

Effect of Biennial Wormwood (*Artemisia biennis*) Interference on Sunflower Yield and Seed Quality

Derek W. Lewis, Andrea Cavalieri, and Robert H. Gulden*

Biennial wormwood has been increasing in Manitoba, Canada, but information is limited on the potential of biennial wormwood to reduce crop yields. Field experiments were conducted over 5 site-yr throughout southern Manitoba in 2010 and 2011 to determine the effect of biennial wormwood density and relative time of biennial wormwood seedling recruitment on sunflower growth, development, yield, and seed quality. Biennial wormwood was broadcast on the soil surface at six densities into sunflowers planted in 75-cm rows, either at the same time the sunflower crop was planted (early weed seedling recruitment) or when the sunflowers were at about the four-leaf stage (late weed seedling recruitment). When biennial wormwood emerged at about the same time as the sunflowers, yield was reduced by up to 46%. Early-recruiting biennial wormwood had minimal effect on sunflower growth and development, but sunflower achene size and individual achene weight were reduced, even when no effect on sunflower yield was observed. Biennial wormwood plants that recruited after the four-leaf stage of the sunflower crop had no effect on sunflower yield or seed quality.

Nomenclature: Biennial wormwood, *Artemisia biennis* Willd., ARTBI; sunflower, *Helianthus annuus* L.

Key words: Crop–weed interference, plant competition, rectangular hyperbola, yield loss.

Sunflower is a native species of North America and one of the four major crops grown for edible oil in the world (Putt 1977). In Canada, the production of sunflower is confined to the province of Manitoba (40,500 ha in 2012), where much of the crop is grown for oil production (70%), with a smaller proportion of confection sunflowers (30%) (Anonymous 2014). Weeds can cause significant sunflower yield reductions, especially when interference occurs during the early stages of crop development. Lewis and Gulden (2014) showed that sunflower yield was reduced by up to 76% when kochia [*Kochia scoparia* (L.) Schrad.] emerged at about the same time as the sunflower crop. Sunflower yield loss ranged from 35 to 54% under competition with wild oat (*Avena fatua* L.) (Chubb and Friesen 1985), hemp sesbania [*Sesbania herbacea* (P. Mill.) McVaugh] (Woon 1987), sunflower broomrape (*Orobancha cumana* Wallr.) (Grenz et al. 2008), and mixed weed species (Chubb 1975; Reddy et al. 2008).

Crop yield loss in response to increasing weed density is often best described using the nonlinear rectangular hyperbola function (Cousens 1985). Parameters derived from this and other yield loss

models are often used to estimate action or economic thresholds or the density of weeds at which action is taken to most economically manage weeds. Limited information about weed interference with sunflower is available. Nevertheless, action and economic thresholds for weed management in sunflower have been determined for kochia (Gulden and Lewis 2014), false caraway (*Ridolfia segetum* Moris) (Caranza et al. 1995), wild mustard (*Sinapis arvensis* L.), lambsquarters (*Chenopodium album* L.), and greater ammi (false Queen Anne's lace) (*Ammi majus* L.) (Onofri and Tei 1994). In these studies, action and economic thresholds for weed management in sunflower tended to be low and ranged from < 1 plant m^{-2} to about 5 to 6 plants m^{-2} , with much overlap in the ranges among these weed species.

Biennial wormwood has been identified as one of the 10 most problematic weeds in sunflower fields in weed surveys conducted from Manitoba to Texas (Anonymous 2013), and biennial wormwood has been increasing on the Great Plains of North America (Fronning and Kegode 2004). In North Dakota field surveys conducted in 1978 and 1979, biennial wormwood was not found in any of the 2,819 fields surveyed; however, by 2010, biennial wormwood was found in 10 to 15% of 111 fields surveyed (96 in North Dakota and 15 in Minnesota) and ranked among the top 10 most prevalent weed species (Kandel 2011). In Manitoba, biennial wormwood was found in 1.1% of 631 fields sown to annual crops

DOI: 10.1614/WS-D-15-00076.1

* Graduate Student, Research Associate, and Associate Professor, Department of Plant Science, University of Manitoba, Winnipeg, MB, Canada, R3T 2N2. Corresponding author's E-mail: Rob.Gulden@ad.umanitoba.ca

surveyed in 2002 and was ranked the 43rd most prevalent weed. However, in some areas in Manitoba, biennial wormwood was found at greater frequencies (Leeson et al. 2002). Around Killarney, Manitoba, for example, biennial wormwood was found in 13.6% of fields surveyed and ranked 15th most prevalent weed in that area, and around Pilot Mound, Manitoba, biennial wormwood was found in 11.1% of the fields surveyed and ranked 12th most prevalent weed in that area. In a survey of South Dakota soybean fields, biennial wormwood was found in 92% of the fields surveyed (Zollinger and Ciernia 2007).

The ability of biennial wormwood to proliferate in both conventional and reduced-tillage systems, an extended seedling recruitment period and a preference for moist soil conditions have been identified as potential reasons for the increase in biennial wormwood infestations (Kegode and Darbyshire 2013). Biennial wormwood also is easily misidentified as common ragweed (*Ambrosia artemisiifolia* L.), which also may have contributed to its success through the erroneous application of ineffective herbicides.

A search of the literature reveals that little information is available about the competitive ability of biennial wormwood in field crops. In one study, biennial wormwood interference at densities of 10 plants m^{-2} reduced soybean yield by 44% in North Dakota (Nelson and Kegode 2000). Interference of biennial wormwood can also result in indirect yield losses at harvest, because biennial wormwood matures late in the growing season and farmers may completely avoid harvesting densely infested areas. In a replacement series greenhouse study using soybean and biennial wormwood, biennial wormwood plants grew taller when grown in combination with soybean compared with when biennial wormwood plants were grown alone, whereas soybean plants were shorter when grown in combination with biennial wormwood compared with when grown alone (Nelson and Kegode 2005). In the same study, biennial wormwood aggressivity increased with increasing nitrogen fertility.

An experiment was conducted in 2010 and 2011 at three locations in southern Manitoba. The objective of this experiment was to determine the effect of biennial wormwood interference on sunflower growth, development, yield, and seed quality. The yield loss data were used to determine an action threshold for biennial wormwood control in sunflowers.

Materials and Methods

Field experiments were conducted in 2010 and 2011 at three locations throughout Manitoba. These included the University of Manitoba Ian N. Morrison Research Farm at Carman, MB (49.50°N, 98.03°W), the University of Manitoba at Winnipeg, MB (49.80°N, 97.15°W) and near the town of Melita, MB (49.27°N, 101.00°W). The soils at Carman and Winnipeg were a Rignold fine sandy loam and a Riverdale silty clay, respectively. At Melita, the soils were a Cameron fine sandy loam in 2010 and a Waskada sandy loam in 2011. The Carman and Melita site-years were established on cereal stubble and the Winnipeg site-year was fallow the previous year. In 2010, a second experiment was established at Carman (designated Carman2) 27 d after the first experiment was planted. Soil nutrient status was determined before seeding each year, and nitrogen and phosphorus fertilizers were applied as recommended (Anonymous 2007). The experimental design was a two-way factorial, laid out as a split-block, randomized complete plot design, with four replicates. The split-block design was easier to manage. Main blocks were the time of biennial wormwood seedling recruitment relative to the sunflower crop. The recruitment of biennial wormwood was targeted at the same time as the sunflowers (early seedling recruitment), or after the four-leaf stage of the sunflowers (late seedling recruitment). The six subplots in the experiment were the different levels of biennial wormwood density. Each subplot was 3 m wide by 4 m long and consisted of four rows of sunflowers planted 75 cm apart.

The initial sample of biennial wormwood seed was obtained by collecting mature plants from field margins and ditches in southern Manitoba in the fall of 2009. The plants were hand threshed, and the material was passed through a 3/64 round hand sieve to obtain the seed sample. Biennial wormwood seeds are extremely small, at about 13,000 seeds g^{-1} (Mahoney and Kegode 2004); therefore, it was not possible to sieve the threshed plant material effectively to obtain a pure seed sample without losing a large amount of seed. The seed material spread over the plots was a combination of seed and other plant residue. A 1.0-g sample of the seed material planted in soil in the greenhouse produced 605 seedlings. The biennial wormwood seeding rates were 1 ×, 5 ×, 10 ×, 20 ×, 40 ×, and a control treatment (0 ×) that was maintained weed free for the entire growing season. In 2010, the 1 × seeding rate of biennial wormwood was 0.2 g seed material m^{-2} ,

Table 1. Summary of sunflower and biennial wormwood planting dates for 5 site-yr of research examining the effect of biennial wormwood interference on sunflower yield, growth, and development in Manitoba.

Site-year	Sunflower	Early wormwood recruitment	Late wormwood recruitment
Carman 2010	May 18	May 19	June 23
Carman2 2010	June 14	June 14	July 8
Melita 2010	June 2	June 9	June 28
Carman 2011	May 19	May 19	June 17
Winnipeg 2011	May 20	May 20	June 20

and in 2011, the $1 \times$ seeding rate was increased to $0.3 \text{ g seed material m}^{-2}$ to achieve greater biennial wormwood densities.

Before planting, a weed-free seed bed was prepared by cultivation or glyphosate application at the rate of $1.33 \text{ kg ae ha}^{-1}$. Sunflower cultivar 6946 (Seeds 2000, P.O. Box 200, 115 North 3rd Street, Breckenridge, MN 56520, USA) was planted with a small plot seeder at a rate of $54,340 \text{ seeds ha}^{-1}$ when field conditions were favorable in the spring (Table 1). After sunflower emergence, grassy weeds were controlled using sethoxydim at 500 g ai ha^{-1} with the required adjuvant at 0.5% (v/v). Broadleaf weeds (except biennial wormwood) were removed by hand.

Sunflower and biennial wormwood data were collected over the course of the growing season. Sunflower plant population was determined by counting all plants in the center two rows of each plot at the eight-leaf stage (about 50 d after seeding). All other measurements were conducted excluding the first and last meter of the plot and the two outer rows. Biennial wormwood density was determined at the same time as the sunflower plant populations by counting the biennial wormwood plants in two 0.25-m^2 quadrats, placed in an area that was representative of the plot. The number of leaves per sunflower plant were counted on five plants per plot at anthesis, and plant height was determined during the seed-filling stage by measuring the height of five randomly selected plants per plot; at the same time, sunflower plant stem diameter was measured on 10 plants (at 30 cm above the soil surface) with a digital caliper. Sunflower head diameter was determined at harvest by measuring the diameter of 10 randomly selected heads per plot.

Seed yield was determined from sunflower heads that were hand harvested at physiological maturity, when bracts on the back of the heads were brown. The heads, collected from 2 m in each of the middle two rows (excluding the first and last meter) in each subplot, were bagged, air dried, and threshed with a stationary small plot combine. Harvested samples were cleaned and weighed, and achene size and

thousand kernel weight (TKW) were determined for each plot using Assess 2.0 Image Analysis Software (The American Phytopathological Society, 3340 Pilot Knob Road, St. Paul, MN 55121, USA), as previously described in Lewis and Gulden (2014). Sunflower yield and TKW were adjusted to 10% gravimetric moisture content.

Daily average temperature and daily precipitation data were obtained from the closest Environment Canada or Manitoba Agriculture Food and Rural Development weather station. At the end of the growing season, average temperature and total precipitation were determined for each month during the growing season.

Statistical analysis was conducted using the PROC NLIN in SAS 9.2 software (SAS Institute, 100 SAS Campus Drive, Cary, NC 27513, USA). Before calculating treatment average, outliers were removed based on studentized residuals (Lund 1975). Initially, site-year were analyzed individually because of variability in biennial wormwood recruitment. The relationship between yield loss and biennial wormwood density was determined using a nonlinear, rectangular hyperbola regression model (Cousens 1985) that is commonly used in weed interference studies (e.g., Blackshaw 1993, Knezevic et al. 1994, Lewis and Gulden 2014). The yield loss relationship is described by Equation 1,

$$Y_L = Id / (1 + Id/A) \quad [1]$$

where Y_L is the percent yield loss relative to the weed-free treatment, I is the percent yield loss per weed as weed density approaches zero, d is weed density, and A is the percent yield loss as weed density approaches infinity. Coefficients of determination for each site-year were determined (Kvalseth 1985). A linear regression approach extracting linear and quadratic components was used when the conformation to the rectangular hyperbola model could not be achieved. A combined analysis was conducted that only included site-year with similar coefficients. The extra sums of squares method (Lindquist et al. 1996) was used to determine which site-year could

Table 2. Average observed weed-free sunflower yields, yield loss parameter estimates (standard errors of the means in parentheses) for the rectangular hyperbola yield loss model, P values and root mean square error (RMSE) for the model, and biennial wormwood plant densities that resulted in 5% sunflower yield loss for increasing densities of early-recruiting biennial wormwood in sunflowers across 5 site-yr in Manitoba.

Site-year	Observed weed-free yield ^a kg ha ⁻¹	Model parameter ^b		P value	RMSE	5% action threshold plants m ⁻²
		<i>I</i>	<i>A</i>			
Carman 2010	2,393 b	NS	NS	0.607	—	—
Carman2 2010	1,639 c	NS	NS	0.123	—	—
Melita 2010	2,263 bc	0.13 (0.08)	46.4 (22.1)	0.006	26.03	44
Carman 2011	2,716 b	0.15 (0.03)	34.3 (3.8)	0.004	23.25	41
Winnipeg 2011	4,415 a	2.41 (1.09)	34.6 (4.2)	0.001	18.56	3
Combined analysis (Melita 2010 and Carman 2011)	—	0.14 (0.05)	39.8 (9.7)	0.001	16.88	40

^a Means followed by different letters are considered significantly different using Fisher's protected least significant difference.

^b *I* is the percent yield loss per weed as weed density approaches zero. *A* represents the asymptote, which is the percent yield loss as weed density approaches infinity. NS indicates that the data did not conform to the rectangular hyperbola yield loss model.

be included in a combined analysis. Action thresholds were determined at the 5% yield loss level as described in Lewis and Gulden (2014). The relationships between sunflower plant height, sunflower stem diameter, the number of leaves per sunflower plant, sunflower head diameter, sunflower achene size, and sunflower TKW and biennial wormwood plant density were determined using the GLM procedure to determine the significance of linear and quadratic components. A mixed-model ANOVA was conducted on sunflower yields in weed-free treatments only among the site-years. Site-year was considered the fixed effect in this analysis, and replicates nested within site-year were considered the random effect. Means were separated using Fisher's protected least significant difference (LSD_{0.05}).

Results and Discussion

All biennial wormwood in this study assumed an annual life cycle. When biennial wormwood plants emerged at about the same time as the sunflowers, the rectangular hyperbola model adequately described sunflower yield loss in 3 of the 5 site-yr (Table 2). At Carman 2010 and Carman2 2010, low overall recruitment (only about 200 biennial wormwood plants m⁻² at the highest planting densities) and inconsistent or complete lack of yield losses among these low biennial wormwood densities (data not shown) contributed to these data not conforming to the rectangular hyperbola nor the linear regression yield loss models. Only 10 × weed seedling density data (817 plants m⁻²) from Carman 2011 conformed to the rectangular hyperbola yield loss model. Biennial wormwood seedling recruitment was greatest in this experiment, and seedling

recruitment densities achieved at 10 × in this experiment were greater than the highest seeding rates in all other experiments. Interestingly, at the two highest biennial wormwood densities of 20 × and 40 × (1,362 and 1,732 plants m⁻², respectively), the percent yield loss in sunflower was less than at the 10 × seeding rate. These results suggest that intraspecific competition and likely self-thinning in biennial wormwood were sufficient to reduce interference with the sunflower crop.

For the early-recruiting biennial wormwood, the values for model parameter *A* ranged from about 34 to 46% among the conforming site-years, whereas the values for model parameter *I* ranged from 0.13 to 2.41% among the conforming site-years (Table 2). Based on the extra sum of squares (Lindquist et al. 1996), the model parameters for Carman 2011 and Melita 2010 were not different from each other, whereas both *I* and *A* parameter estimates for Winnipeg 2011 were significantly different from those of the other two experiments. The shape of the curve and, specifically, the biennial wormwood density at which the yield loss relationship became asymptotic (as illustrated by the different *I* parameters) likely contributed to this. Weed-free sunflower yield also was significantly greater at Winnipeg 2011.

Interestingly, the *I* parameter, which represents the percent yield loss caused by individual biennial wormwood plants at low densities, was greatest at Winnipeg 2011, even though, based on the long-term average (LTA), it was a relatively warm, dry growing season in Winnipeg that year (Table 3). Biennial wormwood tends to proliferate in saturated soils, and its ability to withstand low-moisture environments seems limited (Kegode and Darbyshire

Table 3. Summary of monthly growing season temperatures and monthly growing season precipitation for each site-year followed by the average growing season temperature and total growing season precipitation and deviation from the 30-yr average for each site-year in parenthesis.

Month	Growing season conditions							
	Carman 2010		Melita 2010		Carman 2011		Winnipeg 2011	
	C	mm	C	mm	C	mm	C	mm
May	11.6	159	11.0	98	10.4	72	11.1	57
June	16.3	63	16.5	106	16.7	59	16.8	44
July	19.6	48	19.2	68	20.3	38	20.9	10
August	18.7	139	18.5	90	19.3	12	20.1	27
September	11.8	107	11.7	35	14.0	65	16.7	68
Average	15.6 ^a (−0.3) ^b	525 ^c (+190) ^d	15.4 (−0.5)	397 (+42)	16.1 (+0.2)	246 (−89)	17.1 (+1.2)	206 (−29)

^aThe average growing season temperature from May 1 to September 30.

^bThe deviation from the long-term average temperature based on data from the closest Environment Canada weather station from May 1 to September 30 for the years 1971 to 2000.

^cThe average total growing season precipitation from May 1 to September 30.

^dThe deviation from the long-term average total growing season precipitation based on data from the closest Environment Canada weather station from May 1 to September 30 for the years 1971 to 2000.

2013). The soil texture at the Winnipeg site was more fine than all other site-years, which may have contributed to these results. The effects of soil texture on recruitment and growth in biennial wormwood are not known.

In sunflower, variation in model parameter estimates among site-years has been documented for increasing densities of other weeds (e.g., Onofri and Tei 1994, Lewis and Gulden 2014), although perhaps not to the same extent as observed with biennial wormwood. The weather conditions observed in our experiment differed substantially among the two growing seasons. During the 2010 growing season, the growing conditions at Carman and Melita were cooler and wetter than the LTA, whereas in 2011 at both Carman and Winnipeg, it was warmer and dryer than the LTA (Table 3). The extended seedling recruitment periodicity of biennial wormwood (Zollinger and Ciernia 2007), in conjunction with different weather conditions, likely contributed to the differences observed in the model parameters among the site-years. Despite differences in model parameters among site-years, Onofri and Tei (1994) observed more consistent yield loss in sunflower in response to increasing weed densities in wild mustard and false Queen Anne's lace than we observed in biennial wormwood. Whether this was due to less variation in environmental conditions among the site-years or represents a fundamental difference in the biology among these weed species and how they interact with sunflower is not known.

The differences in the estimated model parameters contributed to two distinctly different action thresholds among the three experiments (Table 2).

A yield loss of 5% has been used in previous weed interference research as an acceptable amount of yield loss before control measures should be initiated (Knezevic et al. 1994). For the 2 site-yr that could be combined (Melita 2010 and Carman 2011), the action threshold for biennial wormwood in sunflower was about 40 plants m^{−2}, whereas in the higher yielding Winnipeg 2011 experiment, a 10-fold lower action threshold was observed. In the remaining two experiments an action threshold could not be determined. Taken together, these results indicate that the effect of biennial wormwood on sunflower yield was difficult to predict consistently using wormwood densities alone. Other factors that influence sunflower yield and time of recruitment of biennial wormwood also appear to play a significant role.

When biennial wormwood seedlings recruited after the four-leaf stage, yield loss in sunflower could not be described adequately using the rectangular hyperbola yield loss model, even at biennial wormwood densities as high as 3,358 plants m^{−2} at Carman 2011 (data not shown). When the biennial wormwood seedlings that were planted at the four-leaf sunflower stage recruited, the sunflower canopy was already well established. The crop canopy closed about 7 wk after sunflower planting (based on visual observations), and at that time, the biennial wormwood seedlings were still too small to interfere effectively with the sunflowers. Biennial wormwood remained in the understory of the crop for the rest of the growing season.

In general, results on early and late recruitment of biennial wormwood mirrored those reported by Lewis and Gulden (2014) in a similar experiment

Table 4. Summary of the regression equation describing the effect of early-recruiting biennial wormwood plants on sunflower seed size, thousand kernel weight, and head diameter. In the regression model, x represents biennial wormwood density (plants m^{-2}). Regression coefficient R^2 and P values are indicated.

Parameter	Site-year	Equation	R^2	P value
Sunflower seed size (cm^2 seed $^{-1}$)	Carman 2010	$y = 0.83 - 0.0002x$	0.64	0.055
	Winnipeg 2011	$y = 0.79 - 0.0004x$	0.66	0.048
Thousand kernel weight (g [1,000 seeds] $^{-1}$)	Carman 2010	$y = 129.3 - 0.09x$	0.71	0.035
	Carman2 2010	$y = 106.7 - 0.174x + 0.0008x^2$	0.99	0.001
	Winnipeg 2011	$y = 127.9 - 0.09x$	0.83	0.011
Sunflower head diameter (cm)	Carman 2010	$y = 17.1 - 0.007x$	0.78	0.020
	Winnipeg 2011	$y = 17.6 - 0.016x$	0.78	0.019

conducted on the interference of kochia with sunflower. Interference of early-recruiting kochia reduced sunflower yield on average by 33% (A value), and no yield reduction in response to increasing densities of kochia was observed when kochia recruited after the four-leaf stage of sunflower.

Sunflower seedlings can be susceptible to weed interference when weeds recruit before or at the same time as sunflower; however, sunflowers tend to be quite resilient to yield loss from later flushes of weeds (Johnson 1971). The importance of maintaining a weed-free environment in the early stage of a crop to prevent yield loss is well established in sunflower (e.g., Durgan et al. 1990) and other crops (e.g., Swanton and Weise 1991). Few options to manage biennial wormwood in sunflower are effective. Biennial wormwood has a natural tolerance to many PRE and POST broadleaf herbicides, making chemical management rather ineffective (Zollinger and Ciernia 2007). Mechanical control of biennial wormwood before crop emergence or using interrow cultivation after crop emergence could be an alternative (Pennacci and Tei 2014), but farmers in western Canada generally do not employ interrow cultivation, and intrarow weeds can still be problematic.

Early-recruiting biennial wormwood plants affected sunflower achene quality at some site-years, including at those in which no relationship to yield loss was detected. Reductions in achene size without reductions in yield suggests that much of the interference between biennial wormwood and sunflower occurred late. The growth of biennial wormwood seedlings is slow initially (Kegode and Fronning 2005), which would delay its ability to interfere with sunflower until later stages of sunflower development. Our results showed that achene size was reduced at Carman 2010 and Winnipeg 2011, and TKW was reduced at Carman 2010, Carman2 2010 and Winnipeg 2011 by increasing biennial wormwood densities (Table 4). In sunflower, test weight, achene size, and insect damage or damage from storage are requirements for seed grading and

quality (Anonymous 2011). Lewis and Gulden (2014) showed that head diameter in sunflower was more sensitive to interference with kochia than size of the sunflower achenes. In contrast to interference with kochia, the number of leaves per sunflower plant and height and stem diameter of the sunflower plants were not affected by the increasing densities of early-recruiting biennial wormwood. In comparison, interference of the parasitic weed sunflower broomrape with sunflower resulted in reductions in sunflower biomass, head size, stems, leaves, and achene number and size (Grenz et al. 2008).

Although increasing late-recruiting biennial wormwood plants did not reduce sunflower yield, interference by increasing densities of this weed did affect sunflower growth and development. At Melita 2010, a quadratic response in the number of leaves per sunflower plant with increasing biennial wormwood densities was observed ($y = 24.04 + 0.07x - 0.0003x^2$, $R^2 = 0.94$, $P = 0.025$). At Carman2 2010, the response in stem diameter to increasing biennial wormwood densities was quadratic ($y = 27.79 + 0.1159x - 0.0012x^2$, $R^2 = 0.99$, $P = 0.008$). Again, these results agree with those previously observed by Lewis and Gulden (2014) but are likely of little practical significance because sunflower yield was unaffected. The range in the response in sunflower yield components to interference with biennial wormwood suggested that maximum interference between biennial wormwood and sunflower varied greatly among site-years.

In summary, differences in weather, edaphic factors, and the inherent biology of biennial wormwood contributed to inconsistencies in sunflower yield loss caused by biennial wormwood. As a result, predicting yield loss in sunflower in response to increasing densities of biennial wormwood alone was difficult. Lack of yield loss did not preclude effects of biennial wormwood on sunflower seed quality. This and the inconsistency in predicting yield loss in sunflower made developing action thresholds for biennial wormwood in sunflower difficult, and critical

contributing factors other than density need to be investigated to understand better biennial wormwood interference with sunflower.

Acknowledgments

This research was funded by a grant from the National Sunflower Association of Canada (NSAC) and the Agri-Food Research and Development Initiative (ARDI) to R.H.G. The authors also thank Scott Day and Scott Chalmers of the Western Agricultural Diversification Organization (WADO) at Melita, MB, for their technical assistance.

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Received May 15, 2015, and approved August 7, 2015.

Associate Editor for this paper: John L. Linquist, University of Nebraska.