## SUGARBEET TOLERANCE TO COMPLEX MIXTURES IN 2020

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## Summary

- 1. Ethofumesate preemergence (PRE) followed by postemergence (POST) herbicides alone or in combinations did not increase sugarbeet injury in the field.
- 2. High surfactant methylated oil concentrate (HSMOC) increased growth reduction injury from Lorsban plus Stinger applied with glyphosate, ethofumesate and Outlook, 7 days after treatment (DAT). HSMOC with herbicide combinations did not increase growth reduction or impact fresh weigh at 14 DAT.
- 3. Stinger plus Lorsban mixed with glyphosate, ethofumesate and Outlook caused greater growth reduction injury compared with Outlook plus glyphosate and ethofumesate.
- 4. HSMOC rate should be reduced when Lorsban is mixed with glyphosate, ethofumesate and a chloroacetamide. HSMOC should be eliminated from the mixture when/if Stinger and Lorsban are mixed with glyphosate, ethofumesate and a chloroacetamide herbicide.

#### Introduction

Sugarbeet herbicides may be tank mixed legally if all herbicides in the mixture are registered for use on sugarbeet and if no prohibitions against tank mixes appear on a label. Combinations of postemergence herbicides can improve the spectrum of weeds controlled and provide greater total weed control, compared with individual treatments. Mixtures also improve time efficiency as compared with making individual applications. However, the risk of sugarbeet injury also increases with combinations, so combinations should be used with caution. Glyphosate is frequently combined with other herbicides including ethofumesate, Stinger, or a chloroacetamide herbicide (Dual, Outlook, or Warrant) in sugarbeet. On occasion, growers may mix as many as five active ingredients into a single mixture.

Observations of malformation and necrosis injury from POST Betamix and Stinger applied in combination with glyphosate, ethofumesate, and S-metolachlor were assessed in a field near Amenia, ND in 2019. We later learned the sugarbeet field had also been treated with ethofumesate PRE at 3 pt/A. Researchers have reported ethofumesate PRE may change the texture of surface waxes thus increasing the sensitivity of sugarbeet to POST herbicides (Abulnaja et al. 1992).

We have coined the term 'complex mixtures' to describe combinations of three or more herbicides applied POST to sugarbeet. We anticipate two outcomes for the immediate future. First, ethofumesate PRE will be used on more acres for control of waterhemp and kochia in sugarbeet. Second, complex mixtures will be more commonplace in our pursuit of broad spectrum and effective control of glyphosate-resistant weeds.

## Objective

The objective of this research was a) to investigate sugarbeet injury from ethofumesate PRE followed by POST mixtures with glyphosate and b) to investigate the role of HSMOC in relation to sugarbeet injury when applied with complex mixtures.

#### **Materials and Methods**

*Field.* Experiments evaluating sugarbeet injury from ethofumesate PRE followed by POST mixtures with glyphosate were conducted near Christine, ND and Prosper, ND in 2020. The experimental area was prepared for planting by applying the appropriate fertilizer and tillage. Sugarbeet was seeded in 22-inch rows at about 62,000 seeds per acre with 4.6 inch spacing between seeds. Herbicide treatments were applied on May 12 and June 11, and May 30 and June 18 at Christine and Prosper, respectively, with a bicycle wheel sprayer in 17 gpa spray solution through 8002 XR flat fan nozzles pressurized with  $CO_2$  at 43 psi. The treatment list can be found in Table 1. Visible sugarbeet necrosis, malformation, and growth reduction injury was evaluated at both field locations. All evaluations were a visual estimate of injury phenotypes in the four treated rows compared to the adjacent, two-row, untreated strip. Experimental design was randomized complete block with four replications. Data were analyzed with the ANOVA procedure of ARM, version 2020.2 software package.

Preemergence			Sugarbeet
(PRE) Treatment	Postemergence (POST) Treatment	Rate (fl oz / A)	stage (lvs)
_1	Glyphosate + Nortron <sup>2</sup>	32 + 12	2-4
-	Glyphosate + Nortron + Stinger	32 + 12 + 6	2-4
-	Glyphosate + Nortron + Stinger + Outlook	32 + 12 + 6 + 21	2-4
-	Glyphosate + Nortron + Stinger + Outlook + Mustang Maxx	32 + 12 + 6 + 21 + 4	2-4
-	Glyphosate + Nortron + Stinger + Outlook + Mustang Maxx + Betamix	32 + 12 + 6 + 21 + 4 + 32	2-4
Nortron <sup>3</sup>	Glyphosate + Nortron	32 + 12	PRE / 2-4
Nortron	Glyphosate + Nortron + Stinger	32 + 12 + 6	PRE / 2-4
Nortron	Glyphosate + Nortron + Stinger + Outlook	32 + 12 + 6 + 21	PRE / 2-4
Nortron	Glyphosate + Nortron + Stinger + Outlook + Mustang Maxx	32 + 12 + 6 + 21 + 4	PRE / 2-4
Nortron	Glyphosate + Nortron + Stinger + Outlook + Mustang Maxx + Betamix	32 + 12 + 6 + 21 + 4 + 32	PRE / 2-4

Table 1. Herbicide treatment, rate, and application timing at Christine and Prosper, ND in 2020.

<sup>1</sup>- indicates that no PRE herbicide was applied but that POST applications were applied at the leaf stage shown.

<sup>2</sup>All POST entries included Destiny HC (HSMOC) + N-Pak Liquid AMS at 1.5 pt/A + 2.5% v/v. Glyphosate used was Roundup PowerMax.

<sup>3</sup>Nortron was applied at 3 pt/A PRE.

*Greenhouse.* Greenhouse experiments were conducted in 2019, 2020, and 2021 to evaluate sugarbeet injury from complex mixtures POST with or without ethofumesate PRE as well as complex mixtures with or without HSMOC. Greenhouse experiments were a randomized complete block design with a factorial treatment arrangement and three or four replications. Treatment factors were herbicide treatment and PRE herbicide treatment or adjuvant depending on the experiment. Herbicides were applied PRE to 2-4 leaf sugarbeet. Plants were grown at 24 to 27C for a 16 h photoperiod under natural light supplemented with artificial lighting. Plants were watered and fertilized as necessary. Herbicide treatments were applied using a spray booth (Generation III, DeVries Manufacturing, Hollandale, MN) equipped with a single 8001 XR nozzle calibrated to deliver 11 gpa spray solution at 40 psi and 3 mph. The herbicide treatment lists are found in Tables 2 and 3.

Preemergence (PRE)		Rate	Sugarbeet
Treatment	Postemergence (POST) Treatment	(fl oz / A)	stage (lvs)
_1	Glyphosate + Nortron <sup>2</sup>	32 + 12	2-4
-	Glyphosate + Nortron + Stinger	32 + 12 + 6	2-4
-	Glyphosate + Nortron + Stinger + Dual Magnum	32 + 12 + 6 + 20	2-4
	Glyphosate + Nortron + Stinger + Dual Magnum	32 + 12 + 6 + 20	2-4
-	+ Betamix	+ 32	
	Glyphosate + Nortron + Stinger + Dual Magnum	32 + 12 + 6 + 20	2-4
-	+ Betamix + Lorsban	+32 + 16	
Ethofumesate 4 SC <sup>3</sup>	Glyphosate + Nortron	32 + 12	PRE / 2-4
Ethofumesate 4 SC	Glyphosate + Nortron + Stinger	32 + 12 + 6	PRE / 2-4
Ethofumesate 4 SC	Glyphosate + Nortron + Stinger + Dual Magnum	32 + 12 + 6 + 20	PRE / 2-4
Etheferrente A CC	Glyphosate + Nortron + Stinger + Dual Magnum +	32 + 12 + 6 + 20	PRE / 2-4
Ethofumesate 4 SC	Betamix	+ 32	
Eth former to A CC	Glyphosate + Nortron + Stinger + Dual Magnum	32 + 12 + 6 + 20	PRE / 2-4
Ethofumesate 4 SC	+ Betamix + Lorsban	+32 + 16	

<sup>1</sup>- indicates that no PRE herbicide was applied but that POST applications were applied at the leaf stage shown.

<sup>2</sup>All POST entries included Destiny HC (HSMOC) + N-Pak AMS at 1.5 pt/A + 2.5% v/v. Glyphosate was Roundup PowerMax. <sup>3</sup>Ethofumesate 4 SC was applied at 3 pt/A PRE.

Table 3. Herbicide treatment, rate	and application timing in th	e greenhouse in 2020 and 2021.

			Sugarbeet stage
Postemergence Treatment <sup>1</sup>	Rate (fl oz / A)	Adjuvant	(lvs)
Glyphosate + ethofumesate	32 + 12	-	2-4 lvs
Glyphosate + ethofumesate + Outlook	32 + 12 + 21	-	2-4 lvs
Glyphosate + ethofumesate + Outlook + Lorsban	32 + 12 + 21 + 16	-	2-4 lvs
Glyphosate + ethofumesate + Outlook + Lorsban + Stinger	32 + 12 + 21 + 16 + 6	-	2-4 lvs
Glyphosate + ethofumesate	32 + 12	HSMOC <sup>2</sup>	2-4 lvs
Glyphosate + ethofumesate + Outlook	32 + 12 + 21	HSMOC	2-4 lvs
Glyphosate + ethofumesate + Outlook + Lorsban	32 + 12 + 21 + 16	HSMOC	2-4 lvs
Glyphosate + ethofumesate + Outlook + Lorsban + Stinger	32 + 12 + 21 + 16 + 6	HSMOC	2-4 lvs

<sup>1</sup>All mixtures contained N-Pak Liquid AMS at 2.5% v/v. Glyphosate used was Roundup PowerMax and ethofumesate was Ethofumesate 4SC.

<sup>2</sup>HSMOC=Destiny HC at 1.5 pt/A.

Visual sugarbeet injury evaluations (0 to 100% with 100% reflecting complete sugarbeet death) were completed 3, 7, and 14 ( $\pm$ 3) DAT. Above-ground fresh weight (g pot<sup>-1</sup>) were collected at the conclusion of the experiment or after the 14 DAT evaluation. Data were analyzed with the ANOVA procedure of ARM, version 2020.4 software package.

#### Results

*Field.* The Christine experiment was discontinued due to poor sugarbeet stands. At Prosper, PRE ethofumesate had minimal effect on sugarbeet injury across POST treatments (Factor A) or ethofumesate did not increase sugarbeet injury from postemergence herbicides, even when Betamix was part of the mixture (Factor A  $\times$  B) (Table 4).

			Grow	th Reduct	tion
Preemergence Herbicide	Postemergence (POST) Herbicide	Rate	10 DAT <sup>1</sup>	20 DAT	Mean <sup>2</sup>
	<b>z</b> , ,	fl oz/A		%	
-	Glyphosate + Nortron <sup>4</sup>	32 + 12	5	0	5
-	Glyphosate + Nortron + Stinger	32 + 12 + 6	0	0	0
-	Glyphosate + Nortron + Stinger + Outlook	32 + 12 + 6 + 21	26	9	20
-	Glyphosate + Nortron + Stinger + Outlook + Mustang Maxx	32 + 12 + 6 + 21 + 4	30	25	26
-	Glyphosate + Nortron + Stinger + Outlook + Mustang Maxx + Betamix	32 + 12 + 6 + 21 + 4 + 32	58	28	47
Nortron <sup>3</sup>	Glyphosate + Nortron	32 + 12	3	0	4
Nortron	Glyphosate + Nortron + Stinger	32 + 12 + 6	10	9	13
Nortron	Glyphosate + Nortron + Stinger + Outlook	32 + 12 + 6 + 21	12	10	16
Nortron	Glyphosate + Nortron + Stinger + Outlook + Mustang Maxx	32 + 12 + 6 + 21 + 4	31	21	33
Nortron	Glyphosate + Nortron + Stinger + Outlook + Mustang Maxx + Betamix	32 + 12 + 6 +21+ 4 + 32	67	20	41
P-Value, Factor A	PRE ethofumesate		0.2847	0.5560	0.6842
P-Value, Factor B	POST Herbicide treatments		0.0001	0.0001	0.0001
P-Value, Factor A×B	PRE herbicide × POST Herbicide treatment		0.1954	0.5112	0.6258

Table 4. Sugarbeet growth reduction in response to preemergence and postemergence herbicide treatments at
Prosper, ND in 2020.

<sup>1</sup>DAT=Days after POST treatment. <sup>2</sup>Average of growth reduction 5, 10, and 20 DAT.

<sup>3</sup>Nortron was applied at 3 pt/A.

<sup>4</sup>All POST entries included Destiny HC (HSMOC) + N-Pak Liquid AMS at 1.5 pt/A + 2.5% v/v. Glyphosate used was Roundup PowerMax.

Sugarbeet injury 10 DAT, 20 DAT or the average across evaluations was greater when the number of herbicides mixed with glyphosate and ethofumesate increased, averaged across ethofumesate PRE (Table 5). Growth reduction

injury was negligible when Stinger was mixed with glyphosate plus ethofumesate but increased when Mustang Maxx was combined with glyphosate, ethofumesate, Stinger and Outlook. Necrosis and malformation damage varied from plant to plant in plots. Sugarbeet injury was greatest or tended to be greatest when Betamix was combined with glyphosate, ethofumesate, Stinger, Outlook and Mustang Maxx. Sugarbeet necrosis injury from mixtures including Betamix was not consistent but generally was negligible (data not presented). Malformation injury was greater when Outlook, Mustang Maxx or Betamix was mixed with glyphosate, ethofumesate and Stinger (data not presented).

Table 5. Sugarbeet growth reduction in response to postemergence herbicide treatments with or without
ethofumesate PRE at Prosper, ND in 2020.

		Gro	wth Reduction	1
Postemergence (POST) Herbicide <sup>1</sup>	Rate	$10 \text{ DAT}^2$	20 DAT	Mean <sup>2</sup>
	fl oz/A		%	
Glyphosate + Nortron	32 + 12	4 c	0 c	5 d
Glyphosate + Nortron + Stinger	32 + 12 + 6	5 c	4 bc	6 d
Glyphosate + Nortron + Stinger + Outlook	32 + 12 + 6 + 21	19 b	9 b	18 c
Glyphosate + Nortron + Stinger + Outlook + Mustang Maxx	32 + 12 + 6 + 21 + 4	30 b	23 a	29 b
Glyphosate + Nortron + Stinger + Outlook + Mustang Maxx + Betamix	32 + 12 + 6 + 21 + 4 + 32	62 a	24 a	44 a
P-value		0.0001	0.0001	0.0001

<sup>1</sup>All POST entries included Destiny HC (HSMOC) + N-Pak Liquid AMS at 1.5 pt/A + 2.5% v/v. Glyphosate used was Roundup PowerMax.DAT=Days after POST treatment.

<sup>2</sup>Average of growth reduction 5, 10, and 20 DAT.

*Greenhouse*. Ethofumesate 4SC at 3 pt/A PRE did not affect sugarbeet malformation or growth reduction from POST herbicide treatments and, in general, did not have any effect on sugarbeet necrosis (Table 6).

# Table 6. Sugarbeet necrosis, malformation, and growth reduction injury from postemergence herbicide treatments with and without Ethofumesate 4SC PRE at 3 pt/A in the greenhouse in 2020.

	Necrosis <sup>2</sup>		Malformation		Growth Reduction	
Herbicide treatment <sup>1</sup>	No PRE	PRE	No PRE	PRE	No PRE	PRE
	%%%%%					
Base <sup>3</sup>	1 c <sup>4</sup>	1 c	3	5	2	3
Base + Stinger	0 c	2 c	17	15	2	4
Base + Stinger + Dual Magnum	7 bc	0 c	12	10	0	4
Base + Stinger + Dual Magnum + Betamix	11b	11 b	30	27	22	11
Base + Stinger + Dual Magnum + Betamix + Lorsban	23 а	13 b	25	27	18	19
P-Value	0.02	241	0.91	59	0.15	94

<sup>1</sup>All POST entries included Destiny HC (HSMOC) + N-Pak Liquid AMS at 1.5 pt/A + 2.5% v/v.

<sup>2</sup>Necrosis, malformation and growth reduction averaged across evaluations.

 $^{3}$ Base = Roundup PowerMax at 32 fl oz/A + Ethofumesate 4SC at 12 fl oz/A.

<sup>4</sup>Means within a main effect not sharing any letter are significantly different by the LSD at the 10% level of significance.

Due to the lack of effect from Ethofumesate 4SC PRE, data were combined to the POST treatment level (Table 7). The addition of Betamix and Lorsban increased sugarbeet necrosis, malformation, and growth reduction injury compared with glyphosate plus ethofumesate or glyphosate plus ethofumesate plus Stinger.

Herbicide treatment <sup>1</sup>	Necrosis <sup>2</sup>	Malformation	Growth Reduction
		%%	
Base <sup>3</sup>	$1 c^4$	4 c	3 b
Base + Stinger	1 c	16 b	3 b
Base + Stinger + Dual Magnum	3 c	11 bc	2 b
Base + Stinger + Dual Magnum + Betamix	11 b	28 a	17 a
Base + Stinger + Dual Magnum + Betamix + Lorsban	18 a	26 a	18 a
P-Value	0.0001	0.0001	0.0001

Table 7. Sugarbeet necrosis, malformation, and growth reduction injury in response to postemergence herbicide treatments averaged across PRE herbicide in the greenhouse in 2020.

<sup>1</sup>All POST entries included Destiny HC (HSMOC) + N-Pak Liquid AMS at 1.5 pt/A + 2.5% v/v.

<sup>2</sup>Necrosis, malformation and growth reduction averaged across evaluations.

 $^{3}$ Base = Roundup PowerMax at 32 fl oz/A + Ethofumesate 4SC at 12 fl oz/A.

<sup>4</sup>Means within a main effect not sharing any letter are significantly different by the LSD at the 10% level of significance.

The second greenhouse experiment considered both the visual assessment of sugarbeet growth reduction injury and sugarbeet fresh weight (g/pot) in response to herbicide mixtures both with and without HSMOC. Sugarbeet injury from glyphosate + ethofumesate + Outlook + Stinger + Lorsban was greatest 7 DAT and was greater or tended to be greater when HSMOC was added with the mixture (Table 8). Injury decreased with time and HSMOC, when added to herbicide mixtures, did not influence growth reduction or fresh weight at 14 DAT.

Visible sugarbeet growth reduction injury at 7 and 14 DAT increased when Outlook or Outlook + Lorsban +/-Stinger was mixed with glyphosate plus ethofumesate (Table 9). Growth reduction injury tended to be less 14 DAT than 7 DAT indicating that plants were starting to recover from their injury. Sugarbeet fresh weight per pot tended to be reduced as the complexity of mixtures increased.

Table 8. The effect of herbicide mixtures both with and without high surfactant methylated oil (HSMOC) on
visual sugarbeet growth reduction injury and fresh weight averaged across two greenhouse runs in 2020 to
2021.

		Growth	Reduction	Growth F	Reduction		
Herbicide treatment	Rate	$7 \text{ DAT}^1$		14 DAT		Fresh Weight	
		No		No		No	
		HSMOC	HSMOC	HSMOC	HSMOC	HSMOC	HSMOC
	fl oz/A	%		g/pot			
Base <sup>2</sup>		$6 ab^3$	1 a	6	12	32.6	30.3
Base + Outlook	21	18 c	15 bc	17	23	30.3	27.8
Base + Outlook and Lorsban	21 + 16	22 c	34 d	19	23	29.4	26.3
Base + Outlook, Lorsban and Stinger	21 + 16 + 6	38 d	49 e	32	39	29.8	28.0
P-Value		0.0	257	0.9	401	0.9	869

<sup>1</sup>DAT=Days after POST treatment.

<sup>2</sup>Base= Roundup PowerMax at 32 fl oz/A + Ethofumesate 4SC at 12 fl oz/A + N-Pak Liquid AMS at 2.5% v/v.

<sup>3</sup>Means within a main effect not sharing any letter are significantly different by the LSD at the 10% level of significance.

Table 9. The effect of herbicide mixtures averaged across both with and without high surfactant methylated
oil (HSMOC) on visual sugarbeet growth reduction injury and fresh weight averaged across two greenhouse
runs in 2020 to 2021.

		Growth Reduction	Growth Reduction	Sugarbeet Fresh	
Herbicide treatment	Rate	$7 \text{ DAT}^2$	14 DAT	Weight	
	fl oz/A	0/0		g/pot	
Base <sup>2</sup>		4 d <sup>3</sup>	9 c	31.4	
Base + Outlook	21	16 c	20 b	29.0	
Base + Outlook and Lorsban	21 + 16	28 b	21 b	28.9	
Base + Outlook, Lorsban and Stinger	21 + 16 + 6	43 a	35 a	28.1	
P-Value		0.0001	< 0.0001	0.1436	

<sup>1</sup>DAT=Days after POST treatment.

<sup>2</sup>Base= Roundup PowerMax at 32 fl oz/A + Ethofumesate 4SC at 12 fl oz/A + N-Pak Liquid AMS at 2.5% v/v.

<sup>3</sup>Means within a main effect not sharing any letter are significantly different by the LSD at the 10% level of significance.

Malformation injury from Stinger was negligible in these greenhouse experiments (data not presented). However, Stinger did cause greater sugarbeet growth reduction injury when added to Outlook + Lorsban compared with Outlook + Lorsban alone. Sugarbeet growth reduction injury was observed as both stature reduction and speckling of the leaves, presumably from the oils in some of the herbicide formulations as well as in the HSMOC adjuvant.

#### Conclusion

Pesticides (herbicides, fungicides, and insecticides) approved for use in sugarbeet usually are safe to sugarbeet when applied individually. These same pesticides applied in mixtures, however, occasionally injure sugarbeet since each pesticide must be detoxified by the plant. Environmental stressors such as low air and soil temperatures or saturated soil-water content are conditions that often reduce photosynthesis and may reduce energy needed for the developing sugarbeet to metabolize pesticides (Smith and Schweizer 1983), thus increasing the risk of sugarbeet injury. Sugarbeet is better able to manage biotic or abiotic stressors as it develops; sugarbeet with more leaf area have greater metabolic activity, dissipating the effect of herbicides, and other stressors.

These field and greenhouse experiments suggest sugarbeet injury concerns with complex pesticide mixtures. For example, we observed injured phenotypes suggesting Betamix or Betamix plus Lorsban caused sugarbeet injury. However, we do not believe Betamix or Lorsban alone are the culprits since Betamix with glyphosate and ethofumesate caused necrosis and malformation injury 14 DAT similar to glyphosate and ethofumesate (in full disclosure we never evaluated Lorsban plus glyphosate or ethofumesate compared with glyphosate and ethofumesate alone). But rather injury from Betamix and/or Lorsban are exacerbated by 'activators' such as a Stinger combined with glyphosate, ethofumesate and chloroacetamide herbicides in complex mixtures under certain environmental conditions. HSMOC had less effect on sugarbeet injury than the herbicides did and it's unclear how much of the injury from the herbicide can be attributed to the active ingredient versus the oil content of the formulation.

## Literature Cited

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