

INITIAL TESTS FIND MEXICAN LANDRACES IN CIMMYT GENE BANK FREE OF PROMOTER ASSOCIATED WITH TRANSGENES

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El Batán, Texcoco, Mexico—Amid recent reports that transgenic maize has been detected in farm fields in several Mexican states, the International Maize and Wheat Improvement Center (CIMMYT) commenced screening Mexican landraces held in its maize gene bank (such selections are termed accessions). Results of those screenings, obtained on October 15, 2001, show that none of the 28 representative populations that were tested carry the CaMV 35S promoter, which would indicate the presence of an introduced gene (transgene). In addition, in coming days CIMMYT scientists will be screening seed lots recently collected in Oaxaca as part of a study on farmer varieties (but not entered as germplasm bank accessions), for the presence of the promoter.

CIMMYT would like to reiterate its offer to provide its considerable expertise to the appropriate Mexican institutions to (1) help identify the type and source of the introduced gene(s), (2) assess potential impacts to biodiversity, the ecology, and the socioeconomic environment, and (3) to explore possible responses.

Screening process and results

Twenty-eight populations were selected from the CIMMYT maize germplasm bank, spanning accessions collected or regenerated from eleven Mexican states and dating as far back as 1967. Although the first commercial transgenic maize was not released in the United States until 1996, researchers wanted to determine whether any transgenes had made their way into the bank's collection during recent regenerations of the seed stocks.

The 28 populations were screened for the presence of the cauliflower mosaic virus (CaMV) 35S promoter, a fragment of DNA found in most commercial transgenic maize and not found naturally in the maize genome. Thirty plants of each of the 28 populations were planted in CIMMYT's experimental greenhouse and single leaves were subsequently harvested from each of the plants. DNA was extracted, quantified, and mixed in the same tube to form bulks of 15 plants each, thereby ensuring that two bulks would represent the

DNA of each population. The mixtures were amplified using the Polymerase Chain Reaction (PCR), the most sensitive method for detecting DNA fragments, using a primer specific to the CaMV 35S promoter. Amplified DNA was electrophoresed and visualized on agarose gels.

In a control test to measure the sensitivity of the analysis, CIMMYT scientists extracted DNA from a transgenic plant that was known to contain the CaMV 35S promoter and mixed it with DNA from nontransformed plants at a ratio of 1:14 ('transformed DNA' to 'nontransformed DNA'). In all tests, the CaMV 35S promoter sequence was detected in the mixed DNA sample. The use of this mixed DNA ensures that the CaMV 35S promoter will be detected if present in the bulks derived from the gene bank samples.

In the main test of the gene bank germplasm, all DNA samples were amplified by both the CaMV 35S promoter primer as well as a primer corresponding to a fragment of DNA known to exist naturally in the maize genome (a molecular marker called phi96100). Finally, DNA of a positive control, known to contain the CaMV 35S promoter, was amplified to test that the CaMV primer sequence does indeed amplify the expected fragment of DNA in transgenic maize. The results showed that no DNA isolated from the gene bank accessions amplified a fragment with the CaMV 35S promoter; the positive control did amplify the fragment with the CaMV 35S promoter; and all gene bank material amplified the expected fragment using the maize primer phi96100. Thus, all of the reactions worked correctly, and none of the 30 plants from each of the 28 populations contained the CaMV 35S promoter.

Herbicide tests to determine the presence of expressed herbicide-resistant transgenes and PCR-based tests on other materials as described above are ongoing.

Details of the study results may be obtained by contacting David Hoisington, Director of CIMMYT's Applied Biotechnology Center, at d.hoisington@cgiar.org

This document was prepared in English, and a Spanish translation has been prepared for informational purposes. In the event of any inconsistency between the different versions, however, the English version should be considered the authoritative text.