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# EFFECTIVENESS OF USING THE MIXTURES OF HERBICIDES FLUMIOXAZINE AND FLUROCHLORIDONE IN SUNFLOWER CROPS

M. P. Radchenko, Zh. Z. Guralchuk, O. P. Rodzevych,  
M. V. Khandezhina, Ye. Yu. Morderer

*Institute of Plant Physiology and Genetics of the National Academy Sciences of Ukraine,  
31/17, Vasylykivska Str., Kyiv, Ukraine, 03022*

*E-mail: mradchenko.phd@i.ua, azhanna@ukr.net\*, rodzevich2017@gmail.com,  
20111975marika@ukr.net, morderer@ifrg.kiev.ua*

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**Aim.** This work is devoted to the search for potential partners for the integrated application of flumioxazine in sunflower crops. This herbicide is an inhibitor of protoporphyrinogen oxidase (PPO) in weed plants. The resistance to this class of herbicides is not yet common today, compared to other classes. Therefore, the work is aimed at developing flumioxazine-based herbicide compositions that effectively prevent the emergence of herbicide-resistant weed biotypes. **Methods.** The interaction effects, weed control efficiency, and crop selectivity were studied when flumioxazine was used in the mixtures with herbicides acetochlor and propisochlor (long-chain fatty acid synthesis (LCFAS) inhibitors), promethrin (electron transport (ET) inhibitor in photosystem 2 (PS 2) of chloroplasts) and fluoro-chloridone (inhibitor of carotenoid synthesis by blocking the activity of phytoendesaturases (PDS)) both in the experiments on sunflower crops and in greenhouse experiments using the model objects. **Results.** The studies have shown that when flumioxazine is applied with the ET inhibitor, promethrin, the interaction is antagonistic, resulting in poor weed control efficiency and sunflower yield decrease. The tank mixtures of flumioxazine with LCFAS inhibitors acetochlor and propisochlor provide high weed control efficiency but are low in selectivity for sunflowers. The interaction between flumioxazine and fluoro-chloridone within the recommended application rates is additive. The tank mixture of flumioxazine and fluoro-chloridone herbicides at the application rates of 55 and 500 g/ha, respectively, is selective for sunflowers. In terms of control efficiency of annual dicotyledons, this mixture of herbicides exceeded, and in terms of control efficiency of annual cereal weed species was only slightly inferior to the control integrated herbicide (metolachlor + terbuthylazine). In these application rates, flumioxazine and fluoro-chloridone provided sunflower yield on par with this complex herbicide. **Conclusions.** Among the investigated herbicides, the optimal partner of flumioxazine for complex use in sunflower crops was the herbicide, inhibitor PDS, fluoro-chloridone. Given that flumioxazine and fluoro-chloridone differ in their phytotoxicity mechanisms but share a common spectrum of controlled weed species, the use of a mixture of these herbicides is a factor that minimizes the likelihood of the emergence of herbicide-resistant weed biotypes.

**Key words:** *Helianthus annuus L.*, herbicides, resistance, interaction, flumioxazine, fluoro-chloridone, promethrin, propisochlor, acetochlor.

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## INTRODUCTION

The areas under sunflowers are increasing with a rapid tempo, though in 2022, these have been reduced a little as compared to the previous year, but a high index of 4 million 702 thousand hectares has been preserved (Ministry of Agrarian Policy, 2022).

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Ye. Yu. MORDERER, 2022

There is also increasing potential soil clogging, and weediness of crops which may result in the loss of 20–30 % of yield if comprehensive measures are not taken (Shevchenko M, 2007), and the yield drops twofold in the fields with very thick weeds (Masliyov SV et al, 2021). The application of agrotechnical measures, including the control of harmful weeds in the fields of sunflower predecessor, pre- and post-germination harrowing, and interrow hoeing, does not always protect

the fields from weeds in a reliable way (Remeniuk, 2015; Masliyov et al, 2021). In modern conditions of cultivating sunflowers in short (four-, six-field) crop rotations, the protection of this crop from weeds is a relevant element of its cultivation technology (Rozhkovan, 2014). According to the traditional technology, the protection of sunflowers from undesired weeds is based on applying soil herbicides (Morderer et al, 2014; Rudik et al, 2020).

It is known that the long-term application of single-type herbicides leads to the emergence of resistant forms of weeds due to the selective pressure on their populations. If herbicides with the same phytotoxicity mechanism are applied permanently, the weeds-survivors have a considerable advantage and may become dominant in the population, replacing the initial type (Powles and Yu, 2010; Vencil et al, 2012). At present, among 266 species of weeds (113 of which are monocotyledonous and 153 – dicotyledonous weeds), 509 biotypes have been found to be resistant to 164 different herbicides (Heap I, 2022). The emergence of resistant populations decreases the effectiveness of controlling weeds and causes an increase in the rates of herbicide application which entails additional expenses and considerable harm to the environment.

To avoid the development of weed resistance to herbicides, it is important to have the comprehensive application of herbicides with different mechanisms of phytotoxicity (Diggle et al, 2003; Beckie, 2006; Norsworthy et al, 2012; Morderer et al, 2018). The integrated herbicide preparations and mixtures of herbicides, consisting of long-chain fatty acid synthesis (LCFAS) inhibitors and electron transport (ET) inhibitors in photosystem 2 (PS 2) of chloroplasts, including the preparations, the active substances of which are LCFAS inhibitor S-metolachlor and ET inhibitor terbuthylazine are widely used to protect sunflower crops (Morderer et al, 2014). The application of these mixtures and preparations decreases the probability of emerging resistance to some degree, but it is not perfect, since the spectra of action of the components overlap only in part. At the same time, the components of the herbicide composition should have a common spectrum of action to prevent the emerging resistance effectively (Diggle et al, 2003; Beckie, 2006; Norsworthy et al, 2012). Besides, the resistance to herbicides, ET inhibitors, is relatively common. For example, the biotypes, resistant to this group of herbicides, were found among 87 weed species (Heap, 2022). At the same type, the biotypes, resistant to the herbicides, inhibitors of protoporphyrinogen

oxidase (PPO), were found only among 13 weed species (Heap, 2022). Thus, the creation of antiresistant compositions of herbicides, based on PPO inhibitors, to protect sunflower crops is quite a logical step.

The greatest problem in protecting sunflower crops is control over dicotyledonous weed species, as cereal weeds can be effectively controlled during sunflower vegetation using graminicide herbicides. Thus, regardless of the recommendation for flumioxazine, a herbicide that is PPO inhibitor, to be used only to control dicotyledonous species of weeds (Rafalsky et al, 2018), the introduction of this herbicide to the antiresistant compositions of herbicides to protect sunflower crops is quite reasonable. The potential partners of flumioxazine in combined application on sunflower crops may be the herbicides, recommended for the use on these crops, including promethrin – the ET inhibitor, propisochlor and acetochlor – LCFAS inhibitors, and fluorchloridone – the inhibitor of carotenoid synthesis by blocking the activity of phytoendesaturases (PDS) (Rafalsky et al, 2018). It should be noted that the resistance to LCFAS and PDS inhibitors, found in 14 and 15 weed species, respectively, is a less common phenomenon than the resistance to ET inhibitors (Heap, 2022). Since all the mentioned potential partners of flumioxazine are applied in sunflower crops before the germination of this plant, this study envisages the investigation of the effectiveness of the comprehensive application of flumioxazine when introduced into the soil before the germination of sunflower and the weeds. It is noteworthy that flumioxazine is used in mixtures with other herbicides for weed control in agriculture of different countries (Hermann et al, 2017, Schutte, 2018), but there are no current data on its application in the sunflower crops.

Therefore, the aim of this work was to determine the effects of the interaction during the combined application of flumioxazine, the herbicide that is PPO inhibitor, in the mixtures with herbicides, inhibitors of ET, LCFAS, and PDS, to choose the optimal composition of herbicides from the selectivity standpoint regarding sunflower, to control weeds effectively, and to prevent the emergence of herbicide-resistant weeds.

## METHODS

The initial selection of components for the comprehensive application with flumioxazine was conducted in field experiments. The character of the interaction in the combination of herbicides, chosen for further investigation, was checked in greenhouse experiments using model objects. The final evaluation of the effec-

tiveness of controlling weeds and the selectivity of the chosen combination of herbicides for sunflowers was conducted in the field experiments.

The field experiments were conducted in 2016 on the fields of rural enterprises in the village of Sushchany (location 1), the village of Hermanivka (location 2) of Obukhiv district, Kyiv region, in the crops of sunflower hybrid P63LE10, with winter wheat being its predecessor. In 2017–2018, field experiments were conducted on the fields of the experimental farm of the Institute of Plant Physiology and Genetics, the NAS of Ukraine, in the village of Hlevakha, Fastiv district, Kyiv region (location 3), sunflower crops of Karamba hybrid, its predecessors – winter wheat (2017) and soybeans (2018). The sunflower was sown by the technology common for the farms. The herbicides were applied by spraying the soil after sowing but before the sunflower germination, using the integral boom sprayer Agritop with condensed air: (integral boom sprayer), boom width – 2.5 m, number of sprinklers – 5, width between sprinklers – 50 cm, boom travel height – 50 cm, velocity – 5 km/h, working liquid consumption – 300 l/ha. The experiment was laid out in a randomized complete block design in four replicates with the experimental plot size adopted to 12.5 sq.m.

The field experiments were launched by the scheme presented in Table 1; the integrated herbicidal preparation with active substances metolachlor, LCFAS inhibitor, and terbuthylazine, ET inhibitor, was used as a reference for comparison.

The effectiveness of weed control was determined for each specific species via evaluating the number of weeds on the plots, where herbicides were applied, comparing it to the control, and calculating according to the formula (1): (Ivashchenko and Merezhytsky, 2001).

$$E (\%) = 100 - B_2 * K_1 * 100 / (B_1 * K_2) \quad (1),$$

where E (%) – the effectiveness of controlling a specific species of weeds,  $K_1$  – number of weeds per 1 sq.m. at the first registration in the control (initial weed infestation),  $K_2$  – number of weeds per 1 sq.m. at the second (third) registration,  $B_1$  – number of weeds per 1 sq.m. at the first registration on the experimental plot with the herbicides (initial weed infestation),  $B_2$  – number of weeds per 1 sq.m. at the second (or third) registration on the experimental plot with the herbicides.

The weeds were counted 30–40 and 60 days after the application of herbicides and before harvesting sun-

flower seeds. The selectivity of herbicides for sunflowers was estimated 15 days after the application of herbicides and during each counting of the weeds.

The selectivity was assessed by the degree of the phytotoxic impact of herbicides on the crop, which was determined by counting the number of plants with evident signs of the phytotoxic effect of herbicides and the visual assessment of the degree of this damage (a decrease in the linear dimensions of plants, morphological changes, chlorosis of leaves, etc.). The degree of the damage was expressed in percentage terms (0 % – no manifested effect of the herbicide, 100 % – the complete collapse of plants). The phytotoxic effect was calculated by the formula (2) (Ivashchenko and Merezhytsky, 2001):

$$Ph (\%) = \sum B_i * K_i * 100 / K \quad (2),$$

where Ph – phytotoxic effect of herbicides on the crop in percentage terms,  $B_i$  – visual assessment of a specific type of damage in percentage terms,  $K_i$  – number of plants with this type of damage, K – total number of the examined plants.

The yield of sunflower seeds was registered by collecting and shelling sunflower heads from the experimental plots. The seeds were weighed, and the humidity was determined. The yield was estimated in t/ha with the standard humidity of 8 %.

The greenhouse experiment involved the model object, which was the plants of oil radish (*Raphanus sativus d. var. oleifera* Metrg.) (a model of annual dicotyledonous weeds). The plants were grown in plastic pots with 1 kg of soil in the greenhouse of the Institute of Plant Physiology and Genetics, the NAS of Ukraine (north latitude – 50° 39'; east longitude – 30° 49'). The herbicides were applied to the soil layer of 1 cm, used to cover the seeds for sowing. The experiment was

**Table 1.** The scheme of the field experiments (Locations 1 and 2) for the selection of potential partners of herbicide flumioxazine

No.	Rates of application in terms of the active substance
1	Control
2	Flumioxazine (66 g/ha)
3	Flumioxazine (55 g/ha) + promethrin (750 g/ha)
4	Flumioxazine (55 g/ha) + propisochlor (1440 g/ha)
5	Flumioxazine (55 g/ha) + acetochlor (1350 g/ha)
6	Flumioxazine (55 g/ha) + fluorochloridone (500 g/ha)
7	Integrated herbicide (metolachlor (1406 g/ha) + terbuthylazine (844 g/ha))

done in four repeats. The scheme of the greenhouse experiment is presented in Table 2.

Fifteen days after sowing and applying herbicides, the inhibitory effect was determined by the suppression of the growth in the wet weight of the aboveground part of the plant. The inhibitory effect was calculated by the formula (3)

$$I_i = 100 - 100P_i/P_0, \% \quad (3),$$

where  $I_i$  – the phytotoxic effect in percentage terms in this experimental variant,  $P_i$  – the wet weight of the aboveground part of the plant in a certain experimental variant,  $P_0$  – the wet weight of the aboveground part of the plant in the control variant of the experiment.

The interaction effects of the application of the herbicidal mixture were determined by Colby's method (Colby SR, 1967) by comparing the actual inhibitory effect or effectiveness of controlling a certain species

**Table 2.** The scheme of the greenhouse experiment using oil radish to determine the interaction effects in the combined application of flumioxazine and fluorochloridone

No.	Rates of application in terms of the active substance
1	Control
2	Fluorochloridone (375 g/ha)
3	Fluorochloridone (625 g/ha)
4	Flumioxazine (38 g/ha)
5	Flumioxazine (50 g/ha)
6	Fluorochloridone (375 g/ha) + flumioxazine (38 g/ha)
7	Fluorochloridone (625 g/ha) + flumioxazine (38 g/ha)
8	Fluorochloridone (375 g/ha) + flumioxazine (50 g/ha)
9	Fluorochloridone (625 g/ha) + flumioxazine (50 g/ha)

**Table 3.** The scheme of the field experiment (Location 3) to determine the interaction effects in the combined application of flumioxazin and fluorochloridone

No.	Rates of application in terms of the active substance
1	Control
2	Fluorochloridone (375 g/ha)
3	Fluorochloridone (625 g/ha)
4	Flumioxazine (38 g/ha)
5	Flumioxazine (50 g/ha)
6	Fluorochloridone (375 g/ha) + flumioxazine (38 g/ha)
7	Fluorochloridone (625 g/ha) + flumioxazine (38 g/ha)
8	Fluorochloridone (375 g/ha) + flumioxazine (50 g/ha)
9	Fluorochloridone (625 g/ha) + flumioxazine (50 g/ha)
10	Integrated herbicide (metolachlor (1406 g/ha) + terbuthylazine (844 g/ha))

of weeds, which was observed while applying this mixture, to the expected effectiveness, calculated by the formula (4):

$$I_{ij} = I_i + I_j(100 - I_i)/100 \quad (4),$$

where  $I_{ij}$  – expected inhibitory effectiveness of controlling weeds by the mixture of herbicides,  $I_i$  and  $I_j$  – inhibitory effectiveness of controlling weeds by the application of the first and the second components of the mixture.

A field experiment to study the interaction of flumioxazine with fluorochloridone was carried out according to the scheme presented in the Table 3.

The following herbicidal preparations were used in the experiments: Pledge, wettable powder (flumioxazine, 551 g/kg); Gesagard 500 FW, suspension concentrate (promethrin, 500 g/l) and Primextra TZ Gold, suspension concentrate (S-metolachlor, 312, 5 g/l + terbuthylazine, 187.5 g/l); Proponit, emulsion concentrate (propisochlor, 720 g/l); Trophy, emulsion concentrate (acetochlor, 900 g/l); Racer, emulsion concentrate (fluorochloridone, 250 g/l).

The statistical processing of the obtained results was done by the dispersion analysis (ANOVA) using the Tukey test (HSR). The results were presented as mean values and standard deviations ( $m \pm SE$ ). The difference between the data was considered significant if  $p \leq 0.05$ .

## RESULTS

To conduct the initial assessment in the field experiments in Locations 1 and 2 the herbicides were applied on the day following the sowing (Table 1). In Location 1, the application was done on April 11, 2016; it was sunny at the moment of the application, the air temperature was +19 °C, the wind velocity – 3 m/s, the soil surface was dry. Two days after the application of herbicides, there was considerable precipitation, which, combined with insolation and wind, resulted in the formation of the crust on the soil surface. This led to the delay and partial inhibition of the germination of sunflower and weeds and, as the following registrations demonstrated, it impacted the effectiveness of the herbicidal action. In Location 2, the herbicides were applied on April 17, 2016; it was sunny at the moment of the application, the air temperature was 20 °C, the wind velocity – 3 m/s, the soil was wet.

The results of crop examination to assess the effect of herbicides on sunflower plants are presented in Table 4. In the experiment in Location 1, there was more scarce sunflower germination in all the variants, including the



control. Besides, there was leaf roll in some plants, including the control. The highest phytotoxic effect on the crop, conditioned by this type of damage, was observed in the variants where flumioxazine was used in the mixtures with propisochlor and acetochlor. In other variants with the application of herbicides, the insignificant increase in the manifestation of this type of damage as compared with the control was observed only 60 days after the introduction of herbicides. In the experiment in Location 2, an evident manifestation of the impact of flumioxazine was seen in the goffered first and second actual leaves. In the variant with the application of the integrated control herbicide, the

manifestation of the phytotoxic effect on sunflowers was an insignificant decrease in linear dimensions of some plants. It should be noted that plants with goffered leaves and delayed growth were registered in the control as well. The combined application of flumioxazine and herbicides propisochlor and acetochlor, as well as promethrin, enhanced the phytotoxic effect on the crop (variants 3, 4, 5). In the variants with the application of flumioxazine, integrated control herbicide, and the mixture of herbicides flumioxazine and fluorochloridone, there was no observed reliable excess in the manifestation of plant damage as compared to the control.

**Table 4.** The phytotoxic effect (%) of herbicides on sunflower plants 15, 30–40, 60, and 140 days after the application of herbicides (2016)

Location 1				Location 2			
15	40	60	140	15	30	60	140
0.7 ± 0.3 <sup>a</sup>	0.6 ± 0.2 <sup>a</sup>	0.4 ± 0.1 <sup>a</sup>	0.3 ± 0.1 <sup>a</sup>	1.7 ± 0.3 <sup>a</sup>	0.94 ± 0.18 <sup>a</sup>	0.30 ± 0.07 <sup>a</sup>	0.08 ± 0.02 <sup>a</sup>
1.7 ± 0.1 <sup>a</sup>	2.0 ± 0.1 <sup>a</sup>	3.6 ± 0.1 <sup>b</sup>	1.6 ± 0.2 <sup>a</sup>	4.5 ± 0.5 <sup>a</sup>	1.88 ± 0.26 <sup>a</sup>	0.94 ± 0.18 <sup>a</sup>	0.15 ± 0.06 <sup>a</sup>
1.6 ± 0.1 <sup>a</sup>	1.7 ± 0.1 <sup>a</sup>	3.3 ± 0.3 <sup>b</sup>	1.5 ± 0.1 <sup>a</sup>	7.2 ± 0.8 <sup>a</sup>	4.69 ± 0.94 <sup>b</sup>	3.28 ± 0.47 <sup>b</sup>	0.34 ± 0.09 <sup>a</sup>
4.9 ± 0.2 <sup>b</sup>	5.1 ± 0.1 <sup>b</sup>	9.2 ± 0.8 <sup>c</sup>	5.3 ± 1.3 <sup>b</sup>	18.0 ± 1.7 <sup>b</sup>	7.50 ± 1.11 <sup>bc</sup>	4.53 ± 1.0 <sup>b</sup>	0.59 ± 0.24 <sup>a</sup>
7.2 ± 0.7 <sup>b</sup>	7.4 ± 0.7 <sup>b</sup>	12.5 ± 0.5 <sup>c</sup>	6.8 ± 0.1 <sup>b</sup>	24.4 ± 2.4 <sup>c</sup>	9.1 ± 0.5 <sup>c</sup>	4.8 ± 0.4 <sup>b</sup>	0.69 ± 0.21 <sup>a</sup>
1.6 ± 0.1 <sup>a</sup>	1.8 ± 0.2 <sup>a</sup>	3.4 ± 0.1 <sup>a</sup>	1.6 ± 0.1 <sup>a</sup>	3.9 ± 0.6 <sup>a</sup>	3.0 ± 0.5 <sup>a</sup>	0.8 ± 0.2 <sup>a</sup>	0.12 ± 0.01 <sup>a</sup>
1.7 ± 0.3 <sup>a</sup>	1.9 ± 0.4 <sup>a</sup>	3.1 ± 0.1 <sup>a</sup>	1.6 ± 0.1 <sup>a</sup>	2.8 ± 0.3 <sup>a</sup>	1.1 ± 0.2 <sup>a</sup>	0.5 ± 0.1 <sup>a</sup>	0.18 ± 0.04 <sup>a</sup>

Note. The difference is reliable at  $p \leq 0.05$  if the letters are not the same.

**Table 5.** The infestation of the sunflower crop (control plots) with some species of weeds, the stages of their development, and linear dimensions 40 days (Location 1) and 30 days (Location 2) after the application of herbicides

Species	Code	Weed infestation (items/sq.m.)	Stage of development (BBCH)	Height (cm)
<i>Location 1 (40 days after the application)</i>				
Field mustard ( <i>Sinapis arvensis</i> L.)	SINAR	2	30	10–12
Field pennycress ( <i>Thlaspi arvense</i> L.)	THLAR	1–2	30–32	5–10
Herb-Sophia ( <i>Descurainia sophia</i> (L.) Webb.	DESCO	0.2–0.4	32	5–10
Shepherd's purse ( <i>Capsella bursa-pastoris</i> (L.) Medik.	CAPBP	0.1	30	5
Black nightshade ( <i>Solanum nigrum</i> L.)	SOLNI	2–4	15–16	2–5
Goosefoot ( <i>Chenopodium album</i> L.)	CHEAL	0.3	13–15	4–5
Black-bindweed ( <i>Polygonum convolvulus</i> L.)	POLCO	0.5	13–15	3–10
<i>Location 2 (30 days after the application)</i>				
Goosefoot ( <i>Chenopodium album</i> L.)	CHEAL	8–10	12–14	1–2
Field mustard ( <i>Sinapis arvensis</i> L.)	SINAR	1–2	12–14	2–3
Field pennycress ( <i>Thlaspi arvense</i> L.)	THLAR	1–2	12–14	1–2
Yellow foxtail ( <i>Setaria glauca</i> (L.) Pal. Beauv.)	SETPF	1–2	10–12	2–3

Note. The globally recognized BBCH system of 10 stages was used to determine the phases of plant development (Hack et al, 1992).

**Table 6.** The effectiveness of controlling some species of weeds (%) 40 and 60 days after the application of herbicides (location 1)

	40 days						60 days							
	SINAR	THLAR	DESSO	CAPBP	SOLNI	CHEAL	POLCO	SINAR	THLAR	DESSO	CAPBP	SOLNI	CHEAL	POLCO
	58 ± 5 <sup>b</sup>	60 ± 4 <sup>a</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>b</sup>	85 ± 3 <sup>b</sup>	97 ± 2 <sup>a</sup>	30 ± 6 <sup>a</sup>	48 ± 3 <sup>a</sup>	60 ± 4 <sup>b</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>a</sup>	48 ± 5 <sup>a</sup>	65 ± 9 <sup>a</sup>	8 ± 5 <sup>a</sup>
	38 ± 5 <sup>a</sup>	43 ± 8 <sup>a</sup>	99 ± 1 <sup>a</sup>	90 ± 2 <sup>a</sup>	64 ± 6 <sup>a</sup>	99 ± 1 <sup>a</sup>	40 ± 4 <sup>ab</sup>	43 ± 8 <sup>a</sup>	33 ± 6 <sup>a</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>a</sup>	43 ± 6 <sup>a</sup>	86 ± 2 <sup>b</sup>	8 ± 5 <sup>a</sup>
	84 ± 2 <sup>c</sup>	81 ± 8 <sup>b</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>b</sup>	93 ± 1 <sup>c</sup>	99 ± 1 <sup>a</sup>	55 ± 3 <sup>c</sup>	65 ± 7 <sup>b</sup>	75 ± 6 <sup>bc</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>a</sup>	86 ± 4 <sup>c</sup>	93 ± 1 <sup>b</sup>	25 ± 3 <sup>b</sup>
	93 ± 1 <sup>d</sup>	94 ± 2 <sup>bc</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>b</sup>	94 ± 1 <sup>c</sup>	99 ± 1 <sup>a</sup>	55 ± 3 <sup>c</sup>	92 ± 2 <sup>c</sup>	93 ± 1 <sup>d</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>a</sup>	90 ± 2 <sup>c</sup>	91 ± 1 <sup>b</sup>	38 ± 5 <sup>c</sup>
	89 ± 1 <sup>d</sup>	91 ± 2 <sup>b</sup>	99 ± 1 <sup>a</sup>	97 ± 1 <sup>b</sup>	73 ± 5 <sup>ab</sup>	99 ± 1 <sup>a</sup>	45 ± 3 <sup>bc</sup>	71 ± 1 <sup>b</sup>	80 ± 7 <sup>cd</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>a</sup>	53 ± 5 <sup>a</sup>	86 ± 4 <sup>b</sup>	23 ± 3 <sup>b</sup>
	71 ± 8 <sup>c</sup>	97 ± 2 <sup>c</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>b</sup>	84 ± 5 <sup>b</sup>	99 ± 1 <sup>a</sup>	48 ± 5 <sup>bc</sup>	73 ± 8 <sup>b</sup>	92 ± 5 <sup>d</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>a</sup>	70 ± 6 <sup>b</sup>	92 ± 2 <sup>b</sup>	25 ± 3 <sup>b</sup>

Note. The difference is reliable at  $p \leq 0.05$  if the letters are not the same.

Due to the formation of the crust, the weed infestation of control plots in the experiment in Location 1 occurred much slower than in the experiment in Location 2. The infestation of the sunflower crop (control variant) with some species of weeds and the stage of their development at the moment of registering 40 days (Location 1) and 30 days (Location 2) after the application of herbicides is presented in Table 5.

The results of determining the effectiveness of weed control in the experiment in Location 1 are presented in Table 6. In this experiment, the effectiveness of controlling dicotyledonous weeds, field mustard and field pennycress using flumioxazine was found to be rather low which is likely related to the formation of the crust on the soil surface that both inhibited the emergence of weed germination and impacted the effect of flumioxazine. Higher effectiveness in controlling mustard and pennycress was achieved if flumioxazine was used in the mixtures with herbicides propisochlor, acetochlor, and fluorochloridone (variants 4, 5, 6). While applying the mixture of flumioxazine and promethrin (variant 3), the effectiveness of controlling mustard and pennycress was lower compared to the application of flumioxazine (variant 2) alone, which is evidence to the antagonistic interaction between these herbicides. The initial effect of flumioxazine on goosefoot and black nightshade was rather high but not prolonged. During the following registration, the effectiveness of controlling these weeds using the herbicide decreased considerably due to the emergence of new weeds and partially due to the restoration of the vegetation by inhibited but not completely destroyed plants of goosefoot and black nightshade. The plants of black-bindweed were not controlled effectively in any experimental variant. The last registration before harvesting did not demonstrate any principal changes in the nature of weed infestation of sunflower crops.

The results of determining the effectiveness of weed control in the experiment in Location 2 are presented in Table 7. Herbicide flumioxazine controlled annual monocotyledonous weeds with high effectiveness but not 100 %. The only effect of adding promethrin to flumioxazine was an insignificant increase in the effectiveness of controlling goosefoot 30 days after the application (variant 3). As for other periods and other species of weeds, the effectiveness of controlling weeds using the mixture of flumioxazine and promethrin did not differ from the effect of flumioxazine alone, which confirms the presence of the antagonistic interaction between these herbicides. At the same time, the ap-

plication of flumioxazine in the mixtures with other herbicides (variant 4, 5, 6) ensured practically a complete destruction of dicotyledonous weeds. As of the moment of the last registration – before sunflower harvesting (140 days after the application of herbicides), the crop was infested with only two species of weeds – goosefoot (*Chenopodium album* L.) and yellow foxtail (*Setaria glauca* (L.) Pal. Beauv.), which were in the phase of seed formation. As of that moment, the plants of mustard and pennycress completed their vegetation. The degree of infestation of the control variant with goosefoot increased a little – up to 10–12 it./sq.m.

Some goosefoot plants were 170 cm high. The second wave of infestation with goosefoot somewhat decreased the effectiveness of the protection in the variants where flumioxazine was applied alone or in the mixture with promethrin. If flumioxazine was applied in the mixtures with promethrin, propisochlor, and acetochlor, high effectiveness of controlling goosefoot was preserved till the completion of sunflower vegetation. Herbicide flumioxazine alone had practically no effect on the infestation of sunflower crops with yellow

low foxtail. The highest effectiveness of controlling yellow foxtail was achieved by the application of the mixture of flumioxazine and acetochlor. The effectiveness of controlling yellow foxtail using the mixture of flumioxazine and fluorochloridone did not differ reliably from the effect of the mixture of flumioxazine and propisochlor and that of the integrated control herbicide (variant 7), but it was lower than the effect of the mixture of flumioxazine and acetochlor.

The results of determining the yield of sunflower seeds during the application of herbicides are presented in Table 8. The formation of the crust and the sparseness of germinated sunflower plants resulted in a decrease in the yield of the seeds in the experiment in Location 1. Due to a low degree of weed infestation, the effectiveness of the application of herbicides in this experiment was comparatively low. The highest yield of sunflower seeds, which reliably exceeded the yield in all the other experimental variants except for the variant with the integrated preparation, was obtained in the variant with the application of the mixture of herbicides flumioxazine and fluorochloridone. In the variants with

**Table 7.** The effectiveness of controlling some species of weeds (%) 30, 60, and 140 days (before harvesting sunflower) after the application of herbicides (location 2)

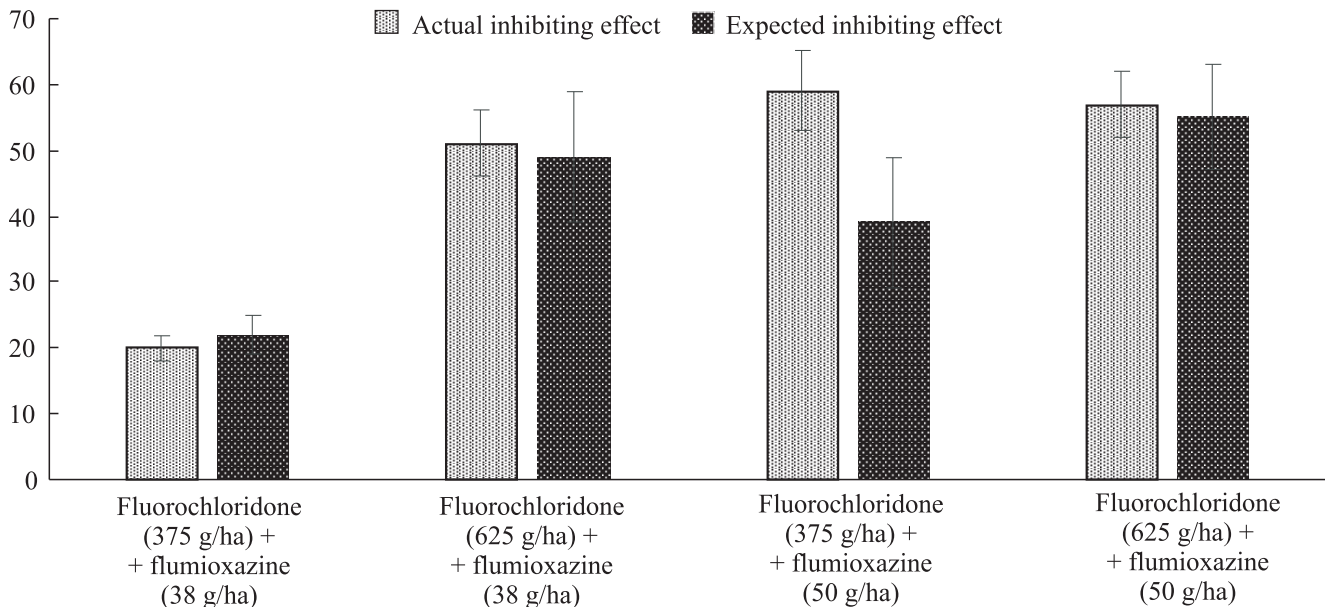
30 days			60 days			140 days	
CHEAL	SINAR	THLAR	CHEAL	SINAR	THLAR	CHEAL	SETPF
89 ± 3 <sup>a</sup>	97 ± 1 <sup>a</sup>	83 ± 3 <sup>a</sup>	81 ± 4 <sup>a</sup>	94 ± 5 <sup>ab</sup>	74 ± 6 <sup>a</sup>	76 ± 6 <sup>a</sup>	13 ± 3 <sup>a</sup>
96 ± 2 <sup>b</sup>	93 ± 4 <sup>a</sup>	84 ± 2 <sup>a</sup>	80 ± 4 <sup>a</sup>	90 ± 5 <sup>a</sup>	73 ± 3 <sup>a</sup>	76 ± 2 <sup>a</sup>	15 ± 5 <sup>a</sup>
97 ± 1 <sup>b</sup>	94 ± 5 <sup>a</sup>	99 ± 1 <sup>b</sup>	95 ± 3 <sup>bc</sup>	99 ± 1 <sup>b</sup>	99 ± 1 <sup>b</sup>	86 ± 4 <sup>ab</sup>	43 ± 3 <sup>b</sup>
99 ± 1 <sup>b</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>b</sup>	97 ± 2 <sup>c</sup>	99 ± 1 <sup>b</sup>	99 ± 1 <sup>b</sup>	95 ± 1 <sup>b</sup>	58 ± 5 <sup>c</sup>
98 ± 1 <sup>b</sup>	99 ± 1 <sup>a</sup>	99 ± 1 <sup>b</sup>	97 ± 2 <sup>c</sup>	99 ± 1 <sup>b</sup>	99 ± 1 <sup>b</sup>	88 ± 3 <sup>ab</sup>	38 ± 5 <sup>b</sup>
97 ± 1 <sup>b</sup>	97 ± 2 <sup>a</sup>	99 ± 1 <sup>b</sup>	91 ± 3 <sup>b</sup>	96 ± 4 <sup>b</sup>	99 ± 1 <sup>b</sup>	78 ± 6 <sup>a</sup>	45 ± 3 <sup>b</sup>

Note. the difference is reliable at  $p \leq 0.05$  if the letters are not the same.

**Table 8.** The yield of sunflower seeds (t/ha) of P63LE10 hybrid during the application of herbicides

Variant	Location 1	Location 2
Control	1.42 ± 0.02 <sup>a</sup>	2.17 ± 0.04 <sup>a</sup>
Flumioxazine (66 g/ha)	1.71 ± 0.03 <sup>bc</sup>	2.61 ± 0.05 <sup>bc</sup>
Flumioxazine (55 g/ha) + promethrin (750 g/ha)	1.63 ± 0.03 <sup>b</sup>	2.66 ± 0.05 <sup>bc</sup>
Flumioxazine (55 g/ha) + propisochlor (1440 g/ha)	1.57 ± 0.04 <sup>b</sup>	2.63 ± 0.04 <sup>bc</sup>
Flumioxazine (55 g/ha) + acetochlor (1350 g/ha)	1.56 ± 0.03 <sup>b</sup>	2.57 ± 0.04 <sup>b</sup>
Flumioxazine (55 g/ha) + fluorochloridone (500 g/ha)	1.90 ± 0.03 <sup>d</sup>	2.78 ± 0.04 <sup>bc</sup>
Integrated herbicide (metolachlor (1406 g/ha) + terbuthylazine (844 g/ha))	1.84 ± 0.04 <sup>cd</sup>	2.80 ± 0.03 <sup>c</sup>

Note. the difference is reliable at  $p \leq 0.05$  if the letters are not the same.



**Fig. 1.** The actual and expected (%) inhibiting effect of the mixtures of fluorochloridone and flumioxazine on the accumulation of wet weight by oil radish plants (the model of annual dicotyledonous weeds). Note:  $m \pm SE$ , the difference between the expected and actual inhibiting effect is statistically insignificant at  $p < 0.05$  (the additive effect of the mixture of herbicides)

the application of flumioxazine in the mixtures with propisochlor and acetochlor, which had higher effectiveness in controlling weeds, the yield was lower than during the application of the mixture of herbicides, flumioxazine, and fluorochloridone, which may be the consequence of the phytotoxic effect on sunflowers. In the experiment in location 2, due to the inhibition by weeds, the sunflower plants in the control variant were considerably lower than the plants in the variants with herbicides. A reliable gain in the yield as compared to the control variant was obtained in all the variants with the application of herbicides. No reliable difference between variants with the application of herbicides was observed except for the variant with the application of the mixture of flumioxazine and acetochlor, where the yield indices were reliably lower than in the variant with the mixture of flumioxazine and fluorochloridone. It is evident that during the application of the mixture of flumioxazine and acetochlor, the phytotoxic effect of herbicides on the crops may eliminate the positive impact achieved by the destruction of weeds.

Thus, the following conclusions can be made from the results of the experiments. Promethrin, a herbicide, ET inhibitor, cannot be used for the combined application with flumioxazine due to the antagonistic interaction between these herbicides. Although the combined application of flumioxazine and herbicides, LCFAS inhibitors, ensures the high effectiveness of weed control, these mixtures cannot be considered promising

means. Firstly, a high probability of crop inhibition is an obstacle to their application. To enhance selectivity, the doses of components could be reduced, but it could lead to the loss of effectiveness. Secondly, the spectra of action of flumioxazine and LCFAS inhibitors on dicotyledonous weeds overlap only in part, so from the standpoint of preventing the occurrence of resistance to herbicides among these weeds, these mixtures do not have principal differences from the mixtures of LCFAS and ET inhibitors. Thus, from the standpoint of selectivity, the effectiveness of weed control, and fighting resistance, the optimal partner to be used together with flumioxazine, a herbicide, PPO inhibitor, is fluorochloridone, a PDS inhibitor. The effect of the tank mixture of flumioxazine and fluorochloridone, used in the application doses of active substances of 55 and 500 g/ha respectively, does not differ from the effect of flumioxazine, applied alone, in terms of selectivity for sunflower. As for the effectiveness of controlling dicotyledonous weeds, the effect of this mixture equals that of the integrated preparation. Here the spectra of action of fluorochloridone and flumioxazine on dicotyledonous weeds are practically identical, so the application of this mixture will definitely be a factor in preventing the emergence of herbicide-resistant biotypes among dicotyledonous weeds.

Considering some negative effects of flumioxazine on sunflowers, the dose of flumioxazine application was somewhat reduced in further studies. At the same



time, a large range of fluorochloridone application doses was used to ensure control over cereal weeds. Due to these changes, it was essential to check the nature of the interaction in the mixtures of flumioxazine and fluorochloridone, depending on the application rates for each component. This study was conducted in conditions of the greenhouse experiment using oil radish as a model of dicotyledons.

Figure 1 presents the results of determining the actual inhibiting effect of the mixtures of flumioxazine and fluorochloridone on the accumulation of wet weight by oil radish plants on day 15 after the application of herbicides as compared to the expected inhibiting effect, which was calculated by formula 3 based on the data regarding the inhibiting effect of some components of the mixture.

The data presented in Fig. 1 demonstrate that under all the ratios of the doses of applying fluorochloridone and flumioxazine, the actual inhibiting effect does not differ reliably from the expected inhibiting effect (in the variant with the application doses of fluorochloridone of 375 g/ha and flumioxazine of 50 g/ha, there was a tendency of the actual effect exceeding the expected effect, but this excess is not reliable). It shows that in a wide range within the recommended application doses, the interaction between flumioxazine and fluorochloridone regarding dicotyledonous weeds is additive. This nature of interaction allows for the assumption that the application of the mixture of flumioxazine and fluorochloridone in sunflower crops should ensure high effectiveness in controlling dicotyledonous weeds.

To check this conclusion, in 2017–2018, field experiments were conducted in the fields of the experimental

farm of the Institute of Plant Physiology and Genetics, the NAS of Ukraine (Location 3). In 2017, the herbicides were applied on April 24, three days after sowing. As of the moment of application, the weather was sunny, the air temperature was +12 °C, the wind velocity was 3 m/s, the soil surface was dry, the moisture was at the depth of 3 cm. In 2018, the herbicides were applied on April 27, the day following sowing. As of the moment of application, it was cloudy (80 %), the air temperature was +16 °C, the wind velocity was 4 m/s, the upper layer of soil was dry, the moisture was at the depth of 1 cm.

No considerable phytotoxic effect of herbicides on sunflowers was found while examining the crops for two years. Only 15 and 30 days after the application of herbicides, some goffering of leaves was observed in an insignificant number of plants on some plots. There was no reliable increase observed in the number of plants with the features of the phytotoxic effect of herbicides as compared to the control.

The nature of weed infestation in sunflower crops (control plots) in the experiments in location 3 after 30 days since the application of herbicides is presented in Table 9. It is evident that in both years, the dominating species of weeds were annual cereal weeds – barnyard grass (2017), and barnyard grass and yellow foxtail (2018). There was some threat to the crops, also posed by annual dicotyledonous weeds – wild radish (2017–2018) and goosefoot (2017).

The results of determining the effectiveness of weed control are presented in Table 10. Under a higher dose of application, fluorochloridone somewhat exceeded

**Table 9.** The infestation of the sunflower crop (control plots) with some species of weeds, the stages of their development, and linear dimensions 30 days after the application of herbicides (Location 3)

Species	Code	Weed infestation (items/sq.m.)	Stage of development (BBCH)	Height (cm)
<i>2017</i>				
Wild radish ( <i>Raphanus raphanistrum</i> L.)	RAPRA	2	30–60	10–20
Goosefoot ( <i>Chenopodium album</i> L.)	CHEAL	5	15–18	3–5
Barnyard grass ( <i>Echinochloa crusgalli</i> (L.) Pal. Beauv.)	ECHCG	20	13–14	3–6
<i>2018</i>				
Wild radish ( <i>Raphanus raphanistrum</i> L.)	RAPRA	2	30	10
Yellow foxtail ( <i>Setaria glauca</i> (L.) Pal. Beauv.)	SETPF	15	12	3–6
Barnyard grass ( <i>Echinochloa crusgalli</i> (L.) Pal. Beauv.)	ECHCG	25	13–14	5–15

flumioxazine in terms of the effectiveness of controlling dicotyledonous weeds. The effectiveness of controlling cereal weeds was lower than the average in the case of applying both herbicides. If the mixtures were used, the effectiveness of controlling both dicotyledonous and cereal weeds increased reliably. The calculation of the expected effectiveness of controlling weeds using the mixtures demonstrated that the actual effectiveness of all the combinations of the application doses of the components for all the species of weeds did not differ reliably from the expected one. Although, in some cases, in particular, in 2018, there was a tendency regarding barnyard grass towards exceeding the value of the actual effectiveness of the mixture as compared to the expected one (Fig. 2), the combination of the obtained data allows for the assumption that in case of the mixtures of flumioxazine and fluorochloridone, the interaction is additive. The highest effectiveness of the mixture was observed for higher doses of introducing the mixture components. In the experiment in 2017, the effectiveness of this mixture in controlling dicotyledonous and cereal

weeds did not differ from the action of the integrated control herbicide. In the experiment in 2018, the effectiveness of the mixture of herbicides in controlling dicotyledon wild radish exceeded, in controlling the cereal weed and barnyard grass – did not differ, and in impacting another cereal weed, yellow foxtail, – was only a little inferior at the end of the sunflower vegetation to the effect of the integrated herbicide metolachlor with terbuthylazine.

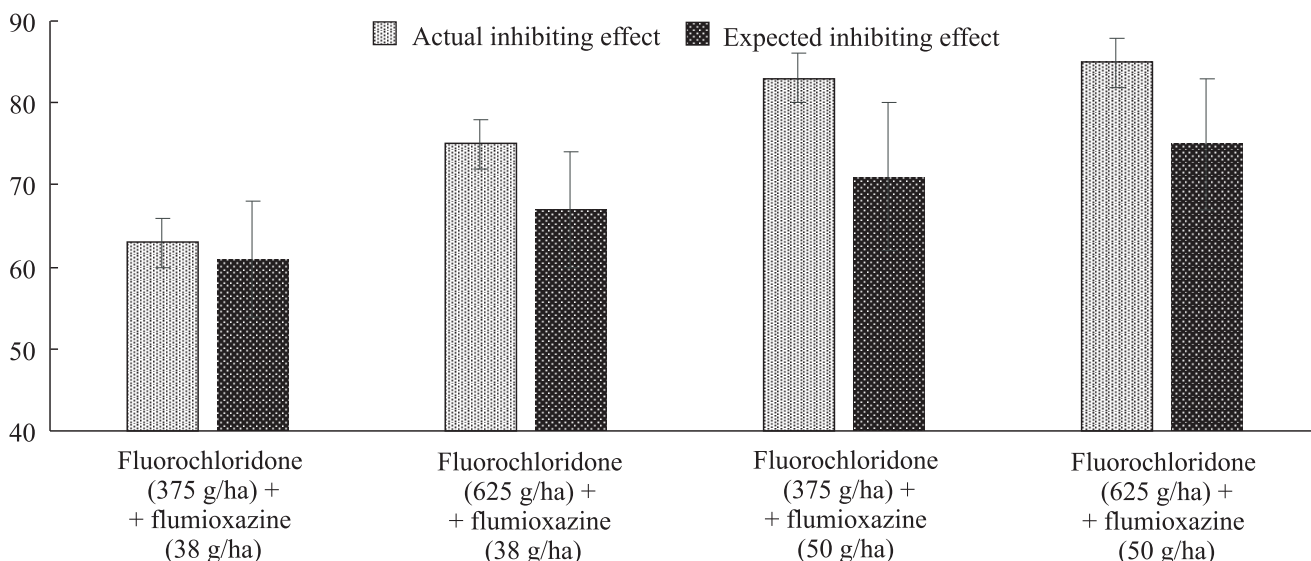
In both years, the yield of the sunflower seeds correlated with the effectiveness of controlling weeds by the herbicides (Table 11). In 2017, a reliable increase in the yield was observed in all the variants with the herbicides, in 2018 – only in the variants with the mixtures of herbicides and at the effect of the integrated herbicide. The largest gain in the yield for the two years of the studies was achieved while applying the integrated herbicide metolachlor with terbuthylazine and the mixture of flumioxazine and fluorochloridone in the maximal doses of the introduction of the components: flumioxazine – 50 g/ha, fluorochloridone – 625 g/ha.

**Table 10.** The effectiveness of controlling some species of weeds (%) 30, 60, and 140 days (before harvesting sunflower seeds) after the application of herbicides (location 3)

30 days			60 days			140 days		
<i>2017</i>								
RAPRA	CHEAL	ECHCG	RAPRA	CHEAL	ECHCG	RAPRA	CHEAL	ECHCG
75 ± 7 <sup>b</sup>	48 ± 7 <sup>a</sup>	10 ± 1 <sup>a</sup>	54 ± 4 <sup>b</sup>	11 ± 1 <sup>a</sup>	29 ± 3 <sup>a</sup>	50 ± 4 <sup>b</sup>	11 ± 1 <sup>a</sup>	21 ± 1 <sup>a</sup>
83 ± 5 <sup>b</sup>	79 ± 3 <sup>b</sup>	30 ± 1 <sup>b</sup>	74 ± 3 <sup>bc</sup>	61 ± 7 <sup>b</sup>	46 ± 2 <sup>b</sup>	71 ± 1 <sup>cd</sup>	53 ± 1 <sup>b</sup>	25 ± 3 <sup>a</sup>
18 ± 1 <sup>a</sup>	45 ± 3 <sup>a</sup>	35 ± 3 <sup>b</sup>	15 ± 8 <sup>a</sup>	15 ± 9 <sup>a</sup>	53 ± 3 <sup>b</sup>	10 ± 7 <sup>a</sup>	10 ± 6 <sup>a</sup>	43 ± 3 <sup>b</sup>
19 ± 2 <sup>a</sup>	48 ± 5 <sup>a</sup>	40 ± 6 <sup>b</sup>	28 ± 3 <sup>a</sup>	26 ± 2 <sup>a</sup>	53 ± 3 <sup>b</sup>	21 ± 1 <sup>a</sup>	21 ± 1 <sup>a</sup>	45 ± 3 <sup>b</sup>
86 ± 1 <sup>b</sup>	73 ± 5 <sup>b</sup>	55 ± 3 <sup>c</sup>	70 ± 4 <sup>bc</sup>	50 ± 4 <sup>b</sup>	55 ± 3 <sup>b</sup>	63 ± 3 <sup>bc</sup>	50 ± 4 <sup>b</sup>	50 ± 1 <sup>b</sup>
90 ± 1 <sup>b</sup>	89 ± 1 <sup>b</sup>	58 ± 3 <sup>c</sup>	85 ± 3 <sup>c</sup>	65 ± 3 <sup>bc</sup>	58 ± 3 <sup>bc</sup>	81 ± 1 <sup>d</sup>	61 ± 1 <sup>bc</sup>	53 ± 3 <sup>bc</sup>
86 ± 4 <sup>b</sup>	81 ± 5 <sup>b</sup>	60 ± 4 <sup>cd</sup>	71 ± 1 <sup>bc</sup>	63 ± 3 <sup>bc</sup>	60 ± 4 <sup>bc</sup>	63 ± 3 <sup>bc</sup>	60 ± 1 <sup>bc</sup>	50 ± 1 <sup>b</sup>
90 ± 1 <sup>b</sup>	89 ± 1 <sup>b</sup>	65 ± 3 <sup>cd</sup>	85 ± 3 <sup>c</sup>	73 ± 3 <sup>c</sup>	65 ± 3 <sup>c</sup>	80 ± 1 <sup>d</sup>	71 ± 1 <sup>c</sup>	61 ± 1 <sup>cd</sup>
90 ± 1 <sup>b</sup>	75 ± 4 <sup>b</sup>	75 ± 3 <sup>d</sup>	80 ± 3 <sup>c</sup>	70 ± 3 <sup>bc</sup>	70 ± 2 <sup>c</sup>	70 ± 2 <sup>cd</sup>	62 ± 2 <sup>bc</sup>	65 ± 2 <sup>d</sup>
<i>2018</i>								
RAPRA	ECHCG	SETPF	RAPRA	ECHCG	SETPF	RAPRA	ECHCG	SETPF
95 ± 3 <sup>ab</sup>	35 ± 3 <sup>a</sup>	31 ± 1 <sup>a</sup>	95 ± 3 <sup>bc</sup>	31 ± 1 <sup>a</sup>	19 ± 1 <sup>a</sup>	91 ± 1 <sup>b</sup>	19 ± 1 <sup>a</sup>	13 ± 3 <sup>a</sup>
99 ± 1 <sup>b</sup>	44 ± 2 <sup>ab</sup>	41 ± 1 <sup>ab</sup>	98 ± 1 <sup>c</sup>	33 ± 1 <sup>a</sup>	21 ± 1 <sup>a</sup>	97 ± 1 <sup>bc</sup>	21 ± 1 <sup>a</sup>	15 ± 3 <sup>a</sup>
90 ± 2 <sup>a</sup>	39 ± 3 <sup>a</sup>	39 ± 3 <sup>a</sup>	91 ± 1 <sup>b</sup>	30 ± 2 <sup>a</sup>	10 ± 1 <sup>a</sup>	80 ± 2 <sup>a</sup>	18 ± 1 <sup>a</sup>	11 ± 1 <sup>a</sup>
91 ± 1 <sup>a</sup>	55 ± 5 <sup>bc</sup>	53 ± 3 <sup>b</sup>	97 ± 1 <sup>bc</sup>	35 ± 2 <sup>a</sup>	23 ± 3 <sup>a</sup>	91 ± 1 <sup>b</sup>	21 ± 1 <sup>a</sup>	15 ± 2 <sup>a</sup>
99 ± 0.3 <sup>b</sup>	63 ± 3 <sup>cd</sup>	60 ± 1 <sup>bc</sup>	99 ± 0.3 <sup>c</sup>	50 ± 4 <sup>b</sup>	43 ± 3 <sup>b</sup>	99 ± 1 <sup>c</sup>	43 ± 3 <sup>b</sup>	35 ± 3 <sup>b</sup>
99 ± 1 <sup>b</sup>	75 ± 3 <sup>de</sup>	e71 ± 1 <sup>cd</sup>	99 ± 0.3 <sup>c</sup>	61 ± 1 <sup>bc</sup>	56 ± 5 <sup>c</sup>	99 ± 0.3 <sup>c</sup>	53 ± 2 <sup>bc</sup>	41 ± 1 <sup>bc</sup>
99 ± 0.3 <sup>b</sup>	83 ± 3 <sup>ef</sup>	80 ± 4 <sup>de</sup>	99 ± 0.3 <sup>c</sup>	58 ± 3 <sup>b</sup>	51 ± 1 <sup>b</sup>	99 ± 0.3 <sup>c</sup>	51 ± 1 <sup>b</sup>	41 ± 1 <sup>bc</sup>
99 ± 0.3 <sup>b</sup>	85 ± 3 <sup>ef</sup>	85 ± 3 <sup>de</sup>	99 ± 0.3 <sup>c</sup>	73 ± 1 <sup>ce</sup>	65 ± 3 <sup>cd</sup>	99 ± 0.3 <sup>c</sup>	64 ± 6 <sup>cd</sup>	50 ± 2 <sup>c</sup>
90 ± 2 <sup>a</sup>	91 ± 2 <sup>f</sup>	90 ± 2 <sup>e</sup>	80 ± 2 <sup>a</sup>	75 ± 3 <sup>c</sup>	70 ± 2 <sup>d</sup>	80 ± 2 <sup>a</sup>	70 ± 2 <sup>d</sup>	61 ± 1 <sup>d</sup>

Note. The difference is reliable at  $p \leq 0.05$  if the letters are not the same.

EFFECTIVENESS OF USING THE MIXTURES OF HERBICIDES FLUMIOXAZINE



**Fig. 2.** The actual and expected effectiveness (%) of controlling barnyard grass using the mixtures of fluorochloridone and flumioxazine (2018). Note:  $m \pm SE$ , the difference between the expected and actual inhibiting effect is statistically insignificant at  $p < 0.05$  (the additive effect of the mixture of herbicides)

**Table 11.** The yield of the sunflower seeds (t/ha) of Karamba hybrid while using fluorochloridone and flumioxazine alone and in the mixtures (location 3)

Variant	2017	2018
Control	2.10 ± 0.04 <sup>a</sup>	2.57 ± 0.05 <sup>a</sup>
Fluorochloridone (375 g/ha)	2.65 ± 0.05 <sup>b</sup>	2.65 ± 0.05 <sup>a</sup>
Fluorochloridone (625 g/ha)	2.76 ± 0.04 <sup>b</sup>	2.71 ± 0.03 <sup>ab</sup>
Flumioxazine (38 g/ha)	2.33 ± 0.04 <sup>b</sup>	2.59 ± 0.04 <sup>a</sup>
Flumioxazine (50 g/ha)	2.36 ± 0.04 <sup>b</sup>	2.64 ± 0.04 <sup>a</sup>
Fluorochloridone (375 g/ha) + flumioxazine (38 g/ha)	2.87 ± 0.06 <sup>c</sup>	2.96 ± 0.05 <sup>b</sup>
Fluorochloridone (625 g/ha) + flumioxazine (38 g/ha)	2.93 ± 0.07 <sup>cd</sup>	3.12 ± 0.07 <sup>bc</sup>
Fluorochloridone (375 g/ha) + flumioxazine (50 g/ha)	2.92 ± 0.03 <sup>cd</sup>	3.09 ± 0.06 <sup>bc</sup>
Fluorochloridone (625 g/ha) + flumioxazine (50 g/ha)	3.10 ± 0.03 <sup>d</sup>	3.26 ± 0.08 <sup>c</sup>
Integrated herbicide (metolachlor (1406 g/ha) + terbuthylazine (844 g/ha))	3.12 ± 0.03 <sup>d</sup>	3.31 ± 0.09 <sup>c</sup>

Note. The difference is reliable at  $p \leq 0.05$  if the letters are not the same.

**DISCUSSION**

In our experiments, we used a herbicide, a PPO inhibitor, with a relatively novel active substance – flumioxazine, which belongs to the class of N-phenyl-phthalimides. This herbicide is used for soil and post-emergence application in sunflower crops, which controls a broad spectrum of dicotyledonous and some cereal weeds. If the preparation is applied to the soil, sensitive weeds either do not germinate or have necrotic damage immediately after germination and perish. When it is applied after germination, sensitive weeds receive remarkable burns and perish (Pledge, 2016).

Herbicides, PPO inhibitors, appeared on the market rather a long time ago (in the 1960s), but the resistance of the weeds to their effect develops rather slowly as compared to, for instance, the herbicides, ALS inhibitors. For instance, at present, there are 13 species of plants, resistant to the herbicides, PPO inhibitors, whereas there are 169 species, resistant to herbicides, ALS inhibitors (Heap, 2022). PPO enzyme (EC 1.3.3.4) is the site of action of the herbicides, which belong to different chemical classes (for instance, diphenyl ethers, heterocyclic phenyl ethers, oxadiazoles, phenylimides, triazolinones and pyrazoles) (Dayan et al, 2014). It participates in the path of the haem synthesis in chloroplasts and mitochondria and chlorophyll in

plastids and catalyzes the transformation of protoporphyrinogen-IX into protoporphyrin-IX. The inhibition of PPO enzyme by the herbicides is accompanied with the abnormal accumulation and release from organelles into cytoplasm of the substrate, protoporphyrinogen-IX, which is transformed into protoporphyrin-IX by the enzymes, associated with the plasmatic membranes with peroxidase activity (Lee and Duke, 1994). Protoporphyrin-IX acts as a photosensibilizer and induces the formation of oxygen radicals with further destruction of cellular membranes and cell components.

The isoforms of protoporphyrinogen oxidase PPO1 (chloroplasts) and PPO2 (mitochondria, sometimes mitochondria and chloroplasts) are coded by two nuclear genes, respectively – *PPXI* and *PPXII* (Dayan et al, 2018). The target resistance of the plants to the herbicides, PPO inhibitors, is related to the mutations of these genes. The mechanism of resistance to the PPO inhibitors in *Amaranthus tuberculatus* (Moq.) Sauer, in which the non-sensitivity to this class of herbicides was first identified, is related to the loss of three consecutive nucleotides, which results in the occurrence of the deletion of glycine residue in position 210 ( $\Delta G210$ ) of the PPO2 enzyme (Patzoldt, 2006; Sarangi et al, 2019) and a change in the architecture of the substrate binding domain (Dyan FE et al, 2010). This mutation was identified in the resistant populations of another species of weeds – *Amaranthus palmeri* S. Watson (Salas et al, 2016). Further studies demonstrated that in addition to the deletion of a glycine residue in some populations of *A. palmeri* the resistance to the herbicides, PPO inhibitors, was caused by the occurrence of other mutations, including the substitution of an aminoacid in position R98 (Giacomini et al, 2017) or glycine for alanine in the catalytic domain PPO2 in position 399 (G399A) (Rangani et al, 2019). It was recently shown that the substitution of the aminoacid Ala-212-Thr in PPO1 adds stability to oxadiazon *Eleusine indica* (L.) Gaertn. (Bi et al, 2020). The resistance of weeds to the herbicides, PPO inhibitors, may also be related to non-target resistance, in particular, caused by the active metabolization of the herbicide (Obenland et al, 2019).

The effect of another herbicide, used in our experiments, – promethrin, similarly to flumioxazine, is also related to the photosynthesis since it is an inhibitor of electron transport in PS 2 of chloroplasts. Promethrin belongs to the chemical group of triazine herbicides. This is a systemic herbicide, working in soil and partially in leaves, designed to control annual dicotyledon and cereal weeds in the fields of a wide spectrum of crops, including sunflower and vegetables. It destroys

annual dicotyledon and cereal weeds, not disrupting the rotation of fields in the rotations. Some annual dicotyledon weeds are better controlled in the phase of early germination (Gesagard, 2022).

In our experiments we also used fluorochloridone, a herbicide that is inhibitor of the phytoendesaturases, key enzymes of carotenoid synthesis, which ensure the transformation of phytoene into  $\zeta$ -carotene. The effect of herbicides, PDS inhibitors, is seen in the accumulation of the phytoene in the plants. As a rule, the herbicides, affecting the biosynthesis of carotenoids, lead to the emergence of “bleached” leaves, which is a result of the initial inhibition of the biosynthesis of carotenoids in combination with the inhibition of chlorophyll biosynthesis and the destruction of the existing chlorophyll (Dan Hess, 2000). The herbicides, which belong to this group of chemical substances, appeared on the market in the early 1970s and make up a small share of herbicides, used in agrarian production (Dayan et al, 2014).

Fluorochloridone is a pre-emergence herbicide of systemic action from the group of piralidones. It is used to control annual dicotyledonous and cereal weeds in the crops of sunflower, chick-pea, potatoes, and carrots. This herbicide is considered the best to control common ragweed (*Ambrosia artemisiifolia* L.). It is highly effective against many dicotyledonous (including the cruciferous) and cereal weeds; it creates a durable protective screen, preserves moisture and nutrients in the soil, minimizing the competition with the weeds. The preparation comes to the plant through the roots and is quickly translocated to the leaf tissues of sensitive weeds. The emerging weeds perish before they appear on the surface or appear to be bleached and perish quickly. However, the effective application of fluorochloridone as a soil herbicide depends on the precipitation during the first 5–7 days after the application (Racer, 2022).

All three abovementioned herbicides are soil herbicides, related to the photosynthesis process but with different sites towards which their action is directed. In the sunflower crops, they control annual dicotyledonous and cereal weeds; the spectra of their action overlap. This is the reason why we chose them for our studies along with other soil herbicides – LCFAS inhibitors propisochlor and acetochlor. Our studies have demonstrated that the combined application of flumioxazine, a herbicide, PPO inhibitor, and promethrin, an inhibitor of electron transport in PS2, shows the antagonistic character of the interaction in controlling



annual dicotyledon weeds. In case of the combined application of flumioxazine, a herbicide, PPO inhibitor, and fluorochloridone, a PDS inhibitor, within the range of the recommended application rates, there is an additive effect on annual monocotyledonous and dicotyledonous weeds. It is also relevant that the tank mixture of flumioxazine and fluorochloridone herbicides at the application rates of 55 and 500 g/ha, respectively, is selective for sunflowers.

## CONCLUSIONS

The optimal partner of flumioxazine, a herbicide that is PPO inhibitor, for complex use in sunflower crops is the herbicide, inhibitor PDS, fluorochloridone. The tank mixture of flumioxazine and fluorochloridone herbicides at the application rates of 55 and 500 g/ha, respectively, is selective for sunflowers. In terms of control efficiency of annual dicotyledons, the mixture of these herbicides exceeded, and in terms of control efficiency of annual cereal weed species was only slightly inferior to the integrated herbicide. Due to the latter fact, it should be noted that in case of a high level of cereal weed infestation in sunflower crops, there may be a need to apply herbicides from the graminicide class regarding the sunflower vegetation on the background of using the mixture of herbicides flumioxazine and fluorochloridone to the emerging plants of the crop. Nevertheless, considering the selectivity for crop and the high effectiveness of controlling dicotyledonous weeds, the application of the tank mixture of herbicides flumioxazine and fluorochloridone in sunflower crops is quite reasonable. In addition, given that flumioxazine and fluorochloridone differ in their phytotoxicity mechanisms but share a common spectrum of controlled weed species, the use of a mixture of these herbicides is a definite factor that minimizes the likelihood of the emergence of herbicide-resistant weed biotypes.

**Compliance with ethical standards.** No experiments, described in this article, involved animals.

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### Ефективність застосування сумішей гербіциду флуміоксазину з флуорохлоридоном у посівах соняшника

М. П. Радченко, Ж. З. Гуральчук, О. П. Родзевич,  
М. В. Хандежина, С. Ю. Мордерер

Інститут фізіології рослин і генетики  
Національної академії наук України,  
Вул. Васильківська, 31/17, Київ, Україна, 03022

E-mail: mradchenko.phd@i.ua, azhanna@ukr.net\*,  
rodzevich2017@gmail.com, 20111975marika@ukr.net,  
morderer@ifig.kiev.ua

**Мета.** Робота спрямована на пошук потенційних партнерів для комплексного застосування гербіциду флуміоксазину в посівах соняшника. Гербіцид належить до інгібіторів ферменту протопорфіриногенаоксидази (ПРОТО) у рослин-бур'янів. Резистентність до даного класу гербіцидів, порівняно з іншими класами, на сьогодні ще не є поширеним явищем. У зв'язку з цим, метою роботи було розробити на базі флуміоксазину гербіцидні композиції, ефективні для попередження виникнення резистентних до гербіцидів біотипів бур'янів. **Методи.** У польових дослідках на посівах соняшнику та у вегетаційних і модельних об'єктах досліджували ефекти взаємодії, ефективність контролювання бур'янів та селективність щодо культури при застосуванні флуміоксазину у сумішах з гербіцидами інгібіторами синтезу жирних кислот з довгим ланцюгом (СЖКДЛ) ацетохлором і пропізохлором, інгібітором транспорту електронів (ТЕ) у фотосистемі 2 (ФС 2) хлоропластів прометрином, інгібітором синтезу каротиноїдів шляхом блокування активності фітоендесагураз (ФДС) флуорохлоридоном. **Результати.** В результаті проведених досліджень показано, що при сумісному застосуванні флуміоксазину з інгібітором ТЕ прометрином взаємодія є антагоністичною, наслідком чого є низька ефективність контролювання бур'янів і зменшення врожайності соняшника. Бакові суміші флуміоксазину з інгібіторами СЖКДЛ ацетохлором і пропізохлором забезпечують високу ефективність контролювання бур'янів, але характеризуються низькою селективністю щодо соняшника. Взаємодія флуміоксазину з флуорохлоридоном у межах рекомендованих норм внесення є адитивною. Бакова суміш гербіцидів флуміоксазину та флуорохлоридону у нормах внесення відповідно 55 та 500 г/га є селективною щодо соняшника. За ефективністю контролювання однорічних дводольних ця суміш гербіцидів перевищувала, а за ефективністю контролювання однорічних злакових видів бур'янів лише незначно поступалася дії контрольного комплексного гербіциду (метолахлор + тербутилазин). За цих норм унесення флуміоксазину та флуорохлоридону забезпечували урожайність соняшника нарівні з даним комплексним гербіцидом. Згідно з цими нормами внесення спостерігали врожайність соняшників нарівні з комплексним гербіцидом. **Висновки.** Серед досліджених гербіцидів оптимальним партнером флуміоксазину для комплексного застосування в посівах соняшника виявився гербіцид інгібітор ФДС флуорохлоридон. Враховуючи те, що флуміоксазин та флуорохлоридон відріз-

няються за механізмами фітотоксичності, але мають спільний спектр контрольованих видів бур'янів, застосування суміші гербіцидів є фактором, який мінімізує вірогідність виникнення резистентних до гербіцидів біотипів бур'янів.

**Ключові слова:** *Helianthus annuus L.*, гербіциди, резистентність, флуміоксазин, флуорохлоридон, прометрин, пропізохлор, ацетохлор.

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