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Identification and characterization of Markers Linked to Resistance Mofifs against Maize Chlorotic Mottle Virus Infection in Tanzanian Maize Germplasm through Amplified Fragment Length Polymorphism Analysis

Abstract

Maize (*Zea mays* L.) is the major staple food crop in Tanzania providing about 60% of dietary calories to the people. Although this cereal has many positive economic attributes, it is beset by environmental and biological stresses that result in significant loss of potential harvest yield each year. The major biological stress facing maize production in Tanzania is maize lethal necrosis (MLN) disease, which is induced by the synergetic infection of *maize chlorotic mottle virus* and any of the cereal potyviruses such as *sugarcane mosaic virus.* Resistance to MCMV in cultivated maize varieties in Tanzania and in East African region has not been reported. Thus it poses significant threats to maize production in the region. This study sought to screen, at the phenotypic and molecular levels, Tanzanian maize landraces and inbreeds lines against MLN in a comparative analysis with promising resistance and susceptible lines from CIMMYT-Kenya and from U.S. origins. One hundred and fifty two Tanzanian maize landraces and 33 maize inbreed lines were artificially inoculated with *maize chlorotic mottle virus* and *sugarcane mosaic virus* isolates in two separate trials to identify resistant phenotype against maize lethal necrosis. Disease evaluation was assessed based on 1-5 maize lethal necrosis rating scale at 14, 28, 42 and 72 days post inoculation (dpi) for landraces and at 7, 14, 21 and 52 (dpi) for inbred lines. Significant phenotypic variations *(p=0.05)* were observed among the landraces in terms of symptoms development and disease severity scores. Landrace TZA-2793 had the lowest severity mean score of 3.5 followed by four other landraces (TZA-3585, TZA-3543, TZA-4505 and TZA-2292), with mean a severity score of 3.75. No significant variations *(p=0.05)* were detected in trials involving inbreed lines, as all were susceptible to maize lethal necrosis. They had scores ranging from 4.5 to 5, except for the resistant check (CML494), which differed from the inbred lines. It had a mean score of 3.75. Twenty two genotypes including selected landraces under artificial inoculation, promising resistance and susceptible lines of CIMMYT and the U.S. were subjected to AFLP analysis with 11 AFLP primer combinations. The analysis yielded 1025 polymorphic AFLP allelic fragments with an average polymorphism percent of 62.75%, whereas genetic similarity coefficients among the genotypes ranged from 0.60 to 0.88. Cluster analysis, using UPGMA based on AFLP data matrix, revealed three clusters that grouped genotypes according to their reaction to MLN disease. Promising resistant/tolerant genotypes were grouped in Cluster I and susceptible genotypes in Clusters II and III. Also landraces were grouped according to agro-ecological locations where they were collected. Eluted polymorphic AFLP fragments were sequenced. Nucleotide BLAST showed the similarity of loci, which are associated with disease resistance genes such as pathogenesis related proteins, Serine/threonine kinase protein, rust resistance protein (rp3-1), receptor kinases and Zea mays putative zinc finger protein genes. Other important loci that are responsible for plant response to stress were also identified. Further studies are recommended to (1) explore the potential of identified AFLP markers, their association to disease resistance, and if the regions of these markers are hosting other genes linked to resistance of other diseases of maize; and

(2) subject the five identified landraces to further MLN tests in order to determine the utility of using these materials in future MLN disease resistance breeding programs.