THE EFFECTS OF *BEET CHLOROSIS VIRUS* (BCHV) ON THE YIELD OF SUGAR BEET

M. STEVENS AND P. B. HALLSWORTH

Broom's Barn Research Station, Higham, Bury St Edmunds, Suffolk, ENGLAND IP28 6NP.

ABSTRACT

Virus yellows remains an important disease of the UK sugar beet crop because the maritime climate favours the overwinter survival of the aphid species that transmit the viruses to the crop. Several different viruses are involved in the disease complex including the poleroviruses *Beet mild yellowing virus* (BMYV) and the recently identified *Beet chlorosis virus* (BChV). Studies have shown that the biological and molecular properties of BChV differ significantly from BMYV; the effects of BChV on the yield of UK sugar beet are unknown. Therefore, in studies at Broom's Barn, the separate effects of BMYV and BChV on the yield of field-grown sugar beet were studied following sequential inoculations from May to July in 1997, 1999 and 2000. Each sugar beet plant within the appropriate plots was infected with virus using at least 10 wingless viruliferous *Myzus persicae* per plant. In all three years, yield losses were negatively correlated with time of infection, with early season (May) inoculations causing 18-27% losses in sugar yield but late season losses only 4-15%. BChV decreased the yield and sugar content of beet following early season inoculations, although the effects on yield were more variable (range 8-24%) and less pathogenic than those caused by BMYV. However, inoculations with BChV in July of each year caused greater root and sugar losses than inoculations with BMYV at that time. Both poleroviruses increased the sodium content of the roots early in the season, although neither virus had an effect on potassium levels at any stage.

ABREGE

La jaunisse virale reste une des maladies les plus importantes touchant la betterave à sucre au Royaume Uni à cause du climat maritime qui favorise la survie des espèces d'aphides transmettant le virus aux cultures. La maladie est due à plusieurs virus dont les polerovirus de la jaunisse modérée (Beet Mild Yellowing Virus (BMYV) et celui, plus récemment découvert, de la chlorose de la betterave à sucre (Beet Chlorosis Virus (BChV). Des études ont montré que les propriétés biologiques et moléculaires du BChV diffèrent de façon significative de celles du BMYV; les effets du BChV sur le rendement en betterave à sucre au Royaume Uni restant inconnus. Par conséquent, des études ont été conduites a Broom's Barn afin de différencier les effets respectifs du BMYV et du BChV sur le rendement des betteraves à sucre cultivées dans des champs expérimentaux après inoculations séquentielles de mai à juillet lors des années 1997, 1999 et 2000. Chaque lot de betteraves, au sein des plants

appropriés, fut infecté avec une dose d'au moins 10 aptères virulifères de l'espèce *Mysus persicae* par plante. Au cours des années tests, les pertes de rendement furent négativement corrélées à la période d'infection. Lors d'infections ayant lieu en début de saison (mai), les pertes de rendement furent de l'ordre de 18 a 27% alors qu'en inoculation en fin de saison, ces pertes furent de l'ordre de 4 a15%. Concernant les effets du BChV à proprement parlé, lorsqu'il fut inoculé en début de saison, les pertes de rendement furent plus variables (8 a 24%) mais les effets furent moins pathogènes que ceux provoqués par le BMYV. En revanche, les inoculations de BChV en juillet de chaque année causèrent des pertes en racines et en sucre plus importantes que lors d'inoculations de BMYV à la même période. Tôt dans la saison, les deux polerovirus furent responsables d'une augmentation de la quantité en sodium au sein des racines. Par contre, à aucun moment, l'un de ces virus n'eut d'effet sur la quantité en potassium.

KURFASSUNG

Vergilbungs-Viren bleiben eine wichtige Krankheit der britischen Zuckerrübenproduktion, da das maritime Klima das Überleben der Blattlausarten begünstigt, die die Viren auf die Rüben übertragen. An dem Krankheitskomplex sind verschiedene Viren beteiligt, darunter das Polerovirus *Beet mild yellowing virus* (BMYV) und das kürzlich identifizierte *Beet chlorosis virus* (BChV). In Untersuchungen wurde gezeigt, dass sich die biologischen und molekularen Eigenschaften von BChV signifikant von BMYV unterscheiden; der Einfluß des BChV-Befalls auf den Ertrag von Zuckerrüben in England sind unbekannt. Deshalb wurden am Broom's Barn die Auswirkungen von BMYV und BChV auf den Ertrag im Feld angebauter Zuckerrüben nach sequentiellen Inokulationen von Mai bis Juli der Jahre 1997, 1999 und 2000 studiert. Jede Zuckerrübenpflanze in den entsprechenden Parzellen wurde mit dem Virus infiziert, wofür mindestens 10 flügellose, virustragende Blattläuse (*Myzus persicae)* pro Pflanze verwendet wurden. In allen drei Jahren bestand eine negative Korrelation zwischen den Ertragsverlusten und dem Infektionszeitpunkt, wobei früh in der Saison (Mai) erfolgte Inokulationen Verluste des Zuckerertrags von 18 - 27 % verursachten, spät in der Saison erfolgte Inokulationen jedoch nur 4 -15 %. BChV reduzierte den Ertrag und Zuckergehalt von Zuckerrüben nach früh in der Saison erfolgten Inokulationen, obgleich die Auswirkungen auf den Ertrag eine grössere Variation (im Bereich von 8 - 24 %) und geringere Pathogenität zeigten als nach Infektion mit BMYV . Inokulationen mit BChV im Juli eines jeden Jahres verursachten jedoch höhere Verluste im Rübengewicht und Zuckergehalt als Inokulationen mit BMYV zu diesem Zeitpunkt. Beide Poleroviren steigerten früh in der Saison den Natriumgehalt der Wurzeln, aber keiner der beiden Viren hatte in irgendeinem Stadium Auswirkungen auf den Kaliumspiegel.

INTRODUCTION

Until recently, the viruses most commonly involved in the virus yellows disease complex were thought to be *Beet mild yellowing virus* (BMYV; genus *Polerovirus*, family *Luteoviridae*) and *Beet yellows virus* (BYV; family

Closteroviridae, genus *Closterovirus*). Both are transmitted by aphids, primarily *Myzus persicae*, and these viruses can seriously decrease the sugar content and root yield of infected plants (Smith & Hallsworth, 1990). In 1989, a virus that exhibited symptoms of paler yellowing rather than the characteristic golden yellow discoloration associated with BMYV was found in the UK. Initially, this virus was described as a second strain of BMYV based on its biological, serological and molecular properties (Stevens *et al.*, 1994; Hauser *et al.*, 2000). Surveys of UK sugar beet crops showed that this new strain of the virus represented 11% of infected leaves; its host range was also narrower and appears to be confined to *Beta* species. Further studies of the biological and molecular properties of this strain showed that it differs significantly from BMYV and has been classified as a new species within the *Polerovirus* genus (Hauser *et al.*, 2002), the name *Beet chlorosis virus* (BChV) recently being accepted.

Each year, the UK sugar beet industry requires information on the effects of virus yellows on the national yield of sugar. As the effects of BChV are unknown a series of trials were conducted to determine the economic importance of this disease.

MATERIALS AND METHODS

Artificially-inoculated field trials were set up at Broom's Barn in 1997, 1999 and 2000. Cultures of BChV and BMYV were established in either sugar beet or *Capsella bursa-pastoris*, respectively, as BChV does not infect *C. bursa-pastoris*.

The field trials were sown between 21 March and 2 April each year. In 1997, untreated seed of the variety Zulu was sown and in subsequent years Roberta was used. Sugar beet were inoculated with either virus in mid-May, June and July respectively, while control plots were left uninoculated. In each trial there were four replicates of each treatment. Each plot was surrounded by guard rows of sugar beet treated with the insecticide imidacloprid ((Gaucho) 90 g a.i. / unit of seed) to minimise virus spread. To inoculate the plots, pieces of virus-infected leaf tissue, infested with at least 10 wingless *M. persicae* were placed into the heart leaves of every sugar beet plant. To avoid cross-contamination of the two viruses inoculations were undertaken on different days, preferably when it was not too windy. Aphids were then killed 48 hours after the second inoculation with pirimicarb (Aphox). The risk of virus spread was minimised by spraying the trial each month from May until August with pirimicarb.

The number of plants showing symptoms were recorded each month from June to September. In late September, 10 leaves from each plot were tested by ELISA to confirm the presence of BChV and BMYV and to determine levels of cross-contamination that may have occurred.

In 1997 and 1999 the trials were machine harvested in November, but in 2000, due to unfavourable conditions during the autumn, this trial had to be harvested by hand in December. Roots from each plot were washed and weighed and their sugar concentration determined polarimetrically. Impurity levels of amino-nitrogen, sodium and potassium were also calculated.

RESULTS

In all three years sugar beet plants started to exhibit symptoms four to five weeks after inoculation, and within two months of infection at least 95% of plants within plots showed typical virus symptoms. Less than six percent of the uninoculated plants showed any virus symptoms by the end of July, although by early October up to 25% of plants showed some yellowing, indicating the difficulties of preventing spread in such trials even under strict hygiene precautions.

BMYV had a significant effect on the yield of sugar beet when plants were inoculated in mid-May and June. Highest yield losses were recorded after May inoculations, which decreased the adjusted tonnes per hectare by 21, 30 and 28% in 1997, 1999 and 2000 respectively. The impact of BMYV infection decreased with time, although in 2000, BMYV still had a significant effect on root yield after infection in July. BMYV also decreased the sugar concentration by up to 27% and increased the levels of sodium and amino nitrogen in the roots, particularly when sugar beet were inoculated in May and June.

In contrast, although BChV significantly decreased the yield and sugar content of beet in both May and June each year (except May 1997) in comparison to the uninoculated control plots, BChV was actually less damaging than BMYV after these early inoculations. Yield losses of between 9 and 25% were observed in May. However, in July inoculations each year, BChV had a greater effect on root and sugar yield than BMYV. BChV also increased the levels of sodium of infected beet, but had no effect on amino nitrogen or potassium content of infected beet.

CONCLUSION

Both BMYV and BChV significantly decreased the root weights and sugar yields of field-grown sugar beet and increased the juice impurity levels, particularly of sodium, in the roots. Greatest yield losses were experienced when sugar beet plants were inoculated at an early growth stage. However, the effects of BChV on yield were more variable than BMYV throughout the three year period. It would appear that BChV is less damaging than BMYV when plants were inoculated during May, but July inoculations with BChV had a greater effect on yield than BMYV. BChV produces paler symptoms than BMYV and often leaf tissue around the midrib and lateral veins remains green, which could enable infected leaves to maintain a greater photosynthetic potential than BMYV-infected leaves. However, it remains unclear as to why BChV was more damaging to sugar beet compared to BMYV later in the season. There was no difference in the rate of symptom development during the season, and further work is required to determine the effects these poleroviruses have on the physiology and photosynthetic efficiencies of infected plants.

Plants within the uninoculated control plots were sprayed with insecticide to prevent the spread of BMYV and BChV. However, several plants within these plots became infected and beet inoculated with BChV sometimes contained BMYV as well. Winged virus-carrying migrant aphids alighting in these plots between insecticide treatments may have established further sources of

infection and spread the virus across the plots. Most virus infection identified in the uninoculated plots did not appear until August and would not have affected the yield significantly, and any yield loss in uninoculated plots would actually lead to an underestimation of the effects of BMYV and BChV on final yield. These problems indicate the difficulties of undertaking such trials with aphid-borne viruses, even when strict hygiene measures are followed.

Each year British Sugar requires information on the factors affecting the growth of the UK sugar beet crop in order to predict the national sugar yield at the end of the season. Actual losses attributed to virus yellows vary from year to year, and are influenced by the levels and timing of infection, causal virus (i.e. BMYV or BYV or a mixed infection) and environmental conditions that influence plant growth. These trials have shown that the variation in yield losses will also depend on the species of polerovirus present in the crop.

ACKNOWLEDGEMENTS

This work was funded by the British Beet Research Organisation (BBRO).

REFERENCES

1. SMITH, H. G. & HALLSWORTH, P. B.: The effects of yellowing viruses on yield of sugar beet in field trials, 1985 and 1987. *Annals of Applied Biology 116*, 503-511, 1990.

2. STEVENS, M., SMITH, H. G. & HALLSWORTH, P. B.: Identification of a second distinct strain of BMYV using monoclonal antibodies and transmission studies. *Annals of Applied Biology 125*, 515-520, 1994.

3. HAUSER, S., STEVENS, M., MOUGEL, C., SMITH, H. G., FRITSCH, C., HERRBACH, E. & LEMAIRE, O.: Biological, serological and molecular variability suggest three distinct polerovirus species infecting beet or rape. *Phytopathology 90*, 460-466, 2000.

4. HAUSER, S., STEVENS, M., BEUVE, M. & LEMAIRE, O.: Biological properties and molecular characterization of beet chlorosis virus (BChV). *Archives of Virology 147*, 745-762, 2002.