AN AFLP APPROACH TO INVESTIGATE SEX DETERMINATION IN WATERHEMP. Dean S. Volenberg and Patrick J. Tranel, Postdoctoral Research Associate and Associate Professor, Department of Crop Sciences, University of Illinois, Urbana, IL 61801.

There are several molecular techniques which generate molecular markers. Certain molecular techniques have advantages over others depending on the research question being addressed. The amplified length polymorphism (AFLP) technique generates large numbers of molecular markers and provides a rapid method for scanning the genomes of different individuals for sequence variation. The AFLP technique can be used to quantify genetic variation, assist in marker assisted breeding programs, and identify sex specific markers in plant species which are sexually dimorphic.

Waterhemp is a weed species which exhibits sexual dimorphism. Historical (1930s) genetic work involving crosses between waterhemp and monoecious amaranth species suggested that males are the heterogametic sex. In other words, males have an XY chromosome pair, whereas females have an XX chromosome pair. However, sex chromosomes in waterhemp have never been distinguished from the autosomes. The objective of this study was to evaluate DNA from female and male waterhemp plants using AFLP technique to identify female- or male-specific molecular markers.

A dioecious waterhemp accession was tested for polymorphisms between full-sib-male and -female plants using the AFLP technique. Equal amounts of DNA from five female or male plants were bulked. Eighteen primer combinations were tested, and two of these primer combinations yielded a male-specific molecular marker. The experiment was repeated on DNA from individual male and female plants to verify sex specificity of the molecular marker.

AFLP band patterns in waterhemp showed a high level of variation between individuals, which were unrelated to sex. The eighteen primer combinations were scored to quantify the level of polymorphism in waterhemp. In total, 1928 bands were polymorphic out of 3079 bands scored, producing an average band polymorphism of 62.6%.

AFLP technique in waterhemp generated very low level (0.06% of total scorable bands) of male specific markers. The primer combinations of MseI-CTA/EcoRI-AGC and MseI-CTA/EcoRI-ACA each generated one male specific band of approximately 202 and 252 bp, respectively. These bands were present in all male plants, but absent from all female plants tested. The eighteen primer combinations failed to yield a female-specific molecular marker.

Future research will:1) evaluate other unique primer combinations, 2) clone and sequence male-specific bands(s), and 3) design primers to amplify male-specific molecular markers.