EFFECTS OF COMBINE CROP HARVESTING ON WEED SEEDS AND SUBSEQUENT POTENTIAL FOR EMERGENCE OF HERBICIDE RESISTANT BIOTYPES

By

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INTRODUCTION

Each of the parts of this thesis is a separate manuscript to be submitted for publication; Part I in <u>Weed Science</u>, a Weed Science Society of America publication, and Part II in <u>Weed Technology</u>, a Weed Science Society of America publication.

PART I

EFFECTS OF COMBINE HARVESTING ON GERMINATION OF WILD BUCKWHEAT (<u>POLYGONUM</u> <u>CONVOLVULUS</u>) ACHENES AND SUBSEQUENT POTENTIAL FOR EMERGENCE OF HERBICIDE RESISTANT BIOTYPES

Effects of combine harvesting on germination of wild buckwheat (<u>Polygonum convolvulus</u>) achenes and subsequent potential for emergence of herbicide resistant biotypes¹ RANDALL S. CURRIE AND THOMAS F. PEEPER²

<u>ABSTRACT</u>. Wild buckwheat (<u>Polygonum convolvulus</u> L. #³ POLCO) achenes harvested with standing hard red winter wheat with six grain harvesting combines exhibited differing degrees of scarification. Combine harvesting removed the persistent calyx from 6 to 29% of wild buckwheat achenes and removed the ovary walls from up to 28% of achenes collected in the grain bin. Germination of the achenes with ovary walls removed was 19 to 99% greater than hand harvested achenes. From 25 to 55 percent of the sound mature achenes that entered combine headers entered the grain bins. Treatments of chlorsulfuron (2-chloro-N-[[(4methoxy-6-methyl-1,3,5-triazin-2-y1)amino]carbonyl] benzenesulfonamide), bromoxynil (3,5-dibromo-4-hydroxybenzonitrile) plus MCPA (2methyl-4-clorophenoxyacetic acid) and picloram [4-amino-3,5,6trichloropicolinic acid] plus 2,4-D (2,4-dichlorophenoxy acetic acid) reduced wild buckwheat achene production in wheat by 91 to 100%.

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³Letters following this symbol are a WSSA-approved computer code from Composite List of Weeds, Weed Sci. 32, Suppl. 2.

Although combine scarification altered the germination of some wild buckwheat achenes, integration of the factors affecting the emergence of herbicide resistant biotypes revealed that the time required for resistance to appear should not be greatly influenced by combine harvesting.

<u>Additional index words</u> Dormancy, scarification, seed germination, POLCO.

INTRODUCTION

Because wild buckwheat reduces crop yields and encumbers harvesting and tillage operations on six continents, it is frequently the target of herbicide application (5, 6, 7, 29). Factors that could affect the appearance of herbicide resistance in this species should be better understood. Triazine resistant biotypes of wild buckwheat have been observed in corn (Zea mays L.) fields in Pennsylvania and Austria (15, 29). The many factors involved in the potential appearance of triazine resistant weed biotypes have been discussed in several recent reviews (9, 10, 11, 12, 13). These factors have been integrated into the model PRY = PRO $[1 + ([F(1/[1-EK])]/SL)]^{Y}$ where PRY is the proportion of resistant individuals in a population after Y years of continuous selection by a single herbicide treatment (8). The proportion of resistant biotypes present in the population in year zero (PRO) is assumed to be the mutation frequency of 1×10^{-7} , while F is the percent fitness reduction of resistant biotypes, EK is the effective kill or percent reduction in seed production due to

application of the herbicide and SL equals the time a seed remains viable in the soil.

The two factors with this model that can be manipulated to affect the rate of appearance of resistant biotype are EK and SL. Since weed stands heavily thinned by herbicide treatment possesses the plasticity to increase seed production of survivors (17), EK data is difficult to obtain and is typically unavailable. The more influential of these factors is SL, since all of the other factors are buffered by the effects of all previous years weed seed production. Production of up to 152 x 10^6 wild buckwheat seeds/ha/year has been measured (6, 28, 31). The germination of these seeds occurs over a period of years so that the individuals present in any 1 year are a collection of many previous years seed production. Previous work has demonstrated that tillage depth, location ecology, insect feeding during maturation, moisture, and temperature affect weed seed dormancy (4, 18, 19, 20, 24). However, the most influential factor controlling wild buckwheat dormancy is the integrity of its black ovary wall (16, 18).

Although factors such as concave clearance, cylinder speed and combine design influence pericarp damage of crop grain during harvesting, little research on combine scarification of wild buckwheat achenes has been conducted (1, 2, 3, 21, 22, 23). Porter (25) examined one sample of commercial alfalfa seed in which 24% of the wild buckwheat achenes had their pericarps removed by harvesting and/or processing. After 8 months of dry storage, these decorticated achenes had 20 ±1% germination. Germination of freshly harvested achenes has been reported to vary from 2.4 to 95% (6, 18, 31). Achene harvesting methods are inadequately described in these reports to permit defining

the role of harvesting method on achene dormancy. The primary objectives of this study were to determine the effects of combine harvesting on wild buckwheat achene germination and determine EK for three herbicide treatments used for wild buckwheat control. A second objective was to enter the data into Gressel's model (8) to evaluate the possible impact of combine harvesting on emergence of herbicide resistance.

MATERIALS AND METHODS

During the summers of 1985 and 1986, samples of grain and wild buckwheat achenes were collected as standing hard red winter wheat was being harvested with six self-propelled grain harvesting combines. These machines were selected to represent the types of threshing mechanisms commonly used for harvesting wheat. The four commercial style grain combines included: a Sperry New Holland TR85³ which has two longitudinally mounted rotors, threshing and separating concaves, a discharge beater and cleaning shoe; a Gleaner N-6⁴ with a transverse mounted single rotary segment threshing concave, a 360° separating cage, accelerator rolls and cleaning shoe; a Gleaner A⁴ with a transverse mounted conventional rasp bar cylinder with closed bottom concave mounted in the feeder housing and a straw walker-double sieve system for grain separation; and a Gleaner C2⁴ which is similar to the Gleaner A except that it is larger and thus has greater capacity. The

³Sperry Rand Corp., New Holland, Penn.

⁴Allis-Chalmers Corp., Independence, MO.

two plot combines, the HEGE 125C⁵ and a KEM SP50⁶ both have transverse mounted conventional rasp bar cylinders located behind the feeder housing. Delivery to the cylinder is with a feeder rake chain on the KEM SP50 and with a flat conveyor belt on the HEGE 125C. Unlike the commercial machines, neither plot combine has a tailings return system. Combines were set and operated at or near full capacity in accordance with manufacturers specifications for harvesting wheat (Table 1). Crops with which wild buckwheat was harvested had received no herbicide treatment.

In 1985, 4000 to 5000 g bulk samples of grain and wild buckwheat achenes were collected in June from the grain bins of these combines as they harvested hard red winter wheat infested with wild buckwheat. Not all combines were used at all locations. The Gleaner A and HEGE 125C were used at the North Agronomy Research Station, Stillwater, OK (location 85-1), the Gleaner C2, Gleaner A, HEGE 125C and KEM SP50 were used at the Agronomy Research Station, Stillwater, OK (location 85-1), the Gleaner C2, Gleaner A, HEGE 125C and KEM SP50 were used at the Agronomy Research Station, Stillwater, OK (location 85-2). Only the Gleaner A was used at the Lake Carl Blackwell Research area, Orlando, OK, (location 85-3), and only the Sperry New Holland TR85 was used in Marion Co., KS (location 85-4). Hand harvested samples of 1000 to 1500 achenes were also randomly collected at each site. Achenes harvested from location 85-1 were segregated by hand into achenes with an intact persistent calyx (brown), achenes with the persistent calyx

HEGE Saazuchtmaschinen., Domaene Hohebuch D-7112, Waldenburg/Wertt, West Germany.

⁶Kincaid Equipment Manufacturing, Haven, KS.

removed (black) and true seeds with the persistent calyx and black ovary walls removed (white). Hand sorting these achenes revealed that distinct differences in achene size existed in the samples. Therefore, all subsequent segregations were divided with a seed blower into lighter and heavier achenes, which weighed 3.6 and 4.6 \pm SE 1.0 mg, respectively. This procedure also removed empty achenes and immature achenes lacking endosperm which weighed < 3 mg. The weight classes were then subdivided by color as described. Four 25 seed replications of each segregate from each field sample were then placed on 1 g of absorbent paper toweling in petri dishes, hydrated with 10 ml of distilled water. Preliminary tests revealed that achenes stored dry at 25°C for approximately 1 month would not germinate under these moist conditions at 20°C in diffuse light. Achenes were therefore stratified by placing these dishes in dark storage at 4°C in a walk-in refrigerator. After 8 weeks an additional 5 ml of distilled water was added to each petri dish and the dishes were then placed in a germinator in a randomized block design at 20°C with near 100% relative humidity in diffuse light. Germination was recorded every 3 days for 15 days.

Wild buckwheat achenes were collected in a similar fashion in June, 1986, except for the following changes in procedures. The locations were near Enid, OK (Location 86-1), 5 km west of Stillwater, OK (location 86-2), at the South Central Research Station, Chickasha, OK (location 86-3), at the North Agronomy Research Station, Stillwater, OK (location 86-4) and in a field of oats on the North Agronomy Research Station, Stillwater, OK (location 86-5). Achenes were harvested with the Gleaner A, HEGE 125C and KEM SP50 at all locations

in 1986. In addition a Gleaner N6 and Sperry New Holland TR85 were used at 86-1, a Gleaner N6 was used at 86-3, and the Gleaner C2 was used at 86-5. Sample size was also increased to approximately 15 kilograms of grain from each combine and samples were collected from four randomly selected portions of each of the fields. Samples were collected as they entered the grain bin while the combines were operating near full capacity. This expanded sampling was intended to define the net effect of combine harvesting on the population of achenes in addition to its effect on individual achenes. Samples were cleaned, classified and counted as in 1985. Although not all seed classes were present in adequate numbers in all samples, where possible, four sets of 25 achenes from each of the six seed classifications for each of the four field samples were germinated as described earlier. Differences in germination rate were determined by using the categorical modeling procedure of S.A.S. to construct a three dimensional contour response surface for the germination of each of the six seed classes through time for each of the combines (27). This procedure uses a Chi square test for lack of fit of these surfaces through time.

To verify that samples collected from the grain bin represented a random portion of the seeds entering the combine, twenty 10,000 achene samples of wild buckwheat were delivered to the header of selected combines as they were harvesting uninfested wheat. The achenes used were collected with a vacuum from the soil surface after the plants had naturally matured and seeds had dehisced. A small seed cleaner was used to remove non-achene material. When the threshing mechanism of each combine was fully loaded with wheat and operating normally, a

10,000 achene sample was introduced into the header. Grain entering the bin was collected for 2 minutes. The combine was then cleaned after each sample by continued operation for several hundred meters in a wheat field that contained no wild buckwheat. This procedure was replicated four times for each machine. The proportion of those achenes that entered the bin was determined. Since the quantity of grain collected from the large machines was substantial, the grain was mixed and the number of wild buckwheat achenes present was estimated from four subsamples from each replicate.

Estimations of EK were made by comparisons of wild buckwheat seed content in wheat harvested from herbicide treated and untreated plots for three herbicide treatments. The four locations selected were unassociated with previously described experiments. Treatments were applied to 1.5 by 6 m plots arranged in a randomized complete block design with four replications. Bromoxynil plus MCPA was applied at 280 + 280 g/ha at two locations in 1984. Chlorsulfuron was applied at 23 g/ha at three locations and 26 g/ha at two locations in 1986. Picloram plus 2,4-D was applied at 26 + 420 g/ha at five locations over 2 years. Applications were made to wild buckwheat in the two to five leaf stage. Plots were harvested with a small plot combine and recleaned to remove materials other than wild buckwheat achenes. EK was determined as the percent reduction in the number of wild buckwheat achenes in 25 g samples of harvested grain.

RESULTS AND DISCUSSION

At location 85-1 combine harvesting removed the brown persistent calyx from 26 to 30% of the wild buckwheat achenes and 2% of the

achenes harvested by the HEGE 125C had complete ovary wall removal (Table 2). All hand harvested achenes had intact calyxes. The achenes with complete ovary wall removal had 55% germination versus 11% or less for all other combine harvested seed. Much less brown and black combine harvested seed germinated than hand harvested seed. This was inconsistent with results of earlier research on pericarp injury (16, 18, 26), and indicates that the combine harvested sample may not have been representative of the entire population.

Although the sampling in 1985 permits inference primarily to the effects of combine harvesting on-individual achenes, some inference to the population can be made based on multiple T tests utilizing the other two locations (85-2, 85-3) where the Gleaner A was used as replications. Using this analysis, the lighter and heavier black achenes had 3 and 37% germination, respectively. Therefore in experiments conducted in 1986, the weight factor of the population of achenes was more fully investigated. As observed in 85-1, combined analysis of Gleaner A harvested achenes from 85-2 and 85-3 showed a pattern of persistent calyx removal similar to that seen in 85-1 with the mean combine harvested sample containing 77, 22, and 1% brown, black and white seeds, respectively.

In 1985, the HEGE 125C, KEM SP50, and the Gleaner C2, removed the calyx from 20 to 28% of lighter weight achenes as the Gleaner A did at 85-1, 85-2, and 85-3. Calyx removal from the heavier achenes varied from 19 to 46%. Of the approximately 1500 achenes examined from each combine, only 0 to 5% had their ovary walls completely removed (Table 3). The SNH TR85 removed the calyx from 61 and 77% and the ovary wall from 0.4 and 13% of the lighter and heavier achenes, respectively. The

greater achene injury with the SNH TR85 may have been influenced by achene maturity. Although the harvesting dates were all within one week, the achenes harvested with the SNH TR85 were less mature in the wheat in Kansas and the calyxes may have been softer.

Germination of hand harvested achenes from the different locations varied from 4 to 33% in 1985 (Table 4). Such wide variation was also reported by Justice (18). At location 85-2, brown achenes harvested by the Gleaner A, HEGE 125C and KEM SP50 had from 17 to 36% and 27 to 38% germination of the lighter and heavier achenes, respectively, compared to 4 and 1% for lighter and heavier hand harvested achenes (Table 4). Examination of these achenes suggested that impact fractures of the pericarp beneath the calyx accounted for this enhancement. Lighter achenes harvested at 85-4 by the SNH TR85 had less germination than hand harvested achenes. The lighter brown and black achenes harvested by the Gleaner A and SNH TR85 at 85-3 and 85-4, respectively, had reduced germination compared to hand harvested achenes. The heavier black achenes harvested by the Gleaner A, HEGE 125C and KEM SP50 at 85-2 had 33 to 50% germination compared to hand harvested achenes with 1%.

Germination enhancement was most pronounced in heavy white seeds. These seeds harvested by the HEGE 125C and KEM SP50 at location 85-2 had 25 and 94% germination respectively compared to 1% for hand harvested achenes. The germination of heavy white seeds harvested by the Gleaner A and the SNH TR85 from locations 85-3 and 85-4, respectively, had 62 and 61% germination, respectively, compared to 27 and 33% for their respective hand harvested checks.

Since the sampling procedure in 1985 limited the size of some segregates and comparisons that could be made, in 1986, the samples collected were increased in size to about 10^4 achenes/combine/location. With the larger sample size, the seed blower was able to segregate each color of achene from each location by weight with non-overlapping 95% confidence intervals except for black achenes from location 86-1 and white seed from locations 86-2, 86-3, and 86-4 (Table 5). The broader confidence intervals of these exceptions were due to the smaller number of achenes in these classifications. Lighter brown and black seeds were 1.4 ± SE 0.3 mg and 1.3 ± 0.2 mg lighter than heavier brown and black seeds, respectively.

With the exception of Gleaner A harvested achenes, there was no clear relationship between achene weight and the percent of achenes with calyx and/or ovary wall removal (Table 6). The combine harvested samples had no greater percentage of lighter achenes than hand harvested achenes except for samples harvested with the Gleaner A at 86-2, 86-3 and 86-4; the KEM SP50 at 86-2 and 86-3; the HEGE 125C at 86-2; and the Gleaner N6 at 86-3.

In 1986, at all locations, over 74% of the combine harvested achenes had their persistent calyx intact. With the exception of all combine harvested samples at 86-1 and samples harvested by the HEGE 125C at 86-3 and KEM SP50 at 86-5, fewer lighter brown achenes were found in the combine harvested samples than in hand harvested achenes. Among lighter achenes there were no differences among combines in the percent of black or white achenes from locations 86-1, 86-2, and 86-4 (Table 6). However at locations 86-3 and 86-5, there were more black achenes among achenes harvested by the Gleaner A than by the KEM SP50. There was no difference in the percent of heavier brown achenes found in samples harvested at 86-1 and 86-5. At locations 86-2, 86-3, and 86-4, there was a greater percentage of the brown heavier achenes in the Gleaner A harvested samples than in the hand harvested samples. This was also true for samples harvested by the HEGE 125C at 86-2 and KEM SP50, and Gleaner N6 at 86-3. The type of machine used did not influence the number of heavier achenes that had their persistent calyx removed by combine harvesting at locations 86-1, 86-3, and 86-5.

<u>Rate of germination</u>. The use of categorical modeling procedure's Chi Square comparisons among the treatments at two locations, 86-4 and 86-5 detected, no significant differences at the 0.95 probability level in the rate of germination for the achenes harvested. After 3 days, the germination of achenes harvested from locations 86-4 and 86-5 was variable. However, after 6 days, $84.6 \pm SE 0.4$ % and $84.0 \pm SE 6$ % respectively of these achenes that germinated during the 15 day period had done so (Figure 1). From 6 through 15 days, achenes harvested from locations 86-4 and 86-5 germinated at the same rate. Thus, alteration of the physical character of the achenes by combine harvesting did not translate directly into an enhanced rate of germination of those achenes that did germinate.

<u>Final germination</u>. Although the rate of germination was not affected by combine harvesting, the final germination of one or more of the six seed classifications was significantly altered. As in 1985, the germination of achenes harvested by hand in 1986 varied substantially from 0.5 to 45%. In 1986, with few exceptions, the germination of brown achenes were uneffected by combine harvesting (Table 7). Lighter black achenes were also uneffected by combine

harvesting with the exceptions that those harvested with the Gleaner A and HEGE 125C from 86-2 had enhanced germination and those harvested with the Gleaner A and Gleaner C2 at 86-5 had lower germination. At no time did combine harvesting depress the germination of heavier black achenes and it frequently enhanced their germination. Also, at all locations except 86-5, combine harvested white seed had from 43 to 99.5% germination enhancement. Lighter white seed harvested by the Gleaner C2 and heavier white seed harvested by the KEM SP50 and Gleaner A at 86-5 had less germination than hand harvested achenes because many of them were damaged by the threshing process.

Based on the frequency and germination of each of the achene classes, there was no substantial net effect of combine harvesting on germination of the seed population sampled at 3 of the 5 locations harvested in 1986 (Table 8). Although the effect of combine harvesting on individual classes of seed was often dramatic, with the exceptions of locations 86-2 and 86-5, too few of the total achenes were altered to significantly change the germination of the population.

Bin sampling was an effective way to draw inference to achenes exiting these commercial combines since approximately 50% of the achenes entering the headers of the Gleaner A, Gleaner N6, and SNH TR85 entered the grain bin (Table 9). Although the plot combines delivered fewer than 50% of the achenes entering the combine to their grain bin, the percentage was high enough to draw inference to the whole population.

For the popular wild buckwheat herbicide treatments evaluated, the EK values did not vary widely as Gressel had speculated for herbicide treatments in general (9). All treatments evaluated reduced achene

production from 91 to 99.5% (Table 10). With bromoxynil + MCPA at 280 + 280 g/ha EK varied from 93.3 to 96.6%. EK with chlorsulfuron varied from 90.8 to 99.5% and EK from picloram + 2,4,D at 26 + 240 g/ha varied only from 95.0 to 96.7%.

Although the range of EK values is narrow, the SL value to integrate with them in Gressel's model (8) is broad and uncertain. Wild buckwheat achenes with complete ovary wall removal would have a SL of 1 to 2 years. The work of Chepil also (4) suggests a SL of 1 to 2 years. In situations of late harvest or harvest of windrowed grain, wild buckwheat achenes dehiscing before passing through the combine should react like hand harvested achenes. These unscarified achenes should have a wide range of SL values as indicated by the first year germination of hand harvested achenes. A range of SL values 3 to 5 years is also possible as suggested by many of the locations and harvest methods used, as well as the work of Forsberg and Best (6). However, the affects of harvesting method and location on first year germination of achenes makes prediction of the upper limits of SL very speculative. As EK values increase, the determination of this upper limit of SL is less important. For example, if the highest EK observed (99.5%) is used and SL is decreased from 5 to 1 year, the model predicts that the time necessary for resistance to evolve would be reduced from 5 to 3 years, as contrasted with a reduction of from 20 to 8 years if the lowest EK value observed (91.4%) is used. The average EK's of chlorsulfuron at 26 g/ha, bromoxynil + MCPA at 280 + 280 g/ha, chlorsulfuron at 23 g/ha, and picloram at 2,4,D at 26 + 420 g/ha are 94, 95, 96, and 96%, respectively. Although the impact of SL is still pronounced at these EK's, SL impact is still greatly reduced even when

EK is only increased from 94 to 96%. This is illustrated using Gressel's estimates for the other parameters and EK's of 94, 95, and 96% and decreasing SL from 5 to 1 year. Under these assumptions, the time required for the evolution of resistance should be reduced from 15 to 7, 14 to 6 and 11 to 5 years for EK's of 94, 95 and 96%, respectively. These high EK values of the herbicides tested, coupled with any reduction in SL, suggest that herbicide resistance may appear much more quickly than has been observed in the past.

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Table 1. Combine settings.

	Cyli	inder Speed		
Combine	Peripheral	Shaft	Cy Bar passes/sec ^a	vlinder Concave Clearance
	(m/sec)	(revs/min)		(mm)
Gleaner C2	33	1275	170	6.4
Gleaner A	32	1250	167	6.4
Gleaner N6	33	980	131	6.4
SNH TR85	26	1150	115	3.2
HEGE 125C	24	1500	150	6.4
KEM SP50	21	1050	140	6.4

^aBars per cylinder x revolutions per second.

<u>Table 2</u>. Effect of harvesting method on persistent calyx and/or ovary wall removal and germination of wild buckwheat achenes collected from location 85-1 in 1985^a.

		Frequenc	су		Ger	Germinatior				
Harvesting method	Brown ^b	Black	White	a de la compañía de l	Brown	Black	White			
	وند وند وند وند وند وند		نه بنند ونه روه سه سه ونه و	- (%) -	ون جند چه چه چه چه چه ه					
Gleaner A	74 b	26 b	0 c		0.1 d	5 cd				
HEGE 125C	70 b	28 b	2 b		6 bc	11 b	55 a			
Hand	100 a	0 c	0° C		59 a					

^aObservations followed by the same letter are not statistically different at the 5% level according to multiple T tests of the least square means.

^bBrown achenes have an intact brown calyx, black achenes have no calyx, and white true seeds have no calyx or ovary wall.

<u>Table 3</u>. Unreplicated observations of the percent of lighter and heavier achenes with their persistent calyx and/or ovary wall removed, collected with various combines, each at a different location in 1985.

Harvesting		Light	er weight	seed	Heavier weight seed				
method	Location	Brown ^a	Black	White	Brown	Black	White		
				(१	5)				
HEGE 125C		76	24	0.10	65	30	5		
KEM SP50		80	20	0.0	52	46	3		
Gleaner C2		73	27	0.0	77	19	4		
SNH TR85		22	77	0.4	27	61	12		
Hand Harvested	all	100	0	0.0	100	0	0		

^aBrown achenes have an intact brown calyx, black achenes have no calyx, and white true seeds have no calyx or ovary wall.

		Achene Seed Weight and Color ^b											
			Lighter			Heavier							
Location	Combine	Brown	Black	White	Brown	Black	White						
		الله جه مور بوری و به به به به به به		(8)								
85-2	Gleaner A	31 bcd	3 fg		38 bc	33 bcd	4 fg						
	HEGE 125C	17 cde	12 defg		34 bcd	50 b	25 bcde						
	KEM SP50	36 bc	15 cdef		27 bcde	48 b	94 a						
	Gleaner C2	3 fg	<l g<="" td=""><td></td><td>2 fg</td><td>8 efg</td><td>2 fg</td></l>		2 fg	8 efg	2 fg						
	Hand Harvested	4 fg			l fg								
85-3	Gleaner A	7с	3 c	17 c	27 b	41 a	62 a						
	Hand Harvested	24 b			27 b								

Table 4	Germination	of	achenes	harvested	from	locations	85-2.	85-3	and	85-4
TUDIC 4.	OCTUTUCTOU	OL.	acticited	TIGT VCDCCG		TOCACTORD	05 21	05 5	una	05 -

		Achene Seed Weight and Color										
			Lighter		Heavier							
Location	Combine	Brown	Black	White	Brown	Black	White					
				(%)							
85-4	SNH TR85	10 b	10 b		38 c	35 c	61 a					
	Hand Harvested	33 c			33 c							

^aObservations within each location followed by the same letter are not statistically

different at the 5% level according to multiple T tests of the least square means.

^bBrown achenes have an intact brown calyx, black achenes have no calyx, and white true seeds have no calyx or ovary wall.

	Lighter				Heavier			
Location	Brown ^a	Black	White	Brown	Black	White		
			(m	g)				
86-1	4.3 to 3.9	4.6 to 2.0	3.1 to 2.7	5.5 to 4.9	4.9 to 3.9	3.7 to 3.2		
86-2	4.0 to 3.6	4.3 to 2.9	5.9 to 2.5	5.7 to 5.2	4.8 to 4.6	5.2 to 1.8		
86-3	4.2 to 3.2	3.5 to 2.7	4.8 to 1.7	5.6 to 4.8	4.6 to 4.3	3.5 to 3.3		
86-4	4.8 to 4.6	3.5 to 3.3	3.4 to 2.0	5.9 to 5.3	5.1 to 4.5	3.9 to 3.3		
86–5	5.1 to 4.7	3.7 to 3.5	3.9 to 3.5	6.7 to 6.5	5.4 to 5.2	4.2 to 4.1		

Table 5. Effect of location on achene weight ranges with 95% confidence intervals.

^aBrown achenes have an intact brown calyx, black achenes have no calyx, and white true seeds have no calyx or ovary wall.

<u>Table 6</u>. Effects of combine harvesting on the percentage of achenes with and without an ovary wall from light and heavy weight achenes harvested from 5 wheat fields in 1986^{a} .

	Lighter				Heavier			
Harvesting	Brown	^b Black	White	Total ^c	Brown	Black	White	Total
		0 an an an an an an an		(१	5)			
				_ (Locati	on 86-1)			
Gleaner A	65a	17a	0.la	82ab	10a	5a	3a	18ab
Hege 125C	79a	5a	0.la	84ab	12a	4a	0.2b	16ab
KEM SP50	59a	8a	0.2a	67b	29a	3a	0.3b	33a
SNH TR85	63a	lla	la "	75ab	17a	7a	lb	25ab
Gleaner N6	81a	9a	0.la	90a	7a	3a	0.1b	10b
Hand	90a	0.0b	0.0b	90ab	10a	0b	0.0c	10ab
				_ (Locati	on 86-2)	<u> </u>		
Gleaner A	23c	6a	0.4a	30c	51ab	14a	ба	70a
Hege 125C	26c	3a	0.3a	29c	54a	13a	3b	7la
KEM SP50	50b	5a	0.2a	55b	41bc	3b	lc	45b
Hand	71a	0.0b	0.0b	71a	29c	0.0c	0.0d	29c
				_ (Locati	.on 86-3)			
Gleaner A	27bc	4a	0.la	32bc	58ab	10a	lb	69ab
Hege 125C	47ab	3ab	0.la	50ab	42bc	6a	lb	50bc
KEM SP50	17c	lc	0.0b	18c	72a	7a	3a	82a
Gleaner N6	25bc	2bc	0.la	27c	63ab	9a	lb	73a
Hand	67a	0.0d	0.0b	67a	33c	0.0b	0.0c	33c

Table 6. Effects of combine harvesting on the percentage of achenes with and without an ovary wall from light and heavy weight achenes harvested from 5 wheat fields in 1986, cont'd.

	Lighter				Heavier			
Harvesting								
method	Brown	Black	White	Total	Brown	Black	White	Total
				(१	5)			
				_ (Locati	lon 86-4)	-		
Gleaner A	27c	6a	2a	35b	49a	lla	5a	65a
Hege 125C	51b	8a	la	59a	29b	9b	3a	41b
KEM SP50	50b	5a	0.4a	55a	38ab	5c	2a	45b
Hand	66a	0.0b	0.0b	66a	34b	0.0d	0.0b	34b
				_ (Locati	ion 86-5)			
Gleaner A	46c	9a	lb	55a	34a	8a	3.0b	45a
Hege 125C	50bc	6ab	lb	57a	32a	9a	3b	44a
KEM SP50	66a	5b	0.1b	72a	20a	7a	lb	28a
Gleaner C2	47c	9a	3a	58a	28a	4a	10a	42a
Hand	70a	0.0c	0.0c	70a	30a	0.0b	0.0c	30a

^aObservations followed by the same letter are not significantly different at the 5% level according to multiple T tests of the least square means. Comparisons should only be made within the same column and within the same location.

^bBrown achenes have an intact brown calyx, black achenes have no calyx, and white true seeds have no calyx or ovary wall.

^cFigures may not add to 100 due to rounding.

<u>Table 7</u>. Effects of persistent calyx and/or ovary wall removal during the harvesting process on germination after 15 days of lighter and heavier wild buckwheat achenes harvested from 5 fields in 1986^{a} .

		Lighter			Heavier	
Harvesting		<u> </u>				
method	Brown	Black	White	Brown	Black	White
			(!	8)		یہ جہ جہ کا جہ جہ ان ان جہ کا جا
			(Locatio	on 86-1)		
Gleaner A	1.6a	3a	68ac*	2a	lla	46b*
HEGE 125C	1.5ab	lab	71a*	lab	7a	45b*
KEM SP50	2.2ab	2a	45abc*	3a	9a	100a*
SNH TR85	1.2ab	2a	25cd	0.3b	2b	18b
Gleaner N6	0.5b	0.3b	lbd	0.2b	lb	54b*
Hand ^c	1			0.5		
			(Locatio	on 86-2)		4
Gleaner A	29b*	25b*	40b*	18ab	26c*	40b*
HEGE 125C	37a*	37a*	47b*	23a*	43a*	6la*
KEM SP50	20c	32ab*	81a*	14b	34b*	70a *
Hand	17			12		
			(Locatio	on 86-3)		
Gleaner A	10b	11b		8b*	18b	50b*
HEGE 125C	17a*	27a*	79a*	8b*	27a*	55ab*
KEM SP50	13ab	9Ъ		18a	28a*	62a*

<u>Table 7</u>. Effects of persistent calyx and/or ovary wall removal during the harvesting process on germination after 15 days of lighter and heavier wild buckwheat achenes harvested from 5 fields in 1986, cont'd.

	Lighter			Heavier			
Harvesting method	Brown	Black	White	Brown	Black	White	
			(8)			
Gleaner N6	10b	15a	54b*	5b *	22ab	60b*	
Hand	10			18			
	<u></u>		(Locati	on 86-4)			
Gleaner A	19a	20a	44b	10.3ab	24b*	44b*	
HEGE 125C	23a	25a	60a	15.4a	30ab*	60a*	
KEM SP50	22a	23a	63a	9 . 7b	32a*	66a*	
Hand	25			8.5			
			(Locati	on 86-5)			
Gleaner A	27b*	5b*	34b	24c	41bc	29c*	
HEGE 125C	39a	17a*	87a*	44a	53a*	80a*	
KEM SP50	41a	9b*	50b	33b	51ab*	28c*	

<u>Table 7</u>. Effects of persistent calyx and/or ovary wall removal during the harvesting process on germination after 15 days of lighter and heavier wild buckwheat achenes harvested from 5 fields in 1986, cont'd.

	Lighter				Heavier		
Harvesting							
method	Brown	Black	White	Brown	Black	White	
	و المراجع الم		(۶	8)	ه چه زه زه زه که که زه که ک		
Gleaner C2	24b*	15ab*	26a*	30bc	45b	45b	
Hand	45			43			

^aLetter comparisons only valid within a seed color, weight and location. Those comparisons followed by the same letter are not statistically significant at the 5% level according to multiple T test analysis of the least square means.

^bBrown achenes have an intact brown calyx, black achenes have no calyx removed by combine harvest, white true seeds have no calyx or ovary walls.

^cComparisons with hand harvested seed are only valid within each location and seed weight. Means followed by an * are significantly different from hand harvested means at the 5% level according to multiple T test analysis of the least square means.

	Loc 86-1	Loc 86-2	Loc 86-3	Loc 86-4	Loc 86-5
			(%)		
Gleaner A	5	27	17	22	28
HEGE 125C	5	36	19	25	43
KEM SP50	7	22	18	25	40
SNH TR 85	2				
Gleaner N6	1		14		
Gleaner C2					29
Hand harvest	2	19	17	24	45
LSD (0.05)	NSD	2	NSD	NSD	9

Table 8. Germination of achenes that entered the combine bin in 1986, calculated from the frequency and germination of each seed classification.

Table 9. Percent of wild buckwheat achenes introduced into the combine header that entered the grain bin.

Combine Type	(१)
Gleaner A	54.9a
Gleaner N6	48.8ab
SNH TR 85	39,3bc
HEGE 125C	32.8c
KEM SP50	25.1c
LSD (0.05)	14.3

.

<u>Table 10</u>. Effective kill (EK) obtained by applications of selected herbicides to wild buckwheat at 12 Oklahoma locations from 1981 to 1986.

Treatment	Rate	Year	EK ^a
	(g/ha)		(%)
Bromoxinil + MCPA	280 + 280	1985	93.3
Bromoxinil + MCPA	280 + 280	1986	96.6
Chlorsulfuron	23	1984	98.6
Chlorsulfuron	23	1986	99.5
Chlorsulfuron	23	1986	90.8
Chlorsulfuron	26	1986	96.6
Chlorsulfuron	26	1986	91.4
Picloram + 2,4,D	26 + 420	1985	96.2
Picloram + 2,4,D	26 + 420	1986	95.0
Picloram + 2,4,D	26 + 420	1986	96.0
Picloram + 2,4,D	26 + 420	1986	98.6
Picloram + 2,4,D	26 + 420	1986	96.6

^a Each observation is a mean of 4 replications.



Figure 1. The rate of germination of wild buckwheat achenes that germinated in 15 days averaged over harvest method, achene weight and color. Dashed line represents location 86-5 and solid line represents location 86-4. Vertical bars represent \pm the standard error for each time interval.

PART II

EFFECT OF COMBINE HARVESTING ON GERMINATION OF CURLY DOCK (<u>RUMEX CRISPUS</u>), VENICE MALLOW (<u>HIBISCUS TRIONUM</u>), AND SLIMLEAF LAMBSQUARTERS (<u>CHENOPODIUM LEPTOPHYLLUM</u>) Effect of combine harvesting on germination of curly dock (<u>Rumex</u> <u>crispus</u>), Venice mallow (<u>Hibiscus trionum</u>), and slimleaf

lambsquarters (<u>Chenopodium leptophyllum</u>)⁺ RANDALL S. CURRIE AND THOMAS F. PEEPER²

<u>ABSTRACT</u>. Seed of three weed species collected from the grain bins of combines while harvesting standing hard red winter wheat had higher germination than hand harvested seed. Combine harvesting increased germination of curly dock (<u>Rumex crispus</u> L. #³ RUMCR) from 4 to 34% compared to hand harvested seed. Curly dock seeds harvested with a commercial type combine had higher germination than those harvested with a small plot combine. Harvesting slimleaf lambsquarters (<u>Chenopodium leptophyllum</u> (Moq) Nutt. ex S. Wats # CHELE) and Venice mallow (<u>Hibiscus trionum</u> L. # HIBTR) seeds with a commercial style combine also enhanced germination compared to hand harvested seed. However, harvesting with a small plot combine did not consistently increase Venice mallow seed germination. Germination enhancement due

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Letters following this symbol are a WSSA-approved computer code from Composite List of Weeds, Weed Sci. 32, Suppl. 2. Available from WSSA, 309 West Clark St., Champaign, IL 61820.

to mechanical crop harvesting could increase the rate at which herbicide resistant biotypes appear.

Additional index words. Herbicide resistance, dormancy, scarification, RUMCR, HIBTR, CHELE.

INTRODUCTION

Weed seed dormancy has a major impact on the appearance of herbicide resistance (13, 14, 15, 16) and dormancy can be dramatically altered by pericarp damage (10, 18, 21, 28, 29). Pericarp damage to wheat during combine harvesting is well documented (2, 3, 4, 17, 22, 23, 25). If weed seeds were also scarified by combine harvesting and thus germinated sooner, crop harvesting procedures would accelerate the emergence of herbicide resistant biotypes by decreasing the buffering effect of all previous year's seed production.

Enhanced herbicide resistance has been widely reported in species from the Polygonaceae, Malvaceae and Chenopodiaceae families (12). These reports include increased triazine tolerance in two species of Malvaceae, velvetleaf (<u>Abutilon theophrasti</u> L.) and cotton (<u>Gossypium</u> <u>hirsutum</u> L.) (1, 8, 33). Also, three species of Polygonaceae, and four species of Chenopodiaceae with enhanced triazine resistance have been reported in corn (Zea mays L.) culture (12).

Species from these families often found in Oklahoma wheat fields include curly dock, Venice mallow and slimleaf lambsquarters. Seed of these species varies substantially in size and shape and thus should interact differently with threshing mechanisms of grain combines. The 1 to 1.5 mg, 1 to 2 mm curly dock achenes (hereafter referred to as seeds) are enclosed in a 3 to 4 mm winged indehisent dry fruits.

Venice mallow seeds are 2 to 3 mm ovoids weighing 3 to 4 mg. Slimleaf lambsquarters achenes (hereafter referred to as seeds) are 1 mm flattened spheroids which weigh approximately 0.5 mg.

Porter and Koos (30) observed that the pericarps of Polygonaceae and Chenopodiaceae achenes are often removed during commercial crop seed cleaning processes and they speculated that pericarp removal could also occur during grain threshing. They found that pericarp removal did not reduce germination of newly ripened seed but that the viability of seeds without pericarps decreased substantially with 3 months storage. Subsequently, Kanipe (20) reported that curly dock seeds with and without fractured pericarps due to mechanical harvesting had equal germination of 84 ± 1 % while seed without pericarps had only 63% germination. The age of the seed without pericarps was not stated. The work of Porter and Kanipe, like most research on harvesting effects on weed seed coats, dealt primarily with defining the minimum seed injury necessary to classify weed seeds as inert matter in crop seed (6, 7, 11, 20, 30). Thus, efforts have been concentrated on obviously visually damaged seed rather than the entire seed population and do not consider how such damage might have occurred.

Investigations of the effects of combine harvesting on the pericarp of wheat have shown that complex interactions of cylinder speed, concave clearance, material flow rate, machine design and other factors affect the degree of pericarp damage (2, 3, 4, 17, 23, 25). However, the influence of mechanical harvesting on weed seeds is poorly understood.

Species from the Polygonaceae, Malvaceae, and Chenopodiaceae commonly have long periods of dormancy and/or complex germination

requirements that can be altered by pericarp injury (18, 19, 21, 26, 27, 28, 29). Curly dock achenes don't enter dormancy, but burial excludes the light necessary for germination (5). Such burial has been reported to preserve the viability of curly dock achenes in soil for 20 to 100 years (24, 27, 32). However, scarification removes the light requirement for germination (18, 31). Acid or mechanical scarification has also been shown to increase germination of Venice mallow and many <u>Chenopodium</u> spp. (10, 21, 28). However, the germination requirements of slimleaf lambsquarters have not been reported. The objective of this research was to determine whether combine harvesting weed seed as hard red winter wheat is being harvested affects germination of curly dock, Venice mallow and slimleaf lambsquarters seed populations.

MATERIALS AND METHODS

During the summers of 1985 and 1986, seeds of curly dock, Venice mallow, and slimleaf lambsquarters were collected as standing hard red winter wheat was being harvested with a commercial and/or two small plot self-propelled grain harvesting combines. The primary threshing mechanism of the commercial combine, a Gleaner A^4 , is a conventional rasp bar cylinder mounted in the feeder housing with a closed bottom concave. It has a straw walker-double sieve system for grain separation and tailings are returned to the front of the cylinder. This basic design has been commonly used in production machines for over 30 years. Seed of all three species was harvested with this combine.

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Unlike the Gleaner A, the KEM SP50⁵ and HEGE 125C⁶ plot combines have conventional rasp bar cylinder threshing mechanisms located behind the feeder housings of the header. The KEM SP50 was modified to feed the cylinder with a feeder rake chain. The cylinder of the HEGE 125C is fed by a flat conveyor belt. Both have open bottom concaves, a beater behind the cylinder and a double sieve separation system with no straw walker or tailings return. The KEM SP50 has plastic sieve components whereas the other machines have metal sieve components. These combines were set and operated in accordance with manufacturers specifications (Table 1) and operated at near full capacity while the crop containing the weed seed was harvested. Harvesting was in the late afternoon in standing hard red winter wheat that had been dead ripe for 2 to 3 weeks.

While these machines were operating, four 4 to 5 kg samples of wheat and weed seed were collected from their grain bins. Seed collected from the bins was considered representative of the total seed population because a substantial fraction of small weed seeds are frequently collected with the crop (9).

In 1985, curly dock seed was harvested at the Agronomy Research Station, Stillwater, OK., with the Gleaner A and KEM SP50 combines and with the Gleaner A at the Lake Carl Blackwell Research Area, Orlando, OK. In 1986, curly dock seed was harvested with all combines at the Agronomy Research Station, Stillwater, OK., and in a field 5 km west of

⁵Kincaid Equipment Manufacturing, Haven, KS 67543

HEGE Saazuchtmaschinen., Domaene Hohebuch D-7112, Waldenburg/Wertt, West Germany

Stillwater. Due to the limited supply of standing wheat at Stillwater in 1986, it was necessary to reduce the swath width to 90% of the header width. Venice mallow seed was harvested both years with the Gleaner A and KEM SP50 combines at the Agronomy Research Station, Stillwater, OK. Slimleaf lambsquarters seeds were harvested with the Gleaner A from the Orlando and Stillwater locations both years. In all cases, seed of each species was collected from four areas of each field with each combine and by hand harvesting.

Weed seed was separated from the grain samples with 2 or 3 mm hand held screens. After separation, the Venice mallow seeds were stored dry in paper envelopes in the dark at 25 ± 3 °C for 2 months before being placed in a germinator. The lambsquarters and curly dock seeds were stored in the same manner for 6 months after harvest. Four 25 seed subsamples from each of four field samples were placed in 9 cm petri dishes on 1 g of absorbent paper toweling hydrated with 10 ml of distilled water. The petri dishes were arranged in a germinator in a randomized complete block design and maintained in the dark at 30°C with near 100% relative humidity for 15 days. Every 3 days germinating seeds were counted and removed.

The experimental design used was a randomized complete block with four replications. In each experiment, the cumulative germination for each successive 3 day interval was analyzed separately and means were compared using LSDs at the 5% level.

RESULTS AND DISCUSSION

Gleaner A harvested curly dock achenes had from 26 to 31% and 15 to 23% germination after 15 days in 1985 and 1986 respectively compared

to corresponding hand harvested seeds with 1 to 7% and 1.5 to 4.2% germination (Fig. 1, 2). The curly dock achenes were briefly exposed to light every 3 days during counting. However, such exposure was apparently insufficient illumination to induce high germination of hand harvested seed, since all hand harvested seed from both locations in both years had less than 3% germination. Of the curly dock seeds that germinated, the rate of germination was rapid. Over half of the germination occurred within 6 days and very little germination occurred after 9 days.

Unlike achenes harvested with the Gleaner A, plot combine harvested seeds did not consistently have higher germination than hand harvested seeds. Harvesting with the plot combines at the location west of Stillwater did not enhance germination whereas seeds harvested at Stillwater with the KEM SP50 and HEGE 125C had 13 and 21% germination, respectively, compared to 1.5% of hand harvested seeds. The total amount of material fed through the machine was less at Stillwater than west of Stillwater which could have resulted in more impacts of seeds with metal surfaces which may explain the higher over all germination of curly dock seeds harvested at Stillwater in 1986. Although the periferal cylinder speed of these two combines was very similar, with less non grain material flowing through the combines at Stillwater the slightly higher periferal cylinder speed of the HEGE 125C could have damaged the seed coat more. Conversely west of Stillwater the greater amount of plant material may have masked the differences between these machines.

As with curly dock, over half of the germination of Venice mallow seeds occurred within 6 days and few germinated after 9 days (Fig. 3,

4). Those harvested with the Gleaner A and the HEGE 125C in 1985 had over 40% germination whereas no hand harvested seed germinated. In 1986 the effect of combine harvesting on germination was much less than in 1985. This may be due to less total material being fed through the combine in 1985 as was the case with curly dock seed harvested at Stillwater in 1986. Even with very low Venice mallow germination in 1986, the effect of combine harvesting was still significant. However, no difference in germination between seeds harvested by the Gleaner A and HEGE 125C was observed until the 15th day.

Slimleaf lambsquarters seed harvested with the Gleaner A in 1985 and 1986 had 9 to 12% germination respectively. None of the hand harvested slimleaf lambsquarters seed collected either year germinated (Figure 5).

The injury to curly dock and slimleaf lambsquarters was visible without magnification but required 20X magnification for accurate description. Many curly dock seeds had one of three seams of the fused ovary walls of the achene ruptured. Very similar damage is visible in Kanipe's illustrations (20). Small irregularly shaped patches of the seed coats of slimleaf lambsquarters had been removed. The illustrations of Fleischman also depict this type of damage in mechanically harvested seeds of other <u>Chenopodium</u> spp. (11). This type of damage would seem consistent with the damage combines cause to crop seeds which are reportedly caused by high velocity impact of seeds with metal.

Without magnification combine harvested Venice mallow seeds appeared undamaged. With 20X magnification, scraping breaks in the thin regularly shaped waxy layer of the seed coat were visible. There

was no physical abrasion of the seed coat itself apparent with 64X magnification.

Although the magnitude of the influence of combine harvesting varied with weed species, location and machine, combine harvesting increased weed seed germination in all situations but one. These results indicate that populations of weed seeds of various sizes that pass through a grain combine can be expected to germinate faster than naturally dehiscing seed. The work of Gressel (13) shows that even minor decreases in the length of weed seed dormancy can dramatically increase the rate of appearance of herbicide resistance. Thus, direct combine harvesting of wheat and weed seed could increase the rate of appearance of herbicide resistant species.

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Table 1. Combine settings.

Combine	Peripheral	Rev/min	Bar passes/sec ^ª	Concave opening
	(m/sec)			(mm)
Gleaner A	32	1250	167	6.4
HEGE 125C	24	1500	150	6.4
KEM SP50	21	1050	140	6.4

^aBars per cylinder x revolutions per second.







Figure 2. The cummulative germination of curly dock seed harvested at two locations in 1986 with a Gleaner A (∞), HEGE 125C (\blacktriangle), or KEM SP50 combines (\bigcirc), or by hand (\blacksquare). Symbols followed by the same letter within the same time interval are not different (P < 0.05).



Figure 3. The cummulative germination of Venice mallow seed harvested by the Gleaner A (\bullet) or HEGE 125C combines (\blacktriangle), or by hand (\blacksquare) in 1985. Symbols followed by the same letter within the same time interval are not different (P < 0.05).



Figure 4. The cummulative germination of Venice mallow seed harvested by the Gleaner A ($\stackrel{(\bullet)}{\bullet}$) or HEGE 125C combines ($\stackrel{(}{\bullet}$), or by hand (\blacksquare) in 1986. Symbols followed by the same letter within the same time interval are not different (P < 0.05).



Figure 5. The cummulative germination of slimleaf lambsquarters harvested with a Gleaner A (\bullet) or by hand (\blacksquare) in 1985 (——) and 1986 (---). Symbols followed by the same letter within the same time interval are not different (P < 0.05).

VITA

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Master of Science

Thesis: EFFECTS OF COMBINE CROP HARVESTING ON WEED SEEDS AND SUBSEQUENT POTENTIAL FOR EMERGENCE OF HERBICIDE RESISTANT BIOTYPES

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