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THE ROLE OF BUFFER ZONES IN ENSURING THE COEXISTENCE OF GM AND NON-GM MAIZE

Viorica URECHEAN[1], Dorina BONEA[2]

[1]Agricultural Research and Development Station Simnic, 54 Balcesti Road, Dolj, Romania
[2]University of Craiova, Faculty of Agronomy, Department of Agricultural and Forestry Technology, 19 Libertatii Street, Dolj, Romania

Corresponding author email: dorinabonea@gmail.com

Abstract

Cultivation of genetically modified (GM) maize MON 810 in the European Union, including Romania, requires specific management. The adventitious presence of genetically modified organisms in conventional crops, can affect their coexistence. Two field experiments with genetically modified maize (MON 810) and conventional maize (non-GM) were placed at A.R.D.S. Simnic- Craiova under the climatic conditions of the agricultural years 2011-2012 and 2012-2013. The rate of cross-pollination (highlighted by percentage of Xenia on the ear) was influenced by: the buffer zones, the isolation distance and the prevailing wind direction. The presence of buffer zones (ten row of Sudan Grass) has reduced the cross-fertilization rate by 40.5% in 2012 and by 20.0% in 2013. As a result, the use of buffer zones can be considered one of the important strategies for ensuring coexistence a the genetically modified maize with non-genetically modified maize, in the same area of culture.

Key words: *buffer zones, cross-pollination rate, GM and non-GM maize, percentage of xenia.*

INTRODUCTION

Biotechnology has major implications in agriculture, food production and processing, and medicine.

Genetically modified maize MON810 resistant to *Ostrinia nubilalis*, obtained by transgenesis is authorized for cultivation in the EU, including Romania.

According to the Ministry of Agriculture and Rural Development (MADR 2017), in 2007, the Romanian farmers cultivated 332.5 hectares of genetically modified maize, after which the surface increased to over 6,100 hectares in 2008, and then the surfaces continuously decreased yearly.

Since 2016, farmers have completely abandoned MON 810, probably because of the very complex rules on product traceability.

To ensure that the development of biotechnology and GMOs, in particular, is made safe, the EU has set up a legal framework comprising several legislative rules.

Coexistence refers to certain rules that provide farmers with the practical possibility to opt for conventional, organic or GM crops, in line with legal obligations for labeling and purity standards (EU, 2006).

Recommendation 2010/C. 200/01 on guidelines for the development of national coexistence measures to avoid the adventitious presence of GMOs in conventional and organic crops, was adopted on 13 July 2010 (MADR, 2010). According to this recommendation, each Member State, individually, to achieve as low adventitious presence of genetically modified organisms (below 0.9%) in conventional crops and other crops, must take into account their specific needs at regional and local level on the cultivation of GMOs.

In Romania, it was adopted Order no. 61/2012 which provides as the main coexistence measure for GM maize with conventional maize, ensuring minimum separation distances of 200 m from neighboring pollen sources (MADR, 2012).

Many field experiments have been carried out in recent years to collect technical data under real coexistence conditions (Marceau et al., 2013, Popescu et al., 2010, Popescu et al., 2011), however, this data is not always taken into account in current legislation.

The adventitious presence of GMOs in conventional maize crops is mainly due to cross-pollination (pollen-mediated gene flow).
Urechean and Bonea (2017), previously reported that the best ways to reduce the GM's adventitious presence in conventional maize crops (below 0.9%) are: delayed sowing (so that there is no coincidence in flowering); a minimum isolation distance of 20 m for consumption maize and 100 m for the lots obtaining hybrid seed maize, and the use of buffer zones.
In this work, our purpose is to evaluate the role of buffer zones in reducing the cross-pollination rate and in the adventitious presence of GMOs in conventional maize crops under real culture and coexistence conditions.

MATERIALS AND METHODS

Two field experiments with genetically modified maize and conventional maize were placed at Agricultural Research and Development Station (A.R.D.S.) Simnic in the climatic conditions of the agricultural years 2011-2012 and 2012-2013
Experience I with an area of 4560 sq meters, where the sweet maize - Deliciul verii (pollen receiver) was sowed between two plots of genetically modified maize - MON 810 (pollen donor) at a distance of 100 m (West and East - 2012 and North and South - in 2013).
Experience II with an area of 4740 sq meters, where the sweet maize - Deliciul verii was sown between two plots of genetically modified maize - MON 810 at a distance of 100 m from it (West and East - 2012 and North and South - in 2013), with buffer zones (10 rows of Sudan Grass) halfway through the isolation distance (Photos 1, 2, 3, 4).
From a climatic point of view, the agricultural year 2011-2012 was an extremely dry year with an excessive pedological drought accompanied by extreme drought and extreme heatduring the blooming period. The agricultural year 2012 - 2013 was, in general, a favourable year for maize crops
For the study of the cross-pollination rate between two types of maize with different colour of the grains, the percentages of xenia were calculated (Watanabe et al., 2006) (Photo 5).

Photo 1. Aspects from experimental fields: MON 810, 2012

Photo 2. Aspects from experimental fields: Deliciul verii, 2012

Photo 3. Aspects from experimental fields: MON 810, 2013

Photo 4. Aspects from experimental fields: Deliciul verii, 2013

Photo 5. Xenia (MON 810 x Deliciul verii)

From every experience, from conventional maize parcels (Deliciul verii) there have been taken 5 consecutive ears from the middle part of the every row and the average of xenia has been determined for them. All the results were expressed as the average per ten row ±Standard error (SE).
The coefficient of variance calculated after Saulescu N.A and Saulescu N.N. (1967).

RESULTS AND DISCUSSIONS

In 2012, in Experience I, when the two plots (West 100m and East 100m) were sown without buffer zones, the average percentage of xenia was the same 0.42% respectively, but higher values are found on R3 and R5 of the plot in the West and on the R7-R10 interval of the East plot (the first rows from the MON 810 plot) (Table 1).
The variability of the percentage of xenia was identical, with very high values in both plots (CV = 35.1%).
Because there were no strong winds during the flowering period, we can say that only the local air currents could have influenced the percentage differences from row to row within the same plot.

In Experience II, when the two plots (West 100 m and East 100 m) were sown with a buffer zone of 10 rows of Sudan Grass, the average percentage of xenia was significantly lower compared to Experience I (0.26% for W and 0.24% for E) (Table 1).

Table 1. Variation of the percentage of xenia in sweet maize (Deliciul verii) cultivated at 100 m distance away from the MON 810 (pollen donor) with or without buffer zones, in 2012

No. row	Experience I		Experience II	
	W 100 m	E 100 m	W 100 m	E 100 m
R1	0.4	0.4	0.4	0.2
R2	0.4	0.2	0.3	0.2
R3	0.8	0.2	0.3	0.2
R4	0.4	0.5	0.3	0.2
R5	0.5	0.3	0.3	0.2
R6	0.3	0.4	0.2	0.2
R7	0.4	0.6	0.2	0.3
R8	0.4	0.5	0.2	0.3
R9	0.3	0.5	0.2	0.3
R10	0.3	0.6	0.2	0.3
Average	0.42	0.42	0.26	0.24
Standard error (±SE)	0.05	0.05	0.02	0.02
Coefficient of variation (CV%)	35.1	35.1	26.9	21.5
Average experience	0.42		0.25	
% reduction	40.5			

W= West; E= East

Higher values of the % of xenia were recorded on the R1-R5 interval for the West plot and on the R7-R10 interval for the East plot, as expected with them being the rows closest the pollen donor (MON 810).
A large variability in the percentage of xenia was observed in both plots (CV = 26.9% and CV = 21.5%), but less than in Experience I.
Comparing the average of the two plots (West and East) in Experience I and Experience II (Figure 1), it is very clear that the presence of buffer zones had an essential role, almost halving the average of the percentage of xenia (- 40.5%).

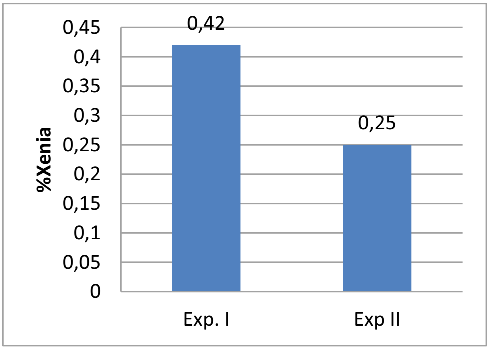

Figure 1. Reducing the percentage of xenia due to presence of buffer zones, 2012

Also, Messéan et al. (2009) reported that the use of buffer zones in combination with isolation distance results in a lower cross-pollination rate.

In 2013, in Experience I, in the two plots that were sown without buffer zones, the average percentage of xenia was very close, respectively 0.41% in South and 0.44% in North (Table 2).

Table 2. Variation of the % of xenia in sweet maize (pollen receptor) cultivated at 100 m distance away from the MON 810 (pollen donor) with or without buffer zones in 2013

No. row	Experience I		Experience II	
	S 100 m	N 100 m	S 100 m	N 100 m
R1	0.6	0.3	0.6	0.2
R2	0.5	0.4	0.5	0.4
R3	0.4	0.3	0.6	0.2
R4	0.4	0.2	0.2	0.5
R5	0.4	0.4	0.3	0.4
R6	0.4	0.5	0.2	0.3
R7	0.4	0.5	0.2	0.1
R8	0.4	0.6	0.2	0.2
R9	0.4	0.6	0.3	0.5
R10	0.2	0.6	0.3	0.6
Average	0.41	0.44	0.34	0.34
Standard error (±SE)	0.03	0.04	0.05	0.05
Coefficient of variation (CV%)	24.2	32.5	48.4	48.4
Average experience	0.43		0.34	
% reduction	20.0			

S = South; N = North

Higher values of the percentage of xenia were recorded on the first rows (R1 - R2) of the southern plot and on the last rows (R6 - R10) on the northern plot, thus on the nearest rows to the MON 810 plot.

The percentage of xenia being somewhat bigger in the North towards the middle of the plot, we can say that this year, only local air currents (which were from North to South) could have influenced a little the cross-pollination rate because the wind speed was small.

The coefficient of variation (CV%) is high for both plots, but something more homogeneous for the plot in the South.

In Experiment II, where the distance between the donor and the pollen receiver was 100 m in the South and North with a buffer zones of 10 rows of Sudan Grass in the middle of the isolation distance (50 m), the average percentage of xenia was lower compared to Experience I (0.34% both in the South and in the North - Table 2).

Higher values were recorded for the southern plot over the R1-R3 interval and in the northern plot for the R9-R10 interval which was expected, these being the first rows closest to the pollen donor (MON 810).

The average values, the standard deviation and the coefficient of variation were identical for both plots.

Comparing the Southern and North plots of Experience I and Experience II (Figure 2), we can say that the presence of protective curtains (10 rows of Sudan Grass) played an essential role, reducing the degree of contamination by 20.0% (in 2013).

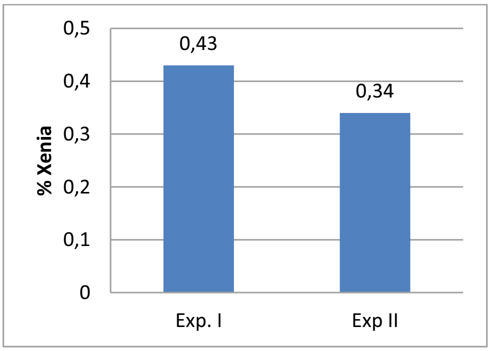

Figure 2. Reducing the percentage of xenia due to presence of buffer zones, 2013

Similar observations on the cross-pollination rate in maize for the distance of 100 m, respectively between 0.1 and 0.4% were reported by Messeguer et al. (2006), Pla et al. (2006) in Spain and by Baltazar et al. (2015) in Mexico.

Comparing the experimental years (2012, 2013), we can say that the predominant wind direction in the area of culture (during pollination period) as well as the intensity and frequency of the local air currents, significantly changes the average percentage of xenia.

Our results on the influence of the wind direction are consistent with those obtained by Weber et al. (2007), who observed that speed and direction of windmay vary between areas and years, so they can not be reliably embedded in strategies to avoid cross-pollination.

Other authors consider that the coincidence of flowering between donor and receiver plants as well as local wind conditions are other major factors of influence in plant coexistence (Hüsken et al., 2007; Warwick et al., 2009).

CONCLUSIONS

The rate of cross-pollination of conventional maize (Deliciul verii) with genetically modified maize (MON 810) was determined by: the presence or absence of buffer zones, the isolation distance and the predominant wind direction during the pollination period.

In 2012, at 100 m West and East from MON 810, the average of the percentage of xenia was 0.42% (Experience I) and 0.25% (Experience II) and in 2013 at 100m South and North distance from MON 810, the average of the percentage of xenia was 0.43% (Experience I) and 0.34% (Experience II)

The presence of buffer zones has led to a reduction in the cross-pollination rate by 40.5% in 2012 and by 20% in 2013.

As a result, the use of buffer zones can be considered as one of the important strategies for ensuring coexistence in the same area for GM maize and non-GM maize.

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