BARGABUS-LARSON, REBECCA L\*<sup>1</sup>, JOHN J. WEILAND<sup>2</sup>, <sup>1</sup>USDA-ARS, Sugarbeet Research Unit, Crops Research Laboratory, 1701 Centre Ave, Fort Collins, CO 80526, <sup>2</sup> USDA-ARS, Sugarbeet and Potato Research Unit, Northern Crop Science Laboratory, 1307 N 18<sup>th</sup> St, Fargo, ND 58105. **RNA silencing for the control of** *Beet necrotic yellow vein virus* infection of sugarbeet.

Beet necrotic yellow vein virus (BNYVV), a multipartite single-stranded RNA benyvirus causing Rhizomania in sugarbeet, is a serious threat worldwide. Tolerant cultivars currently available succumb to Rhizomania under severe disease pressure. The lack of complete control with tolerance prompted investigation into novel means of preventing infection. RNA silencing, a naturally occurring phenomenon, results in the post-transcriptional degradation of aberrant double-stranded RNAs, including mRNAs, preventing protein synthesis. This process has been induced under laboratory conditions for preventing virus infections in numerous plant and animal host-virus systems and may operate in currently deployed genetically enhanced sugarbeets exhibiting BNYVV resistance. In the current study, guide sequences used for eliciting silencing were designed to target blocks of untranslated and coding regions of the RNA1 of BNYVV that encodes for viral replication machinery. The sequences were amplified by reverse transcriptase polymerase chain reaction (RT-PCR), or in the case of small hairpin (hp) RNA, created by direct synthesis of deoxyoligonucleotides with 60 base pair "arm lengths". Silencing constructs were developed by cloning these fragments into a Barley stripe mosaic virus (BSMV) vector. When using either the RT-PCR generated or small hpRNA constructs, a higher degree of silencing was achieved using targets for the untranslated regions of RNA1 when compared to constructs containing RNA1 coding region guide sequences, as determined by disease reduction and ELISA assays.

feeder roots begins in early to mid-fune and commute throughout the growing sensor. Feeding injury causes rignificant crop damage that includes several seedling toproots or badly scarred root surfaces (Yuu 1986). Damaged mots are preduposed to secondary infections inflicted by opportunistic mitrobal pathagens that further contribute to reductions in sugar yields (Campbell et al., 1998; Cooke 1993). Although moderately resistant inters have been released, sugarbert germplasm with high resistance to SBKM is lacking (Campbell et al., 2000). Current control measures rely privarily on chemical insectivities but alternative controls are being sought as the potential for buildup of insectivities but alternative controls are being sought as the potential for buildup of increased resistance to these posticides is antivipered.

Development of efficient insect bioassays is imperative for rapid screening of resistance assetirely in order to design effective approaches for control of insect pests. The inability to completely rear the meet in the laboratory and a need to utilize minure sugarbeet tapoots have madered the development of an effectient bisausay for SBRM larvae. Axenic plant tissues have been used in insect feeding studies to tear some insect interaction and to study plant-insect interactions (Kimmons et al., 1990; Wu et al., 1990), and interaction and to study plant-insect interactions (Kimmons et al., 1990; Wu et al., 1990), and to study plant-insect interactions (Kimmons et al., 1990; Wu et al., 1990), supercot cultures of augarbeet tares that are either susceptible or moderately resistant to sugarbeet in 1990; Campbell et al., 1990; Simgoole et al., 2000; Simgoole et al., 2005). Using these sugarbeet inity not cultures as well as the corresponding stateptible and restatant see ending taget of et al., 2000; Simgoole et al., 2005). Using these sugarbeet inity not cultures as well as the corresponding stateptible and restatant see ending stateptible and restatant to sugarbeet inity not cultures as well as the corresponding stateptible and restatant seedlings, we developed an in vitro SBRM bioassay.

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