ROOT DAMAGES AND ROOT MASS UNDER CONDITIONS OF ARTIFICIAL INFESTATION WITH WESTERN CORN ROOTWORM EGGS

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Abstract. Maize production represents one of the most important segments for agriculture in Serbia. The Western corn rootworm - WCR, Diabrotica virgifera sp. virgifera (Col., Chrysomelidae), is the most dangerous maize pest, causing severe losses worldwide. Field experiment was carried out in Bečej (Vojvodina province, Serbia). In the field, 96 plants were selected, and arranged in 48 pairs. In each pair one plant represented artificially infested plant (D plant), and the second plant was the control plant (C plant). Damages and root mass of the roots were measured and evaluated in September 2016 and August 2017. Root damages were ranked from 1 to 6, according to Ostlie and Notzel (1987) scale. The root mass was measured on a technical balance. According the mentioned scale, 93.75% of D and C plants in 2016 and 95.83% of D and C plants in 2017 were with different level of root injuries. The number of D plants with rate 6 in 2017 was higher for 27.09% then in 2016, while the number of C plants with rate 6 in 2017 was higher only for 2.08% compared to 2016. The number of both, D and C plants with rate 1 in 2016 and 2017 was 3 and 2, respectively. The differences between damages on D and C plants, based on the root damage and root mass were analyzed using non-parametric Kruskal-Wallis test (One-way ANOVA). Statistical analysis of damages and root mass in 2016 shows that there are no significant differences between D and C plants, while in 2017 differences between D and C plants were statistically highly significant, with more damages on D plants. The differences in damages and root mass between D plants in 2016 and 2017 were statistically highly significant. Statistical analysis shows that there are no significant differences between C plants in 2016 and 2017 based on root damages while differences on root mass were statistically hihgly significant.

Keywords: Maize, WCR, level of damages, root mass.

INTRODUCTION

In the last 30 years agriculture production in Serbia has gone through same qualitative changes, which reflected in an increase of the productivity level (STEVANOVIĆ ET AL., 2012). Maize production represents one of the most important segments of agriculture in Serbia (ĐOROVIĆ ET AL., 2006). It is crucial for cattle production and processing industry, which together constitute a precondition for diversification of the agro-industrial sector (ĐOROVIĆ ET AL., 2006). Maize, as one of most important crops in Serbia, occupies 40% of total planted area (KOS ET AL., 2013). Serbia is one of the largest maize producers and exporters in Europe, and belongs to first seven producers (KOS ET AL., 2013). The maize yields on the territory of Serbia during 2012, 2013, 2014, 2015 and 2016 are 3.533.000 t, 5.864.000 t, 7.951.583 t, 5.454.841 t, 7.376.738 t, respectively (Statistical Office of RS). It is used mainly for animal feed (80%), but also for human consumption and in starchy production (KOS ET AL., 2012).

Maize is exposed to a large number of pests in the field, which cause huge losses of yield and reduction of quality of maize stem (OERKE, 2006). One of the most serious and economically most important maize pests is Western corn rootworm, firstly in USA and from the end of XX century in Europe (TOLLEFSON, 2007; MAHMOUD ET AL., 2016). Western corn rootworm (WCR) *Diabrotica virgifera* sp. *virgifera* Le Conte (Col., Chrysomelidae) is an oligophfaguos pest, native in America (BERMOND ET AL., 2012). The first written information

about identification of WCR in Europe are connected to Serbia, former Yugoslavia, in the early '90, near the Belgrade airport (BAČA, 1993). This pest has spread from Serbia to almost every maize field in Europe (HUMMEL ET AL., 2008; LEMIC ET AL., 2015). The rate of WCR adults spread per year is up to 100 km (BAUFELD ET AL., 2001). Both, adults and WCR larvae, attack the maize plants leaving serious consequences. WCR larvae feed on the maize root system causing the most important damages (WESSELER AND FALL, 2010). Adults feed on young leaves and maize silk resulting in smaller damages compared to WCR larvae (IVEZICH ET AL., 2011; RASPUDIĆ ET AL., 2013). WCR larvae feed on the nodal and lateral roots (CHIANG, 1973) and for that reason, they have the significant effect on nutrient assimilation on maize plant and on yield (KAHLER ET AL., 1985). One of the most reliable symptoms that indicate the presence of WCR in field is, so called, symptom of "goose neck" (CHIANG, 1973; WESSELER AND FALI, 2010). Larvae that feed on root system can cause inability of maize to uptake water and nutrients (GAVLOVSKI ET AL., 1992; GRAY, 2009).

Maize tolerance to the presence of WCR larvae in the maize field is reflected as a strong, large root system and the amount of secondary roots (BRANSON, 1986; GRAY AND STEFFEY, 1998). The level of WCR larvae damages are highly depended of environmental conditions, the type of the soil and number of larvae in the soil (CIOBANU ET AL., 2009; SPIKE AND TOLLEFSON, 1989). Beside climatic conditions, maize monoculture represents one of the main reasons for increasing WCR population in the field (SPENCER ET AL., 2009). Inability to harvest the maize kernel because of the plant lodging is one of the reasons for maize yield losses (SPIKE AND TOLLEFSON, 1991; ESTES ET AL., 2015; PEREIRA, 2015). One of the most important reasons for WCR presence in fields is a monoculture (CHIANG ET AL., 1969), which contributes to the increase of WCR population in maize fields (SIVČEV ET AL., 2009). Larval mobility in the soil is less than 50 cm, which points out that they cannot survive in the field with crop rotation (BAČA ET AL., 1998). The presence of WCR in the field can cause plant logging up to 15% (CHIANG ET AL., 1969; ČAMPRAG, 1997) and yield loses up to 70% (LEVINE ET AL., 2002). Maize is a plant with a high ability to grow new roots after larval attack (GRAY AND STEFFEY, 1998), so root regrowth represents the main defense mechanism (RIEDELL AND EVENSON, 1993). WCR shows tolerance to pesticide control (WRIGHT ET AL., 2000).

The aim of this research was to examine the impact of WCR larvae in condition of artificial infestation on maize root system based on root damages and root mass.

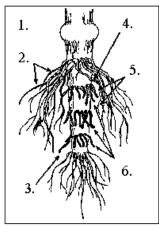
MATERIAL AND METHODS

The field experiment was carried out in Bečej, Vojvodina province in the north of Serbia. It was performed from May 18th to September 10th in 2016 and from May 14th to August 15th in 2017, with serbian cultivar NS-640. The chosen filed for the experiment represents filed with a low WCR natural infestation.

During the experiment, 96 maize plants were selected, labeled and arranged into pairs. The plants are set in two rows with space between labeled plants of 1 m. In each pair, one plant was artificially infested in the root zone with 4mL of WCR eggs in 0.125% agar suspension (D plants). One mL of suspension contained 136 eggs. The other plant from the pair was the control plant, marked with C. In the root zone of the C plant, the same amount of distilled water (4mL) was injected.

During both experimental years, the field was inspected regularly, once a week. In each field inspection the number of maize plants and the presence of "goose neck" symptom were recorded. In the last field inspection, September i.e. August, the maize root damages caused by WCR larvae were evaluated.

The root inspection was conducted as follows: all marked plants were excavated, the soil was removed from roots and the roots were rinsed with water. After the mentioned preparation, root damages were ranked from 1 to 6 (Fig. 1), according to scale recommended by Ostlie and Notzel (1987). Also, the dray root mass was measured on a technical balance (Kern EW 1500-2 M).



Root rating scale (Ostlie and Notzel, 1987)

- 1 No feeding damage
- 2 Visible feeding scars present
- 3 At least one root chewed to within 1.1/2 inches of plant
- 4 One entire node of roots destroyed
- 5 Two nodes destroyed
- 6 Three or more nodes destroyed

Fig.1. Root damages and marks according to scale (Ostlie and Notzel, 1987)

The differences between damages on D and C plants, based on the root damage and root mass, were analyzed using non-parametric Kruskal-Wallis test (One-way analysis of variance) in statistical software SPSS 17.

RESULTS AND DISCUSSION

According to data, only three D plants i.e. 6.25% were with the healthy root system (rate 1). The root injuries rated as level 2 were recorded on seven D plants i.e. 14.58%. The highest number of D plants, 12 i.e. 25%, was with rate 3. Rate 4 was registered on 7 D plants i.e. 14.58%, and rate 5 on eight plants i.e. 16.6%. The strongest and the most visible damages, rate 6, were recorded on 10 D plants i.e. 20.83% (Figure 2.)

From the total number of 48 plants, only three C plants i.e. 6.25% was with rate 1. Root injuries rated as 2, were recorded on three C plants i.e. 6.25%. Rate 3 damages were registered on 13 C plants i.e. 27.08%. Five C plants i.e. 10.41% had the rate 4, while 13 C plants i.e. 27.08% was with rate 5. The most destroyed root system, rate 6, was registered on 10 C plants i.e. 20.83% (Figure 2.)

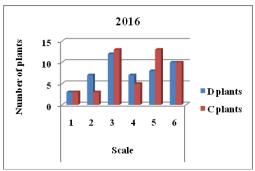
According the OSTLIE AND NOTZEL (1987) root scale, from the total number of plants (96 i.e. 48 D and 48 C plants) during 2016, 93.75% in both categories were with different root injuries caused by WCR larvae.

Statistical analysis in 2016, using non-parametric Kruskal-Wallis test, showed that there were no significant differences between root damages and root mass of D and C plants (Table 1).

The smallest measured root mass during 2016 on D and C plants was 93.12 g and 67.7 g, respectively. In the same year the biggest measured root mass on D and C plants was 563.44 g and 546.03 g, respectively. Average root mass in 2016 on D and C plants were 263.8 g and 233.7 g, respectively.

The obtained results in the vegetation period in 2017 are presented in the Figure 3.

From the total number (48) of infested plants only two, i.e. 4.16% had healthy root system. With rate 2, i.e. 6.25%, three D plants were registered. Root injuries with rate 3 were recorded on eight D plants i.e. 16.6%. Rate 4 and rate 5 were registered on three D plants (6.25%) and six D plants (12.5%), respectively. The highest number of D plants, 23 i.e. 47.92%, was registered with the highest root injuries rate 6 (Fig. 3).



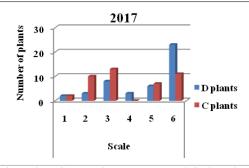


Figure 2. WCR root damages on D and C plants in 2016

Figure 3. WCR root damages on D and C plants in 2017

The recorded number of C plants with healthy root system (rate 1) was two i.e. 4.16% of plants. Ten C plants, i.e. 20.83%, had root damages rate 2. The biggest number, 13 C plants i.e. 27.08%, was with the rate 3, while the rate 4 was not registered in the experimental field. Rate 5 was registered on seven C plants, i.e. 14.58%. The most destroyed root system (rate 6) was recorded on 11 C plants i.e. 22.91% (Fig. 3).

According the OSTLIE AND NOTZEL (1987) root scale, from the total number (96 D and C maize plants) in 2017, the number of destroyed roots with different root injuries was also 93.75%.

The smallest measured root mass during 2017 on D and C plants was 4.46 g and 4.82 g, respectively. In the same year the biggest measured root mass on D and C plants was 44.96 g and 29.61 g, respectively. The average root mass in 2017 on D an C plants was 12.86 g and 12.82g, respectively.

The root damages were high in 2017, and the differences in root damages and root mass between D and C plants were statistically highly significant, at level importance p<0.001 and p<0.01, respectively (Table 1).

Table 1.

Statistical differences between damages and root mass of D and C plants							
V	Parameter	Mean Values		C:-			
Year		D	С	Sig.			
2016	Root mass	$263.8 \pm 123.9345a$	233.7 ±134.0324a	0.164 ns			
2010	Root damage	$3.851 \pm 1.573955a$	4.106 ±1.506999a	0.421 ns			
2017	Root mass	$12.86 \pm 13.88506a$	$12.82 \pm 7.81303b$	0.014 *			
2017	Root damage	5.84 +5.228712a	3.767 +1.688014b	0.005 **			

The different level of root damages on D plants in 2016 and 2017 is presented in Figure 4. The number of D plants with highly destroyed root system (rate 6) in 2017 was higher for 27.09% then in 2016. The number of plants with healthy root system during 2016 and 2017 was similar, 3 and 2, respectively. The most of D plants (12) in 2016 were with root damage rate 3, while during 2017 the most plants (23) were with damages rate 6 (Figure 4.).

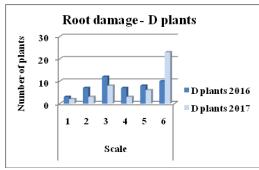


Figure 4. WCR root damages on plants in 2016 and 2017

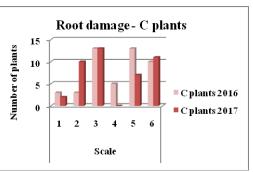


Figure 5. WCR root damages on C plants in 2016 and 2017

The different level of root damages on C plants in 2016 and 2017 is presented in Figure 5. The number of C plants with highly destroyed root system (rate 6) in 2017 was higher for 2.08% then in 2016. During both experimental year the highest number 13 plants, was whit the rate 3. The number of D and C plants with healthy root system in 2016 and 2017 was almost equal i.e. three and two plants, respectively.

Table 2. Differences between damages and mass of root system of D and C plants in 2016 and 2017

Bifferences between damages and mass of foot system of B and C plants in 2010 and 2017						
Year	Parameter	Mean Values		C: a		
i ear		2016	2017	Sig.		
D mlomto	Root mass	$263.8 \pm 123.9345a$	$12.86 \pm 13.88506b$	0.001 **		
D plants	Root damage	$3.851 \pm 1.573955a$	$5.84 \pm 5.228712b$	0.005 **		
C1	Root mass	$233.7 \pm 134.0324a$	$12.82 \pm 7.81303b$	0.001 **		
C plants	Root damage	4.106 ±1.506999a	$3.767 \pm 1.688014a$	0.302 ns		

Although, the root injuries during both experimental years were very high, the differences in root mass and root damages between D plants were statistically highly significant at level importance p<0.001 (Table 2).

Statistical analysis showed that there were no significant differences between C plants in 2016 and 2017 based on their root damages. On the other hand, the differences between the root mass were statistically very significant at level of importance p<0.001 (Table 2).

Artificial infestation with WCR eggs, as reported by TANASKOVIĆ ET AL. (2016), caused 95.7% of root damages on D plants and 47.9% on C plants with different damage rates. In this research, we registered 47.93% less damaged C plants compared to the research of TANASKOVIĆ ET AL. (2016). The level of damaged root of D plants is almost equal with data in TANASKOVIĆ ET AL. (2016).

The smallest root mass in research TANASKOVIĆ ET AL. (2016) registered on D and C plants in 2015, and were 26.88 g and 22.17 g, respectively. The same authors had recorded the biggest root mass 144.4 g and 142.9 g on D and C plants, respectively. Obtained results in 2016 indicate bigger root mass for both plants group comparing with the results TANASKOVIĆ ET AL. (2016).

During experimental period, 2017 was year with more dry and warm days then 2016, which caused higher WCR population, bigger root damages and less root mass. This is in accordid with GROZEA ET AL. (2009) reseatch.

CONCLUSIONS

Comparing the root damages and root mass, statistical analysis shows that there were no differences between D and C plants during 2016. The differences between root damages on D and C plants in 2017 were higher than differences in their root mass. Statistical analysis shows very significant differences in 2016 and 2017 on D plants based on root damages and root mass at level of importance p<0.001. Differences between C plants in both years based on their root damages are not significant, while differences based on root mass were very significant at level of importance p<0.001.

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