results with an elite inbred genotype. Together their reports show that the capacity of maize protoplasts to regenerate into fertile plants is not limited to a narrow germplasm. Both groups emphasize the importance of long-term (several months) propagation of donor cells in suspension culture. Callus cultures, no matter how friable, do not yield totipotent protoplasts.

What remains mysterious is how exactly to induce the competent state in maize cultures. What occurs in the switch from solid to liquid medium that permits cell digestion? Why does it happen in some suspension cultures and not others? While the authors provide complete descriptions of the history of each donor culture, it is unknown what steps were critical in creating the "correct" physiology. And since the two laboratories used different protocols to develop their donor cultures, perhaps none of the individual steps are crucial. Somewhat surprisingly, several aspects of the culture method do not appear important in obtaining regenerable callus. For example, the three groups that have now reported successful regeneration of maize protoplasts used three different methods of protoplast culture.

Despite these individual successes, we are still left with screening as the only means of identifying totipotent donor cultures.

Because the capability of a donor cell suspension culture to regenerate declines with age, the CIBA-Geigy group periodically preserved their cultures in liquid nitrogen. When the regeneration frequency of the cultures began to drop, they could revive an aliquot of frozen culture and continue to produce plants. Can this window in which regeneration potential coincides with protoplast growth be expanded?

Since direct DNA uptake via electroporation or polyethylene glycol works well with maize protoplasts, it should only be a short time before the full potential of these results is realized.

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