ACOSTA-LEAL, RODOLFO* and CHARLES M. RUSH. Texas Agricultural Experiment Station, 6500 Amarillo Blvd. West, Amarillo, TX 79106. **A procedure for rapid detection of resistance breaking variants of *Beet necrotic yellow vein virus* (BNYVV) using real-time RT-PCR allelic discrimination assays.**

## ABSTRACT

Resistant varieties of sugar beets, grown in the Imperial Valley of California, have been increasingly damaged by a new strain of BNYVV. Total RNA was isolated from asymptomatic and symptomatic (severe rhizomania disease) resistant plants and a portion of viral RNA 3, including the p25 gene, was amplified by RT-PCR and sequenced. The analysis revealed two polymorphic sites, A67V and D135E, associated with the capability of the virus to overcome resistance. Based on this data, a set of TaqMan probes was designed for each site to discriminate between wild type (WT) and resistant breaking (RB) variants of BNYVV. The specific fluorescence emitted by each probe was detected and analyzed by an ABI 7000 real-time PCR system and the predicted genotypes were corroborated by DNA sequencing. The capability of this technology to typify numerous isolates facilitated the analysis of the spatial distribution of virus genotypes in the field. Thus, RB variants were mostly baited from yellow strips with high incidence of rhizomania, whereas WT variants predominated in the surrounding green areas. Mixed infections were found mainly in green and transitional zones. The predominance of the RB isolates in yellow strips suggests that they have gained fitness in resistant cultivars and will eventually become the dominant population.