INTERACTIONS BETWEEN BNYVV AND BSBMV AND REMOTE SENSING OF BNYVV

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Beet Necrotic Yellow Vein Virus (BNYVV) and Beet Soil Borne Mosaic Virus (BSBMV) are closely related viruses affecting beet production in the sugar beet growing regions of Minnesota and North Dakota. The viruses are vectored by the same soil borne fungus, *Polymyxa betae*. Since the viruses share the same vector, they are frequently found in the same fields and can both be found infecting the same beet. Their close ecological relationship and their relation at the molecular level suggest that recombination is highly possible. This is a recognized method that viruses use to develop new strains that can differ from the two "parents" in virulence. New sugar beet varieties have been developed that have a high degree of resistance to BNYVV, but they are not resistant to BSBMV. Understanding the relationships between theses viruses will lead to a better understanding of the risks these viruses pose. Understanding the distribution of these viruses and developing better detection methods will lead to better management strategies. We have conducted studies to evaluate the distribution and interaction of BNYVV and BSBMV at the field level. Remote sensing techniques have been used effectively to identify problem regions of fields. We have conducted studies to evaluate remote sensing techniques to determine their ability to detect BNYVV in sugar beet fields and differentiate BNYVV infection from abiotic stress.

Methods

Interactions and distribution of BNYVV and BSBMV- This is a continuation of research initiated in 1999. In 2000, 5 fields in the Southern Minn region, 2 fields in the American Crystal area, and 1 field in Bushland Texas were grid soil sampled. This resulted in 1465 samples being collected. These samples were taken back to the laboratory in Texas and bioassays were initiated by planting sugar beet seed in soil samples obtained from each of the fields. All soil samples from an individual field were planted at the same time. Planting dates for samples from an individual field were staggered at six week intervals and bait plants growing in the soil samples were harvested approximately 10 to 12 weeks after planting. After harvest, plants were tested by ELISA and this data was used to determine the distribution of BSBMV and BNYVV in the field. Viral RNA was extracted from each plant and stored at -20? C for further analysis. Primers have been developed to allow amplification of each virus separately through PCR. We are in the process of amplifying the viral RNA through RT-PCR. These products of PCR will then be subjected to restriction analysis and SSCP. Restriction analysis and SSCP are common molecular techniques that assess the genetic diversity of nucleic acids. The degree of diversity of the genomic nucleic acids of the virus will indicate the variability of the viruses and the possibility of recombination.

Remote sensing of BNYVV- This was the first year of this study. We selected 3 fields in the American Crystal region and 4 fields in the Southern Minn region that showed discreet yellow spots indicative of rhizomania. In each field a yellow spot and a nearby green spot were selected for sampling and georeferenced. These fields were sampled 4 times between August and November. When the weather was clear, 4 readings with a hand held radiometer were taken at each spot. A beet was then selected from directly below where the radiometer reading was taken. Leaves and roots were collected from each beet and transported back to the laboratory in Texas. Roots were analyzed for the presence of BNYVV and BSBMV by ELISA. Leaf spectra were collected with an integrating sphere and an ASD hyper-spectral radiometer. Leaves were then frozen at -20? C. Leaf pigments were extracted from frozen leaves in acetone and read in a scanning spectrophotometer. When pigment extracts are completed, leaf tissue nutrients will be analyzed. When possible satellite and airborne photos were collected. When cloud cover prevented use of the radiometer, samples were still collected for laboratory analysis. Four of these fields were also selected for intensive grid soil sampling.

Results

Interactions and distribution of BNYVV and BSBMV- Geostatistical analysis showed significant levels of autocorrelation between adjacent samples. This indicates a patchy distribution of the viruses. BNYVV was detected

in many areas that showed no symptoms of rhizomania (Figure 1). BSBMV showed a small but significant positive correlation with beet quality. Work to determine the genetic diversity of the viruses is in progress but not yet completed.

Remote sensing of BNYVV- Analysis of spectral readings from either the hand-held radiometer or the ASD integrating sphere showed no distinction between BNYVV infected and non-infected plants in apparently healthy areas, though apparently healthy and apparently diseased areas could readily be distinguished at most wavebands. Spectrophotometer scans of extracted leaf pigments were able to differentiate between infected and non-infected beets in apparently healthy regions (figure 2). There were no differences in soil nitrogen between apparently healthy and apparently diseased areas.

Discussion

The patchy distribution of the viruses suggests that a directed sampling scheme could be developed to accurately detect the viruses. However, the presence of the virus in asymptomatic regions of the fields requires a more sensitive method of detecting the viruses. Spectrophotometric scans of pigment extracts shows promise of being able to develop a quick non-destructive lab based assay for detection of BNYVV. This would allow growers to know which regions of their fields are infected before rhizomania reached damaging levels. These areas could be planted to a resistant sugar beet variety next time the field is planted to sugar beets.

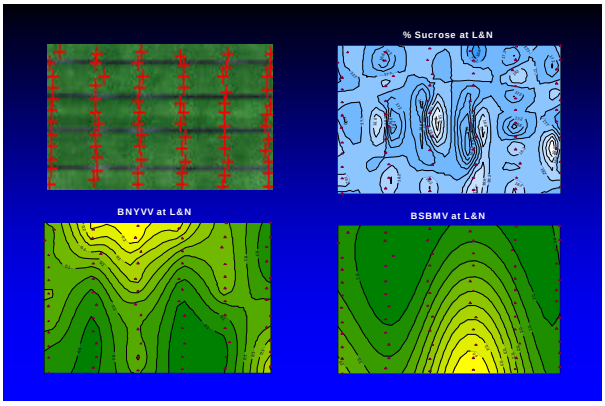

Figure 1. Distribution of BNYVV and BSBMV in relation to visual symptoms and sugar content. Red triangles and crosses represent locations of samples.

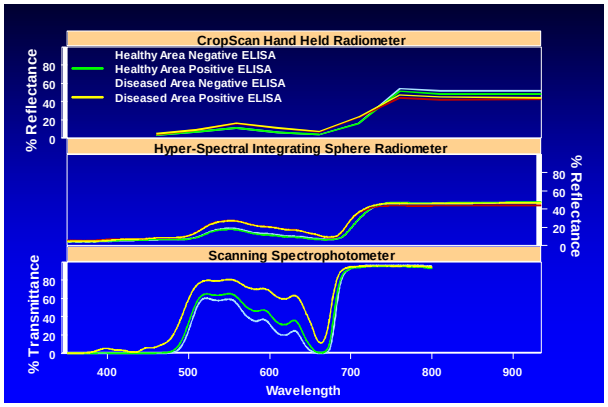

Figure 2. Spectral properties of sugar beet leaves with different instruments. Only the spectrophotometer was able to differentiate between infected and noninfected beets taken from apparently healthy regions of the field.