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STUDIES ON THE EFFECT OF SOME HERBICIDES (SINGLE AND DIFFERENT MIXTURES) ON WEEDS CONTROL AND SOIL QUALITY IN MAIZE

Victor Petcu[1], Georgeta Oprea[2], Costică Ciontu[1], Gheorghe Ştefanic[2]

[1]University of Agronomic Sciences and Veterinary Medicine of Bucharest, Faculty of Agriculture, 59 Marasti Blvd, District 1, 011464, Bucharest, Romania. E-mail: petcuvictor86@yahoo.com

[2]National Agriculture Research and Development Institute Fundulea, Nicolae Titulescu Street, no 1, 915200 Fundulea, Calarasi County, Romania

ABSTRACT

Maize or corn is the third most important crop worldwide; the area occupied by this crop is over 157 million acres. In Romania, corn is the main crop. One of the main problems linked to this crop facing Romania is the weeds control. Different types of pre and post-emergence herbicides are available in the market, but their effectiveness on different weed species in the field still needs to be determined under different agro-climatic conditions.

During the period 2012-2013 on the experimental field of the SC Profarma Holding SRL from Fundulea, under non irrigated conditions on cambic chernozem soil a field experiment with maize (*Zea mays*) was carried out.

The paper presents the results concerning the efficiency of some herbicides (aryloxiacid and different mixtures) on weeds control in maize crop and their impact on soil quality.

In the year 2013 the degree of weed infestation was higher due to heavy rainfall, but we got a good weed control with the combined herbicides in both years. Higher effect of new combined herbicides applied in vegetation against annual weeds, is due to their broad spectrum, including annual "resistant" dicotyledonous, such as *Xanthium strumarium, Solanum nigrum, Sinapis arvensis*.

The treatments with pre-emergence herbicide with dimethenamide P + pendimethalin (trade name Wing P) and post-emergence herbicide with bentazone + dicamba (trade name Cambio), achieved a rate of weed control of 97%, being qualified as having very good efficiency.

The treatments with acetochlor (trade name Guardian) and 2,4D + dicamba (trade name Ceredin) achieved a level of weed control of 88%, because the foxtail (*Setaria* spp.) could not be controlled effectively by a this single post-emergence herbicide.

Corn yields in control variants were 4430 kg ha^{-1} and 4830 kg ha^{-1} in the two years of experimentation. All of the herbicides tested, ensured higher average yields compared to the control. Yields were very significantly correlated with weed control efficiency of herbicides (r = 0.91*; r = 0.93***).**

Cellulolytic activity of soil was influenced by climatic conditions, type of herbicide used and time of applications. Water stress negatively influenced soil cellulolytic activity. In optimal conditions of soil humidity, cellulolytic activity increased, except after applying dimethenamide P + terbuthylazine herbicide (Akris), suggesting an improvement of biological conditions in soil (except for the above mentioned herbicide).

Soil respiration was influenced by herbicides applied. Among the herbicides studied Izoxaflutole + terbuthylazine (Merlin Duo) negatively influenced soil respiration. Herbicides dimethenamide P (Frontier Forte), dimethenamide P + terbuthylazine (Akris) and dimethenamide P + pendimethalin (Wing) had a beneficial effect, suggesting (1) a possible use of herbicides and/or their breakdown products by certain microorganisms in soil as a carbon source, leading to an increase in soil respiration, or (2) the effects of simulation (hormesis) of vital processes from the soil.

Key words: aryloxiacid herbicides, combined herbicides, maize, weed infestation, efficiency, soil cellulolytic activity, soil respiration.

INTRODUCTION

Over the years, worldwide, the chemical weed control in field crops has been in significant progress, which is closely correlated with the synthesis and the emergence of new types of herbicides (Popescu, 2007). The new rules of UE require practicing modern agriculture able to conserve and protect natural resources, cover food needs of a growing

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population and be financially profitable for producers and consumers.

Maize or corn (*Zea mays*) is the third most important crop worldwide; the area occupied by this crop is over 157 million acres. In Romania corn is the main crop, being cultivated on about 3 million hectares, which ranks Romania as the country with the largest area in the EU (27.4% of total). Romania ranks second in total production (13.8%), being surpassed by France, which has a productivity of almost three times higher, 9,100 kg ha^{-1} respectively. One of the main problems linked to maize crop facing Romania is the weed control.

Maize is very sensitive to weed competition during the first two month after emergence, as growth is slow in the first 4-6 weeks, and the small number of plants per square meter (5-7 plants) creates from the outset a fierce competition by weeds. Depending on the weeding level, loss of yield can reach 50 to 90 percent.

Knowledge about the efficacy of new herbicides in weed control, the effect on soil quality and their persistence in soil are extremely important for the proper use of such products in the environment. The more so as the European Union has opted for sustainable agriculture, reduced pesticide use and monitoring of acute and genetic toxicity for the ecosystem (Petsikos-Panagiotarou, 2000)

The paper presents the results concerning the efficiency of some herbicides (aryloxiacid and different mixtures) on weeds control in maize crop and their impact on soil quality under south eastern Romanian conditions.

MATERIAL AND METHODS

The field trials were performed at SC Profarma Holding SRL from Fundulea (situated in south-east part of Romania), under non-irrigated conditions in the years 2012 and 2013. The experiment was designed by the block method, in three replications, size of the experiment plot 25 m^2, on cambic chernozem soil, after preceding crop winter wheat in 2012 and sunflower in 2013. Active ingredient and doses of investigated herbicides are shown in Table 1.

The post-emergence treatments were carried out at the stage BBCH 13-14 of maize (three to four leaves formed).

Determinations concerning the effect of herbicides applied on floristic composition and effectiveness in weeds control in relation with the maize yield were performed.

The efficacy of herbicides against weeds was assessed as percent of damages (0% = no damage, 100% = total damages).

Soil samples were performed from the plough horizon (0-10 cm) and were tested for respiration, cellulose biodegradation potential (Ştefanic methods, 1994, 2006). Cellulolytic activity was expressed in % cellulose degraded and respiration in mg CO_2 from 100 g d.w. soil.

Table 1. Experimental variants

Pre-emergence treatment	Active ingredient	Doses (l/ha)	Post-emergence treatments	Active ingredient	Doses (l/ha)
V1 Control	Untreated		V1 Control	Untreated	
V2 Frontier Forte	Dimethenamide P, 720 g/l	1.2	V2 Cambio	Bentazone, 320 g/l + 90 g/l dicamba	1.2
V3 Guardian	Acetochlor, 820 - 860 g/l + Antidote	2.5	V3 Ceredin	2,4 D, 300 g/l + 100 g/l dicamba	1
V4 Wing P	Dimethenamide P, 212.5 g/l + pendimethalin, 250 g/l	4	V4 Cambio	Bentazone, 320 g/l + 90 g/l dicamba	2
V5 Gardorpim	Metolachlor, 312.5g/l + terbuthylazine, 181 g/l	4	V5 Cambio	Bentazone, 320 g/l + 90 g/l dicamba	2
V6 Akris	Dimetenamide P, 280 g/l + terbuthylazine, 250 g/l	4	V6 Buctril Universal	Bromoxinil, 280 g/l + acid 2,4 D, 280 g/l	1
V7 Merlin Duo	Izoxaflutole, 37.5 g/l + terbuthylazine, 375 g/l	2.5	V7 Adengo	Izoxaflutole, 225 g/l+ tiencarbazon-methyl 90 g/l + ciprosulphamid 150 g/l	0.35

RESULTS

Climatic conditions

The years of experimentation were totally different from the viewpoint of quantity and monthly repartition of rainfall. The averages of monthly temperatures in the growing season of crops were above the annual average, on average 1.9°C in 2012 and 1.2°C in 2013. There was a deficit of rainfall in the early part of the growing season (42.2), followed in May by precipitation that exceeded the normal of the zone and again a deficit of rainfall in June (51.6 mm), insufficient to meet water needs of crops. In 2013 year, the cumulated rainfall exceeded with 124 mm the normal of the zone (Table 2).

*Table 2.*Average temperature (°C) and monthly distribution of rainfall (mm) during the crop vegetation period Fundulea, 2012-2013

	March	April	May	June	July	August	September	Average/Sum
Temperature 2012	5.4	14.2	18.0	23.3	27.2	25.1	19.5	18.96
Temperature 2013	4.9	13.2	18.9	21.7	23.1	23.8	16.8	17.49
Multi-annual average	*4.7*	*11.1*	*16.9*	*20.6*	*22.5*	*22.0*	*17.2*	*16.43*
Differences 2012	0.7	3.1	1.1	2.7	4.7	3.1	2.3	2.53
Differences 2013	0.2	2.1	2.0	1.1	0.6	1.8	-0.4	1.06
Rainfall 2012	4.8	35.1	159.5	20.7	2.0	47.8	49.1	319.00
Rainfall 2013	39.0	38.5	97.1	126.7	96.1	22.2	91.4	511.00
Multi-annual average	*37.5*	*44.6*	*59.0*	*72.3*	*72.2*	*51.0*	*50.1*	*386.70*
Differences 2012	-32.7	-9.5	100.5	-51.6	-70.2	-3.2	-1.0	-67.70
Differences 2013	1.5	-6.1	38.1	54.4	23.9	-28.8	41.3	124.30

Effect of herbicides applied pre and post-emergence in maize on weed density and effectiveness of weed control

In the years of experimentation 2012 and 2013, the weeds from control variants were quite diverse, comprising a total number of 17 species. Among the identified species, some (like *Sonchus arvensis* and *Raphanus raphanistrum*) were isolated, while others had a high frequency and with presence in all plots, like as *Sorghum halepense* (28 pl./m^2), *Setaria glauca* (7.5 pl./m^2), *Setaria viridis* (12.5 pl./m^2), *Amaranthus retroflexus* (6 pl./m^2), *Chenopodium album* (5.5 pl./m^2) and *Cirsium arvensis* (19 pl./m^2). Average of weed infestation in maize crop was quite high, reaching 103.3 weeds/m^2 (Table 3).

Analysis of variance showed a very significant influence of year and treatments on weed density from maize crop. The interaction between year of experimentation and treatments had very significant influence on weed density in maize (Table 4). Higher values were recorded in 2013, indicating favorable conditions for infestation with weeds. Superior effect of new herbicides (mixtures) applied in vegetation for annual weed control, was due to the broad spectrum of annual weed control, including the "resistant" annual dicotyledonous weeds such as *Xanthium strumarium*, *Solanum nigrum*, *Sinapis arvensis*.

The pre-emergence treatment with mixture herbicides based on dimethenamide P + pendimethalin (trade name Wing P) and post-emergence with mixture herbicide based on bentazone + dicamba (trade name Cambio), (variant 4), achieved a very good efficacy (97%), (Table 5).

The treatment with dimethenamide P + terbuthylazine (trade name Akris) and bromoxinil + 2,4 D (commercial name Buctril Universal) (variant 6) was assessed as having very good efficacy (90%), being very close to the treatment with herbicide Wing. The latter (dimethenamide P and pendimethalin) acts very effectively on annual weeds, both mono- and dicotyledonous weeds and to some extent also on perennial weeds, due to an active substance from composition (pendimethalin) which is absorbed by the roots, which made the weeds to die soon after germination and emergence, being eliminated early from the competition.

Table 3. Frequency of weed species in maize control plots. Fundulea, 2012-2013

No.	Species	Frequency ($plants/m^2$)		Average (no. $pl./m^2$)
		2012	2013	
1	*Amaranthus retroflexus*	5	7	6
2	*Chenopodium album*	6	5	5.5
3	*Cirsium arvense*	18	20	19
4	*Echinochloa crus-galli*	5	6	5.5
5	*Galinsoga parviflora*		3	3
6	*Hibiscus trionum*	3	5	4
7	*Portulaca oleracea*	3	2	2.5
8	*Raphanus raphanistrum*		2	2
9	*Setaria glauca*	7	8	7.5
10	*Setaria viridis*	10	15	12.5
11	*Sinapis arvense*	2	4	3
12	*Solanum nigrum*	1	1.5	1.25
13	*Sonchus arvensis*		1	1
14	*Sorghum halepense*	26	30	28
16	*Xanthium strumarium*	2	4	3
17	*Veronica* spp.	0.1	5	2.6
	Total	88.1	118.5	103.3

Table 4. ANOVA for weed density in maize crop

Source of variances	The sum of squares	DF	MS	Factor F and significance
Factor A (Year)	468.745	1	468.75	8835.12***
Erorr A	0.106	2	0.53	
Factor B (Treatments – herbicide)	47551.77	7	6793.11	4518.51***
Interaction A x B	1115.747	7	159.39	106.02***
Erorr B	42.095	28	1.50	

Acetochlor and 2,4 D + dicamba (trade name Guardian and Ceredin) (variant 3) produced a level of weed control of 88%, because foxtail (*Setaria*) could not be effectively controlled by this single post-emergence treatment (Table 5).

In general, the efficacy of the herbicide mixtures was mainly determined by the optimal time for application, the development phase of the weeds, the amount of precipitation after herbicide application (specially in the case of the pre-emergence herbicides), as and the percentages of the various species of weeds.

Table 5. Effect of herbicides applied pre and post-emergence in maize on weeds density and effectiveness of weed control

Variants: Pre- and post-emergence treatments	2012		2013	
	Density of weeds ($plants/m^2$, at 32 DAT*)	Effectiveness of weed control (%)	Density of weeds ($plants/m^2$, at 32 DAT)	Effectiveness of weed control (%)
V1. Control (not treated)	88	0	119	0
V2. Pre: Frontier Forte Post: Cambio	4	94	6	93
V3. Pre: Guardian Post: Ceredin	11	88	15	87
V4. Pre: Wing P Post: Cambio	3	97	3	97
V5. Pre: Guardoprim Post: Akris + Cambio	5	95	6	95
V6. Pre: Akris Post: Universal	14	92	20	90
V7. Pre: Merlin Duo Post: Adengo	6	93	6	95

*Days after treatment.

It is obvious that the effectiveness of weed control was very good in both two years of experimentation and overall explanation is that although 2012 was a dry year, during the period of herbicides application, rainfall recorded was high enough (110.5 mm in May

2012 and 38.1 mm respectively in the same month in 2013), which ensured a good herbicide translocation in plants. On the other hand, new herbicides, such as Wing P, are not like old pre-emergence herbicides, which needed a certain level of soil moisture for maximum control.

According to some authors, of the three factors which cause losses in agriculture - diseases, pests, weeds, the latter causes the greatest damage. Worldwide, the damages caused by pests and diseases were estimated between 9-12% and about 14-15% by weeds (Berca, 2004).

Maize yields in untreated control were 4430 kg ha^{-1} and 4830 kg ha^{-1} in the two years of experimentation. All of the herbicides tested, ensured higher average yields compared to the control. Yields were very significantly positively correlated with effectiveness of herbicides studied (r=0.91***; r=0.93***), (Table 6).

Table 6. The effect of herbicides applied pre and post-emergence in maize on the yield

Variants	Yield (kg ha^{-1})	
	2012	2013
Control not treated	4430	4830
Frontier Forte + Cambio	6840	7200
Guardian + Ceredin	5780	6300
Wing P + Cambio	7170	7770
Gardoprim + Cambio	6920	7120
Akris + Universal	6780	7080
Merlin Duo + Adengo	6750	7100
LSD 5%	270	190
The correlation between yield and weed control efficiency (r)	r=0.91*** y =25.6x + 4362	r=0.93*** y=25.189x + 4761.1

Cellulolytic potential of soil

Many studies have shown that enzyme activities in the soil are sensitive enough to detect the effects of soil pollutants, including heavy metals (Avidano et al., 2005), insecticides (Yao et al., 2006) and herbicides (Sannino and Gianfreda, 2001). Among the various microorganisms from soil, most sensitive to herbicides seem to be cellulolytic bacteria (Ghinea et al., 1998). Studies with different herbicides (ex. glyphosate) showed that this caused a significant reduction (50%) of cellulase activity (Zabaloy and Gomez, 2008).

Our research showed a negative influence of applied herbicide on the enzymatic activity of soil according to the time of treatment and climatic conditions. The reduction was evident in case of the herbicides dimethenamide P + pendimethalin – [Wing P trade name (c-4.24)], dimethenamide P + terbuthylazine – [trade name Akris (c-3.97)] and izoxaflutole + terbuthylazine – [Merlin Duo trade name (c-3.98)], applied pre-emergence in maize (Table 7). The herbicide dimethenamide P (Frontier Forte, applied pre-emergence) and all herbicides applied post-emergence, did not significantly influence the soil cellulolytic activity (Table 7).

In 2013 year, the soil enzymatic activity (including untreated control) was more intense, leading to values between 7.6 (variant 5) and 12-13.5% degraded cellulose/100 g soil for variants V1 (control), respectively V2 (treated with Frontier Forte + Cambio).

For variant 6 (Akris + Buctril Universal) and the variant treated with pendimethalin (variant V4) soil cellulolytic activity was lower compared to the other variants (Figure 1).

These results are in concordance with other studies about the effect of herbicides on soil processes, which have shown that some herbicides reduced, while others stimulated various vital processes from the soil. It was shown that pendimethalin prevents the first step of nitrification - oxidation of ammonium ions to nitrite (Goring and Laskowski, 1982, quoted by Nadasy et al., 2007). Herbicides have a direct effect on nutrients absorbed by the plants by the fact that they have an influence on life processes of plants. This is important because nitrogen absorption is intense in the early stages of the growing season (Nadasy et al., 2007).

Table 7. Influence of pre and post-emergence herbicides for maize crop, on the soil cellulolytic activity (% degraded cellulose) Fundulea, 2012

	Pre-emergence treatment	Bio cellulose (A month after treatment)	Post-emergence treatment	Bio cellulose (A month after treatment)
V1	Untreated control	5.46 a	Untreated control	4.81 a
V2	Dimethenamide P (Frontier Forte)	5.65 a	Bentazone + Dicamba (Cambio)	5.39 a
V3	Acetochlor (Guardian)	4.61 b	2,4 D + dicamba (Ceredin)	4.91 a
V4	Dimethenamide P + pendimethalin (Wing P)	4.24 c	Bentazone + dicamba (Cambio)	4.27 a
V5	Metolachlor + terbuthylazine (Gardoprim)	5.09 b	Bentazone + dicamba (Cambio)	3.04 a
V6	Dimethenamide P + terbuthylazine (Akris)	3.97 c	Bromoxinil + 2,4 D (Buctril Universal)	4.27 a
V7	Izoxaflutole + terbuthylazine (Merlin Duo)	3.98 c	Izoxaflutole + tiencarbazon-methyl + ciprosulphamid (Adengo)	4.64 a
	LSD 0.1%	0.54	LSD 0.1%	2.34

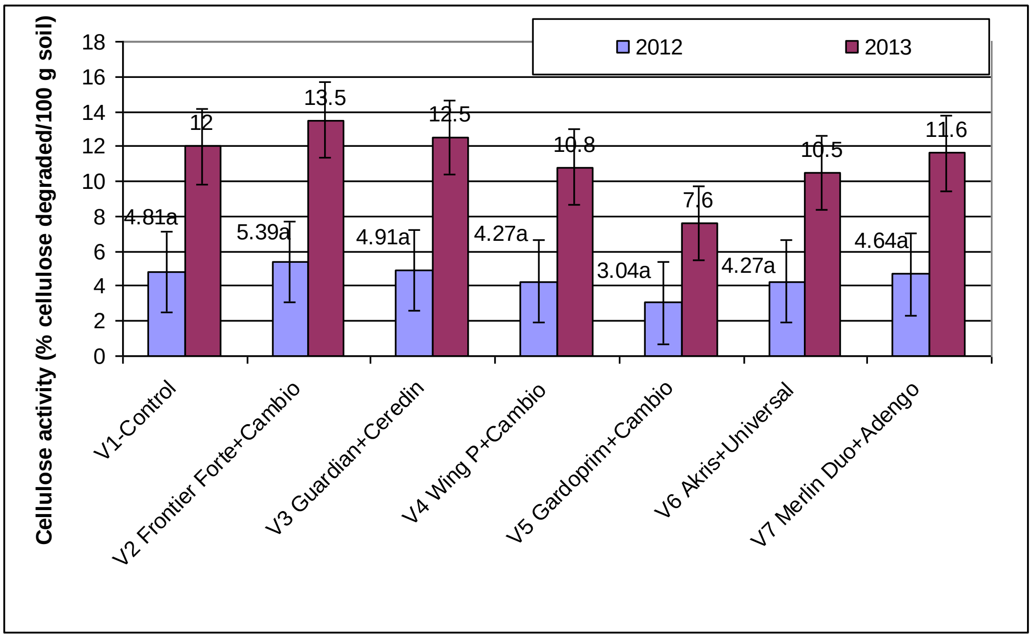

Figure 1. Effect of herbicides applied to maize on soil cellulolytic activity Fundulea, 2012 and 2013

Soil respiration

Soil respiration is a complex process that occurs in all terrestrial ecosystems. Although the results on the effects of herbicides on soil respiration are contradictory, this is considered a sensitive indicator of toxicity of pesticides and heavy metals (Ştefanic and Oprea, 2011).

Our results showed that application of herbicides can stimulate, inhibit or may not affect soil respiration, depending on the time, type of herbicide application and climatic conditions. The herbicides acetochlor and dimethenamide P (pre-emergence application) had no significant effect on soil respiration, values (58.59 and 61.67 mg CO_2/100 g soil) being in the same group (b) with untreated control (60.92 mg CO_2/100 g soil). Herbicides metolachlor + terbuthylazine (trade name Gardoprim) and izoxaflutole + terbuthylazine (trade name Merlin Duo) influenced negatively soil respiration, while herbicides

Akris (dimethenamide P + terbuthylazine) and Wing P (dimethenamide P + pendimethalin) had a beneficial effect in both years of experimentation (Table 8). In case of the post-emergence treatments, stimulation of respiration was identified for bentazone + dicamba herbicide (trade name Cambio) applied after dimethenamide P. Similar values to control were found with treatments performed with Ceredin and Adengo in 2012 and it was obvious that the soil respiration was higher under 2013 year conditions for all treatments, due to high soil humidity which can stimulate CO_2 production in soil (Table 8).

A possible use of herbicides and their degradation products by micro-organisms in the soil as a carbon source, can explain the increase of soil respiration in some treatments, while reducing/inhibition of soil respiration may be an adverse effect of the herbicide used.

Table 8. Influence of herbicides studied, applied to maize, on soil respiration
Fundulea, 2012 and 2013

Pre-emergence treatment	Soil respiration (mg CO_2/100 g soil dry matter)		Post-emergence treatment	Soil respiration (mg CO_2/100 g soil dry matter)	
	2012	2013		2012	2013
Control	60.92 b	63.49 b	Control	62.62 b	73.71 a
Dimethenamide P (Frontier Forte)	61.67 b	63.66 b	Bentazone + dicamba (Cambio)	90.73 a	88.06 b
Acetochlor (Guardian)	58.59 b	61.60 b	2,4 D + dicamba (Ceredin)	42.19 b	73.55 a
Dimethenamide + pendimethalin (Wing P)	75.80 a	70.02 a	Bentazone + dicamba (Cambio)	56.09 b	80.18 a
Metolaclor + terbuthylazine (Gardoprim)	44.87 c	54.98 c	Bentazone + dicamba (Cambio)	38.53 c	67.78 a
Dimethenamide + terbuthylazine Akris	67.96 a	65.96 b	Bromoxinil + 2,4 D (Buctril Universal)	41.00 c	72.63 a
Izoxaflutole + terbuthylazine (Merlin Duo)	42.69 c	53.14 c	Izoxaflutole + tiencarbazon-methyl + ciprosulphamid (Adengo)	55.11 b	79.88 a
LSD 0.1%	11.39	5.16	LSD 0.1%	20.93	12.63

Zabaloy and Gómez (2008) showed that metsulfuron methyl had no effect on microbial respiration of an acidic soil of San Román (pH 6.06), even at the highest rate. However, metsulfuron methyl inhibited microbial respiration in soils of Bordenave (pH 7.44), at a rate of 0.1 mg kg^{-1} soil. Low application rates of glyphosate and 2,4 D produced only transitory effects on CO_2 evolution, whereas the addition of high doses of these herbicides stimulated microbial activity. On the other hand, the addition of fertilizer to soil treated with a high dose of glyphosate temporarily inhibited CO_2 release.

CONCLUSIONS

The degree of weed infestation in maize was high (average 103 weeds/m^2 in the two years of experimentation); the dominant species were *Setaria* spp*.*, *Sorghum halepense* (from rhizomes and seeds) and *Cirsium arvense*.

Among the treatments performed, the mixtures herbicides based on dimethenamide P + pendimethalin and bentazone + dicamba had higher efficiency (97%). This very good efficacy was due to a substance (pendimethalin) which was not present in other combinations. It has a residual activity on weeds, which made the weeds to die soon after germination and emergence, being eliminated early from the competition.

Cellulase activity and soil respiration depended upon the type of herbicide, time of treatment application and climatic conditions. The drought conditions (2012) negatively influenced soil cellulolytic activity. Under optimal conditions of soil humidity, cellulolytic activity was increased, except for herbicide dimethenamide P + terbuthylazine (Akris), suggesting an improvement in soil biological conditions. Other herbicide mixtures [dimethenamide P + pendimethalin (trade name Wing P), dimethenamide P + terbuthylazine (Akris) and izoxaflutole + terbuthylazine (Merlin Duo)] applied pre-emergence in maize had a negative influence on soil cellulolytic activity compared with dimethenamide P (Frontier Forte) and acetochlor (Guardian), which did not influence this indicator.

The soil respiration was higher under 2013 year conditions for all treatments, due to high soil humidity which can stimulate CO_2 production in soil.

Herbicides mixtures izoxaflutole + terbuthylazine (Merlin Duo) and metolachlor + terbuthylazine (Gardoprim) negatively influenced soil respiration, significantly in 2012. Herbicides dimethenamide P (Frontier Forte), dimethenamide P + terbuthylazine (Akris) and dimethenamide P + pendimethalin (Wing P) had a beneficial effect. This benefit suggested a possible use of these herbicides and/or their degradation products by certain micro-organisms from soil as a carbon source, leading to an increase in soil respiration, or an "hormesis" effect for vital processes.

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