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Sweet maize hybrids' yield response to adult western corn rootworm (*Diabrotica v. virgifera* Leconte, Col.: Chrysomelidae) silk feeding and IPM implications thereof

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Abstract. The western corn rootworm [*Diabrotica virgifera virgifera* LeConte (WCR)] is an important pest of maize in USA and Canada, and Europe. Its primary damage is due to larvae feeding on maize roots; yield and quality also occur due to WCR adult silk clipping and interference with ear pollination. WCR economic thresholds for sweet maize hybrids are not available. We aimed to determine whether silk feeding and artificial silk cut in various sweet maize hybrids could be correlated to potential WCR adult levels. The two-year field study was conducted in Hungary using seven different sweet maize hybrids. The silk was cut back to 0, 1, and 2 cm, measured, and compared daily to the silk length of uncut control plants. As a separate treatment, WCR adults were placed in cages on the uncut Suregold hybrid to assess silk clipping activity. After harvest, cob weights were measured, and fertility levels were assessed. Results showed that sweet maize hybrids can tolerate some silk cutting (simulating high WCR density and silk feeding). The action threshold for these hybrids is much higher than that for inbred lines or commercial grain maize, in line with prior studies for the Suregold hybrid (8-12 adults per year). Meteorological conditions and multiple pest presence, phytosanitary regulations, and IPM considerations may help finetune adult WCR control intervention decisions.

Keywords: *Zea mays*, WCR silk feeding, silk cutting, yield parameters, hybrid response, IPM.

Introduction

Maize yield can be considerably affected by the western corn rootworm [*Diabrotica virgifera virgifera* LeConte (WCR)] in North America and in Europe (Gray et al. 2009, Edwards & Kiss 2012, Bažok et al. 2021). This pest is univoltine, and its eggs overwinter in the soil of maize fields. The larvae hatch in the spring and feed on the maize root system (Krysan & Miller 1986). First adults emerge from mid to late June, continuing into September in some locations, and protandry dominates this pattern (Meinke et al. 2009, Toth et al. 2020). They feed primarily on maize pollen, silk, and grain (Chiang 1973) or, if not available, on maize leaves or pollen of other plant species, including weeds in and around maize fields; however, many combinations of maize and other food source are disadvantageous for adult survival (Moeser &Vidal 2005, Toepfer et al. 2015). The WCR adults are active flyers; therefore, population feeding on silk can also depend on pest population levels of other maize fields (Szalai et al. 2011).

The main WCR damage is caused by larval feeding on the maize root system. The adults feeding on the silks of maize, referred to as silk clipping, may cause significant damage resulting in reduced fertilization and kernel set (Culy et al. 1992, Tuska et al. 2002, 2003).

Silk formation, pollination, and subsequent fertilization in maize is an important complex process that highly impacts the yield and quality of grain. Pollen production, viability, and shed are discussed in publications of the Division of Extension, University of Wisconsin-Madison 2006 and Fonseca &Westgate 2004. Pollen grains arriving at the silks adhere to the papilla hairs, germinate, and rapidly develop pollen tubes for delivering sperm cells to the ovules (Zhou et al. 2017). Successful seed set depends on several environmental factors, such as air temperature and relative humidity (Schoper et al. 1986), and the physical presence of silks, as well as on the silk length, which may be impacted by several pests such as WCR adults (Culy et al. 1992, Gyeraj et al. 2021), and on silk water status (Schoper et al. 1986). Therefore, silk feeding, especially combined with heat and water stress, can lead to reduced kernel number and weight and altered kernel size and shape, i.e., maize yield and quality losses.

The damage caused by WCR adult silk feeding depends on the maize variety (thus, genetic background). Inbred lines (for seed production) are highly sensitive to silk clipping due to their lower silk density and vigor. Even one adult per ear may lead to economic losses in inbred lines produced under Central European conditions (Tuska et al. 2002). This aligns with the 1-3 WCR adult threshold for the USA Corn Belt (Culy et al. 1992). Commercial grain hybrids with more dense silks than inbred lines generally tolerate WCR clipping better. Thus, the economic threshold is about 4-6 adults per ear, depending on environmental conditions, in commercial maize in Hungary (Tuska et al. 2003). Irrigation of maize (resulting in higher air humidity) is likely to increase this tolerance by increased pollen viability and silk elongation, as densities of up to 20 adults per ear did not significantly reduce the irrigated maize yield in Colorado, USA (Capinera et al. 1986). Some studies demonstrate that grain maize hybrids respond differently to silk length reduction when assessed for silk growth rate. During these tests, the silks were cut back daily to 25 mm above the ligule of the outer leaf of the husk after the silk appearance. Silk elongation was most intense in the first days, and in some hybrids, the growth rate was 1,5-2 times more rapid than in other hybrids (Bassetti & Westgate 1993).

Sweet maize is a high-value-added commodity; thus, the risk tolerance level of Central European farmers for WCR adults during the silking period of sweet maize is usually low. The sweet maize silking period is challenging due to limitations for chemical insecticide applications considering insect pollinators or biological control agents to manage pests other than WCR. The feasible timing of these applications may differ in 2-4 days depending on the target pest, type of control agent, and insecticide active substance. Therefore, establishing economic thresholds for farmers' control decisions is crucial for integrated pest management (IPM).

Literature data suggest that maize hybrids respond differently to pest damage on their root system (Abel et al. 2000, Ivezic et al. 2006). Thus, the question can be raised whether varieties respond differently to WCR adult silk clipping. We hypothesized that various sweet maize hybrids respond differently to WCR adult silk clipping, and therefore, the action threshold for reducing WCR adult populations may differ. We established parallel field tests with WCR adults in cages and artificial silk cutting. Our approach assessed the presumably high tolerance of sweet maze hybrid Suregold to WCR adult density level and subsequent silk clipping, which might be extrapolated to other sweet maize hybrids. To develop an easier silk clipping field test instead of input demanding caged WCR test, we used artificial silk cutting, a method that could generate applicable information. Suregold sweet maize hybrid with caged WCR adults at various densities and, at the same time, artificial silk cutting was investigated to assess silk damage and the correlation between the adult density and silk cutting. The field test was established with six other sweet maize hybrids with artificial silk cutting only to demonstrate a variable hybrid response to these cuttings and to support establishing WCR adult density action thresholds accordingly.

Material and methods

The study was conducted on seven sweet maize hybrids from 2017-2018 on plots without irrigation in the PlasmoProtect Ltd. research area (47°20′N; 18°51′E) near Martonvásár, Hungary.

The sweet maize plants were grown in an area of 3m (four rows) x 9.2 m (= 27.6 m^2) each study year with a planting density of 55000 seeds per ha. In addition, four rows (3 m) were planted as a border for the study site. Four replicates (length of 9.2 m, 1 row each) were established, where five consecutive plants were selected for silk clipping. Silk clipping levels of 0, 1, and 2 cm for the hybrids were randomized within the replicates. This totaled 20 studied plants, i.e., ears, per each silk cutting level and year (Table 1).

Agronomic and crop management practices were similar in both years and followed the usual practices of the region. Tefluthrin soil insecticide was applied (150 g a.i./ ha) on 29 March 2017 and 10 April 2018. The precrop was oat (*Avena sativa*) in both years.

Weather conditions differed by study year. It was dryer in 2017, and higher temperatures during the pollination and grain-filling period were unfavorable for pollen viability and fertilization. In 2018, the weather during the study period was also dry, but daily maximum temperatures were moderate, and air humidity was adequate during the grain-filling period (Figure 1).

Table 1. Silk clipping and manual harvest dates in the seven investigated sweet maize hybrids.

Hybrids	2017		2018	
	Silk clipping/cutting experiment	Manual harvest	Silk clipping/cutting experiment	Manual harvest
Suregold	04-18 July	30 July	03-19 July	27 July - 01 Aug
Kinze	04-18 July	29-30 July	01-15 July	24-27 July
Mv Július	25 June - 12 July	24 July	21 June - 06 July	18-22 July
GSS 5649	06-21 July	31 July - 01 Aug	30 June - 15 July	23-27 July
GSS 8529	06-21 July	02-03 Aug	04-17 July	27-29 July
Moreland	09-23 July	01 Aug	05-18 July	28 July - 01 Aug
GH 11754	06-21 July	31 July	02-18 July	28 July - 01 Aug

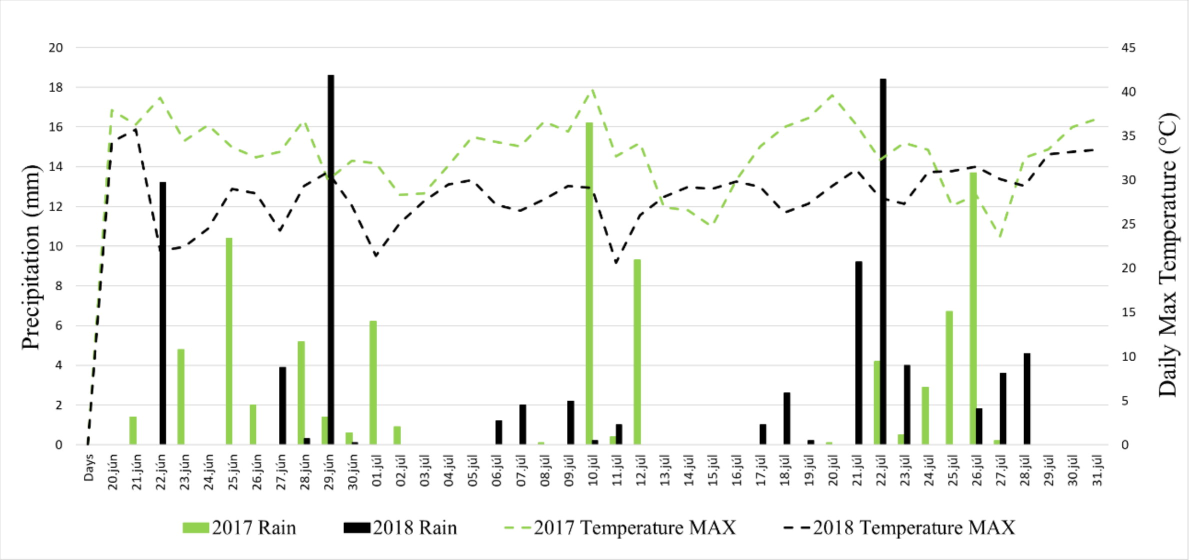

Figure 1. Daily precipitation and maximum temperatures of the study location during the silk clipping experiments, the actual silking period in each hybrid (see also Table 1), in 2017 (green) and 2018 (black), Martonvásár, Hungary.

The sweet maize plants (with adult WCRs and artificial silk cutting) were monitored daily at the tasselling stage VT from the end of June (Abendroth et al. 2011). This stage is when the tassel is completely visible, the plant has reached its full height, and it begins to shed pollen. The development of individual ears was followed, and when silks appeared, the daily silk cutting commenced the R1 stage (Abendroth et al. 2011). The R1 stage occurs about three days after the tasseling stage. This was followed to ensure that the experiment started before the first silks emerged from the tip of the husk leaves, i.e., right before the beginning of the R1 stage of each studied plant. Ears (except for the caged WCR test on Suregold) were not isolated in the experiment because we also wanted to monitor the natural WCR adult infestation. We cut back the silks above the tip of the husk leaves to 0, 1, and 2 cm to simulate different infestation levels and subsequent silk feeding by WCR adults and measured silk length on uncut control plants daily until the end of silking in 2017 and 2018. Twenty uncut ears of control plants were also marked as untreated each year. To monitor the natural WCR adult infestation in the experimental plots, three Pherocon AM yellow sticky traps (Trécé Inc, USA) were placed in the research area, and the number of captured adults was recorded weekly since this may have affected uncaged silk length. The traps were replaced weekly.

All investigated sweet maize cobs were harvested manually, separated, stored in crates, and transported to the laboratory (Table 1). The weight of the husked cobs was measured for each treatment and hybrid.

We also estimated hypothetical WCR adult densities for each cob based on weight. First, we determined the pest silk clipping – weight loss relationship in Suregold hybrid plants for both years: we fitted two linear regression models (Figure 3, left panel). Then, we calculated the weight difference of the clipped cobs vs. the corresponding uncut control cobs (corresponding year and hybrid), and these weight loss values were used to insert in the above-mentioned regression models to estimate the hypothetical WCR adult densities for silk cut hybrids. Data visualization and regression model fits were performed using R (R Core Team, 2019)

Results

Over the two years of the study, there were low to moderate natural WCR adult population levels in the area during the maize silking period. After the appearance of the silk on the earliest sweet maize hybrid, we monitored weekly the number of natural WCR adult infestations with the Pherocon AM trap until the silk drying of the latest hybrid (Table 2).

Table 2. Average natural WCR adult infestation as a rounded number of trap captures.

2017 trap check dates	01-July	08-July	15-July	22-July
Diabrotica adults per Pherocon AM trap	5	11	15	23
2018 trap check dates	28-June	05-July	12-July	19-July
Diabrotica adults per Pherocon AM trap	3	6	7	4

The moderate WCR adult density (3 – 23 adults/trap/week on Pherocon AM cards) corresponds to an estimated 0.25-0.5 adult/maize plant (Bazok et al. 2011) and, thus, was not likely to bias the results of our experiment.

Effect of artificial silk cutting on yield parameters of the seven sweet maize hybrids:

Suregold: In 2017 (dry season, less rainfall), there were no significant yield differences among uncut silks, and those cut back to 1 and 2 cm. However, those cut back to 0 cm resulted in a yield decrease like that measured at 8 to 12 adult WCR/ear infestation levels. In 2018, there were no significant differences among treatments (cutting or WCR adult density) due to more favorable weather conditions.

Kinze: In 2017, silks cut back to 0 and 1 cm resulted in a significant yield decrease, while in 2018 (favorable weather conditions), there were no yield differences.

MV Július: There were no differences both in 2017 and 2018 among treatments.

Moreland: Silk cut back to 0 cm resulted in a significant yield decrease in both years.

The same hybrid response was measured for hybrids GSS5649, GSS8629, and GH11754. For the latter, weak plant establishment due to early drought did not allow data generation in 2018.

It is important to note that the impact of weather conditions in the growing season on yield, specifically in the silking period, is important for each hybrid (Figure 2).

The yield response of the Suregold hybrid to different WCR adult densities suggests that this hybrid may tolerate WCR adult silk clipping up to 8-12 adults per ear; for details, see Gyeraj et al. (2021).

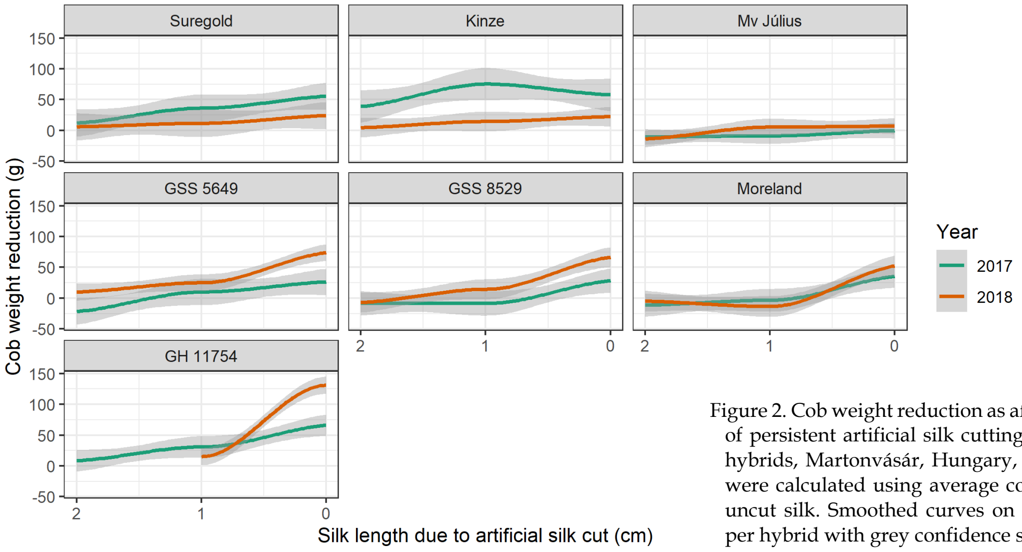

Figure 2. Cob weight reduction as affected by 0, 1 and 2 cm of persistent artificial silk cutting in seven sweet maize hybrids, Martonvásár, Hungary, 2017-2018. Reductions were calculated using average control cob weight with uncut silk. Smoothed curves on 120 data points (cobs) per hybrid with grey confidence stripes were plotted.

Comparisons of the effect of WCR adult densities and artificial silk cutting on yield parameters for Suregold hybrid: This hybrid tolerated the cutting reduction of silk length well. Cutting back silks to 1 and 2 cm did not result in significant cob weight loss in either of the two years. Cutting back to 0 cm of length may result in considerable cob weight loss only in unfavorable (dry) weather conditions (Figure 3).

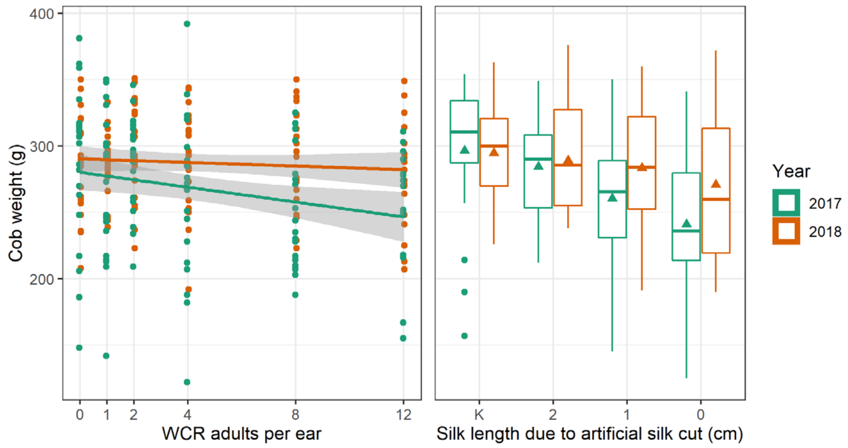

Figure 3. Correlation of cob weight in Suregold sweet maize hybrid by WCR adult damage (left panel) and artificial silk cutting (right panel) to 0, 1 and 2 cm, as well as data of cobs with uncut silk (K) in 2017 and 2018, Martonvásár, Hungary.

Discussion

The two-year study was conducted on seven different sweet maize hybrids in a non-irrigated sweet maize field. There were low to moderate WCR adult population levels in the study area during the silking period in both years. After the appearance of the silks, we started with the artificial silk cutting and cut the silks above the tip of the husk leaves to 0, 1, and 2 cm to mimic/simulate different infestation levels and subsequent silk feeding by WCR adults. Silk length was measured on uncut control plants daily until the end of silking. Since quality requirements for harvested sweet maize are much higher than grain maize, establishing action thresholds for various pests directly or indirectly impacting the quality is necessary.

The WCR economic damage threshold levels determined for seed maize or non-irrigated commercial grain maize were much lower (Tuska et al. 2002, 2003) than the WCR adult levels that caused slight fertilization decreases in our sweet maize study. Moreover, the observed silk clipping did not lead to cob weight reductions even at 12 WCR adults per ear pest density, while under certain conditions, much lower WCR adult number (in seed maize 1-3 adults per ear and in grain maize 4-6 adults per ear Tuska et al. 2002, 2003) may affect grain yield and quality. Pest action thresholds and farmers' risk tolerance levels are often lower in value-added crops. However, our results suggest the opposite for sweet maize compared to grain maize because of sweet maize silk regeneration and the increased probability of fertilization. Of the study years, the dry conditions in 2017 negatively influenced silk regeneration after the adult damage and had a negative effect on fertility and yield results. This is in line with the findings of Schoper et al. (1986). Yet, these dry conditions showed less impact on ear fertilization than noted in inbreds and commercial grain maize. Since sweet maize is mostly cultivated under irrigation, dry weather conditions are compensated by irrigation. Our results justify that irrigation timing may contribute to reducing silk clipping consequences and therefore decrease insecticide application during fertilization. In addition, if biological control solutions are used during the silking period, knowledge of high WCR action threshold levels associated with hybrid tolerance to adult WCR silk clipping helps farmers reduce synthetic insecticide applications.

However, genetic differences can lead to different responses among maize hybrids, as shown for WCR larval damage by Abel et al. (2000) and Ivezic et al. (2006). Therefore, information on yield loss response for WCR by various hybrids under specific regional and/or local conditions may need to be conducted along with varietal tests or demonstration trials.

Artificial WCR silk clipping (silk cutting) showed different responses based on maize hybrid. Surprisingly, silk cutting to 1 cm resulted in moderate yield reduction. Significant yield reduction was measured only at 0 cm cutting. Thus, the action threshold, in general, for adult control only is justified at high adult population levels for sweet maize hybrids. Response of individual sweet maize hybrids to WCR adult damage may be slightly different; thus, this should be considered according to local conditions (multiple abiotic and biotic stressors, e.g., other pests), but usually, there is no need for WCR adult control treatments. This favors reduced insecticide application, environmental load, and increased consumer safety.

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