Maize Tolerance to Western Corn Rootworm Larval Feeding: Screening through Five Years of Investigation

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Summary

Research on maize tolerance to rootworm larval feeding was conducted in Osijek, Croatia in period from 2001 to 2005. Seven Croatian commercial maize hybrids were evaluated by measuring traits associated with resistance: root injury, root size and root regrowth. Hybrids were grown in monoculture in field trials in four replicates. Root injury was rated according to Iowa Node Injury Scale (0-3), and the Eiben 1-6 Scale, reversed, was used for root size and regrowth assessment. No significant differences were observed for root injury between the hybrids. However, injury was significantly different across the years and more damaged roots were observed in 2003 (1.606), 2004 (1.281) and 2005 (0.940) compared to 2001 (0.081) and 2002 (0.099). Root size and root regrowth differed significantly across the years of investigation, while differences between the hybrids occurred only for root regrowth. Hybrids OsSK 617, OsSK 602 and OsSK 596 appeared to be the most tolerant through all years of investigation measured by root regrowth.

Key words

western corn rootworm, larva, maize hybrids, tolerance

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Introduction

Damage in maize fields caused by Western corn rootworm (WCR) (*Diabrotica virgifera virgifera* LeConte) larvae becomes more severe and intense with every subsequent vegetation period. By feeding on the roots, larva is reducing the ability of the plant to absorb water and nutrients from soil. Additionally, plants are lodged and difficulties in harvest occur. WCR is a major maize pest in the United States but recently it became a serious threat to European maize production as well. Once detected, it is very difficult to eradicate as well as to control the pest. American farmers are fighting this pest over the past 60 years. Insect has shown an ability to evolve resistance to several control measures such as insecticides (Meinke et al., 1998; Wright et al., 2000) and agricultural practices (Levine et al., 2002).

In Europe, the first record of WCR was reported from Yugoslavia in 1992 (Bača, 1994). The pest was identified in Croatia in 1995 and infested area is considered to be of approximately 23.500 km^2 (Igrc-Barčić & Bažok, 2004). Currently, the main areas of economic damage in Europe are southeastern countries like Croatia, Serbia, Hungary, Slovakia and Italy (Hummel et al., 2008). Already in 1999, some maize fields in Eastern Croatia, close to Serbian border, were infested with 0.37 to 0.51 beetles per plant (Ivezić et al., 2001). However, there were no visible lodging. The first visible damages on larger scale in Croatia were reported in 2002 (Dobrinčić et al., 2002; Igrc Barčić et al., 2003).

Research on resistance to WCR started 70 years ago in USA, in Corn Belt as an alternative to insecticides or other management tactics (Bigger et al., 1938, 1941). The same authors report the difference in plant reaction to larval feeding already in 1920's however in 1941 certain lines had superior tolerance to rootworms. The resistance by Owens et al. (1974) has been found due to larger root system and greater secondary root development. Tolerance, conferred by larger root size and greater root regrowth, has been identified as the primary plant defense mechanism. A tolerant plant sustains as much feeding damage as a susceptible plant, but still it is able to develop and produce high grain yield regardless of the injury (Riedell & Evenson, 1993).

The potential and severity of WCR spreading is recognized by research units in EU infested countries and many of them initiated the research to discover the most efficient control measures for their production systems. The first research concerning classical resistance breeding to WCR in Europe was carried out in Croatia at the Faculty of Agriculture in Osijek (Moeser and Hibbard, 2005). Ivezić et al. (2006) conducted comparable investigations in US and Croatia, and reported results of several Croatian commercial hybrids that have shown tolerance to WCR larval feeding. Many studies report on response of maize to WCR larval feeding. However, Bohn (2006) reported that there were no germplasm sources with superior WCR resistance under moderate to high insect pressure.

Recently, genetically engineered maize (*Bacillus thuringiensis* Berliner (Bt)) was developed as a modern solution for WCR control and an alternative to chemical control and crop rotation. Bt maize for corn rootworm control is not registered for cultivation in Europe, so development of maize hybrids resistant to WCR would represent environmentally friendly and sustainable WCR management tool as well as a challenge and necessity for the scientists and maize growers to produce maize with no need for insecticide appliance.

The objectives of this research were to evaluate tolerance of seven commercial Croatian hybrids that are widely used by farmers under natural infestations with corn rootworms larval feeding through five years of investigation.

Material and methods

Field experiments were conducted in period of five consecutive years from 2001 to 2005 at Agricultural Institute Osijek, Croatia (45°32"N, 18°44"E). Fields with history of natural WCR infestation were chosen. Monthly precipitation data was obtained from country's Meteorological and hydrological service. Population of WCR beetles was monitored weekly by pheromone traps (PAL) placed in each replication, and replaced every 25 days. The experimental design at each location was a randomized complete block with four replications. Hybrids were grown in monoculture and planted each in four rows plots 6 m long with 2 seeds per hill, 25 cm between hills, and 70 cm row spacing. Plots were thinned to one plant per hill. Maize hybrids (Agricultural Institute Osijek, Dept. for Maize Breeding and Genetics, Osijek, Croatia) commercially often planted in Croatia are chosen for current research: Os 499, OsSK 444, OsSK 552, OsSK 596, OsSK 602, OsSK 617 and OsSK 644.

Five competitive plants in each plot were chosen, tagged, and dug from the soil in the middle of July. The root systems were cleaned with pressurized water and rated for rootworm larvae feeding injury, root size and root regrowth. Root injury was rated according to the Iowa State University 0-3 Node-Injury Scale as follows: 0.00 – no feeding damage; 1.00 – one node, or the equivalent of an entire node, eaten back to within approximately 5 cm of the stalk; 2.00 – two nodes eaten; 3.00 – three or more nodes eaten. Damage between complete nodes eaten is noted as the percentage of the node missing, e.g., 1.50 = one and half nodes eaten and 0.25 = one quarter of node eaten (Oleson and Tollefson, 2000). The Eiben 1-6 Scale, reversed, (Rogers, 1975) was used to rate root size (root system) and regrowth (developed secondary roots). Scales for root size and regrowth were created by choosing roots form the smallest to the largest roots. On the reversed scales a rating of 1 indicated a large root system or well developed secondary roots, and a rating of 6 indicated a small root system or poorly developed secondary roots. Data for root injury, root size, and root regrowth rating were analyzed by location and hybrids using analysis of variance. Response variables were analyzed as least squares means, and means were separated using the least significant differences (LSD) of the least squares means.

Results and discussion

Results of monitoring of WCR beetles on traps were as follows: 950 in 2001, 1082 in 2002, 950 in 2003, 2138 in 2004, and 656 beetles in 2005. Population levels varied among the years. Interestingly, we caught the highest population level of WCR beetles in 2004, after very dry 2003 year, reveling capability of WCR eggs to survive dry summer months before fall and winter rainfall.

In an analysis of variance combined across years, root injury and root size differed significantly only between the years, while root regrowth differed significantly both among the hybrids and the years (Tab. 1).

Root injury was found to be significantly different between the years of investigation, while no differences occurred among hybrids (Tab. 2 and 3). In 2001 and 2002, injuries were significantly lower compared to the following three years, as expected since these were the first two years of monoculture.

Table 1. Combined ANOVA and LSD test

Variable	df	Root Injury	Root Size	Root Regrowth
Hybrids	6	n.s.	n.s.	**
Years	4	**	**	**

** P<0,01; n.s. no significant differences

Table 2. Average hybrids ratings for root injury, root size and root regrowth

Hybrid	Root Injury	Root Size	Root Regrowth
Os 499	0.825	2.660	3.410
Os SK 444	0.818	2.888	3.930
Os SK 552	0.843	2.944	2.660
Os SK 596	0.754	2.760	2.290
Os SK 602	0.774	2.738	2.120
Os SK 617	0.698	2.570	1.700
Os SK 644	0.899	2.690	2.510
LSD 0.05	n.s.	n.s.	0.605
LSD 0.01	n.s.	n.s.	0.829

Table 3. Root injury, root size and root regrowth ratings

Year	Root Injury	Root Size	Root Regrowth
2001	0.081	1.529	2.671
2002	0.099	2.193	2.171
2003	1.606	2.829	2.479
2004	1.281	3.343	2.236
2005	0.940	3.857	3.743
LSD 0.05	0.241	0.381	0.403
LDS 0.01	0.317	0.501	0.531

In 2003, almost all hybrids, except OsSK 596, had the most damaged roots indicating non favorable conditions for maize plants in the year of production (Tab. 5). Plants suffered drought stress in 2003, since it was the year with the lowest precipitation during the vegetation period with only 230 mm of rainfall. Root injury ratings in 2005 were significantly lower then in 2003 and 2004, and significantly higher compared to 2001 and 2002 (Fig. 1). Greater root damage increases plant lodging and potentially reduces yield (Gray and Steffey, 1998).

Table 4. Grain yields (t ha^{-1})

Hybrid	2001	2002	2003	2004	2005
Os 499	11.89	11.60	7.72	10.82	9.26
Os SK 444	11.27	9.70	5.90	10.46	7.90
Os SK 552	11.94	11.40	6.15	10.82	8.96
Os SK 596	11.50	11.70	7.15	10.18	10.93
Os SK 602	13.39	13.20	9.35	11.97	10.70
Os SK 617	12.66	12.20	7.28	11.76	10.20
Os SK 644	11.12	11.40	7.88	8.78	10.69

Table 5. Precipitation (mm) in Osijek during the vegetation period

Month	2001	2002	2003	2004	2005
April	72	71	12	149	53
May	60	135	18	74	50
June	239	36	44	71	110
July	77	58	60	40	173
August	7	80	42	94	237
September	195	81	54	49	75
Total	650	461	230	477	698

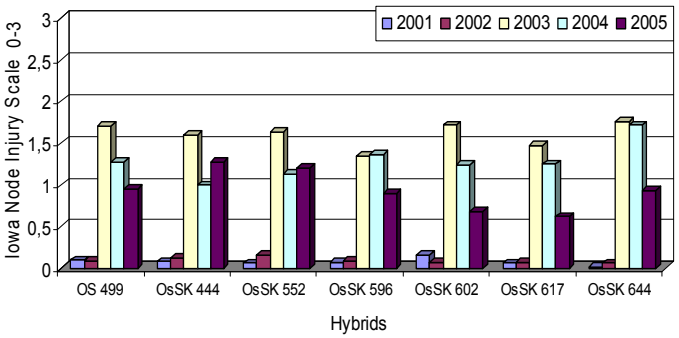

Figure 1. Root injury ratings

Root size differed significantly between the years of investigation, while there were not significant differences among hybrids (Tab. 2 and 3). Root size ratings per each hybrid and years are given in Figure 2. External factors, such as weather conditions, or undertaken agro technical measures, may have impact on root size. Size of a root system as shown in Tab. 2, decreased in every subsequent year of monoculture, implying that well WCR population might have been well established and have had influence on root size.

Root regrowth as a main parameter indicating tolerance, since it appears to be strongly related to corn rootworm, differed significantly between years and hybrids (Tab. 2 and 3). According to Gray and Steffey (1998), root regrowth can be considered a parameter that potentially can predict yield. For all years of investigation hybrid OsSK 617 had proved to have

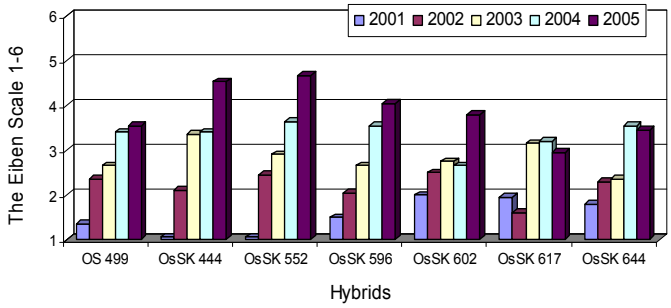

Figure 2. Root size ratings

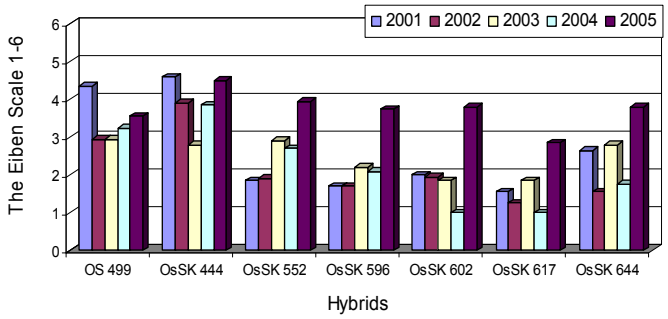

Figure 3. Root regrowth ratings

the highest root regrowth rating (1.700), i.e. well developed secondary roots (Fig. 3) with satisfactory grain yield (Tab. 4). Same hybrid had the lowest root injury (0.698) and the largest root size (2.570) in the fourth year of monoculture. However in 2003, in year that was characterized with unfavorable environmental conditions (Tab. 5), hybrid OsSK 602 proved to be the most productive with highest grain yield achieved (9.35 t ha^{-1}) (Tab. 4). Hybrid OsSK 602 had the second best root regrowth rating and seemed to be more tolerant to drought. Similar results were reported in investigation by Ivezić et al. (2006). Selection for larger root systems may result in superior secondary root development, reduced feeding damage, and reduced lodging (Gray and Steffey, 1998).

Conclusions

Hybrids OsSK 617, OsSK 602 and OsSK 596, according to the root regrowth ratings, were the most tolerant in the first four years of monoculture. Therefore, these hybrids may be recommended to the farmers who grow maize in continuous cropping. Nevertheless, monoculture should be avoided since tolerant plants do not affect WCR population build up. High root regrowth seemed to provide plants with additional ability to assimilate nutrients after injury especially for hybrid OsSK 602, which managed to develop secondary roots and achieve the highest yields in the year with inadequate precipitation level. Research of the response of maize to WCR larval injury will provide information for creation of new, more tolerant or resistant hybrids to WCR. Detected tolerant hybrids should be involved in future research in order to detect the source of tolerance.

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