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Full Length Article

Control of Smoothseed Alfalfa Dodder (*Cuscuta approximata*) in Alfalfa (*Medicago sativa*)

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Abstract

This study aimed to determine the most appropriate control methods for smoothseed alfalfa dodder (*Cuscuta approximata* Bab.), infesting alfalfa (*Medicago sativa* L.) in Van, Turkey. The effects of imazethaphyr and imazamox herbicides and their application times, including three cutting heights of alfalfa, on controlling dodder were investigated. The study had a split-plot design with four replications, and the trial was conducted under field conditions from 2010 to 2013. The results indicated that pre-emergence applications of imazethaphyr, pre- and post-emergence applications of imazamox, suppressed dodder during the first year after application; however, the effect decreased in the following years. In 2013, a substantial decrease in the hay yield and raw protein ratio of alfalfa was found. In addition, the control of dodder was not significantly affected by different cutting heights. © 2017 Friends Science Publishers

Keywords: Alfalfa; Alfalfa dodder; Imazethaphyr; Imazamox; Cutting height; Plant protection; Crop science; Weed science

Introduction

Alfalfa (*Medicago sativa* L.), belongs to Fabaceae family and is a perennial, herbaceous, and deeply rooted forage crop with wide range of adaptability. The feed value of alfalfa is higher than all other forage crops (Ahlgren, 1956; Chrisman *et al.*, 1982). Alfalfa is the most widely grown forage crop worldwide (32 m ha) and in Turkey on 629 thousand hectares (FAO, 2013; TSI, 2013).

Alfalfa crop is usually free of weeds owing to strong competition. However, this does not hold true for alfalfa dodder. Smoothseed alfalfa dodder (*Cuscuta approximata* Bab.) reduces alfalfa yields by preventing plant growth, a major dodder species (Uygur, 1991). Smoothseed alfalfa dodder is an annual holoparasitic plant belonging to the Cuscutacea family (Löffler *et al.*, 1997). Dodder twines around stems, uses numerous small haustoria to penetrate the tissues of the host plants, and feeds from the vascular system. Consequently, dodder leads to a significant decrease in the yield and yield quality, keeping the crops weak and powerless (Dawson *et al.*, 1994; Mishra, 2009). In addition, it is responsible for the transfer of viruses, mycoplasmas, and macromolecules from one host to another, and it may be involved in horizontal gene transfer (Cali *et al.*, 1993; Costea, 2014; Hull, 2014). There are ~200 dodder species in the world, but only 16 species are found in Turkey. Smoothseed alfalfa dodder is the most important species of alfalfa in Turkey and has been dispersed worldwide from southern Europe. Each fruit contains one to four seeds, with colours varying from yellow to greenish-brown, and seeds mature before the first alfalfa cutting period. One side of the seeds is clearly angular and its average size is 1.0 × 0.7 mm (Kadioglu, 1992). The seed survival time in the soil is quite long and, therefore, it is extremely difficult to control (Carey *et al.*, 1993).

Quality forage from ranges, pastures, and forage crops in Turkey is needed to obtain the desired yields in livestock production. The production of forage crops has been supported by the State since 2000 to prevent a forage deficit (Acikgoz *et al.*, 2005; Celik and Demirbag, 2013). There has been a significant increase in alfalfa production owing to this policy in Turkey. The alfalfa cultivation areas covered 246,000 ha in 1999, increased to 535,000 ha in 2007, and reached 629,000 ha in 2013 in Turkey. The presence of dodder increased in the alfalfa fields in the city of Van as alfalfa production increased (Yildirim and Tepe, 2014; Anac *et al.*, 2011). Because of the increase in alfalfa production, its seeds contaminated with dodder seeds have been widely distributed. The problem has grown owing to the uncontrolled sale and use of contaminated seeds (Acikgoz, 2001).

The control of dodder is very important in increasing the yield and quality of alfalfa, and approaches included preventive measures against contamination, crop rotation, flaming, mechanical and chemical controls (Radi, 2007; Tepe, 2014). There are some chemicals, atrazine, simazine,

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chlorprophame, and propyzamide, that control dodder, but at present, the possibility of a chemical control is limited, and only some herbicides, such as imazethaphyr and imazamox, are recommended (MFAL, 2009).

Dodder may be dispersed in many different ways. For this reason, research on appropriate control methods, such as integrated pest management, is important (Cunningham and Brown, 2006). While considering the effects of inappropriate and unnecessary herbicide use on human health and the environment, choosing suitable control methods is important.

The aim of this study was therefore to determine the most appropriate control methods for smooth seed alfalfa dodder, which infests alfalfa fields. The effects of two herbicides and their application times, as well as cutting heights of alfalfa, were investigated for controlling dodder.

Materials and Methods

Trials were established in 2010 in the experimental research area (38°33′47″N, 43°18′00″E, 1660 m asl) of the Faculty of Agriculture at the Yüzüncü Yil University, and the studies were conducted for 4 years. The alfalfa (*M. sativa* L.) cultivar Bilensoy 80 well adapted to the region was used in the study (Sengul *et al.*, 2003).

The soils of the experimental areas are classified as Entisols according to the United States Department of Agriculture (USDA) taxonomy. Soil samples were taken from 0 to 40 cm depth, and analysed chemically and physically. The soils of the trial area are a loamy texture with a high $CaCO_3$ content (15.5%), low levels of organic matter (1.83%) and nitrogen (N), moderate levels of available phosphorus (P), and a sufficient potassium (K) content. They had a low salt content (0.02%) and were slightly alkaline (pH 7.8). Some local climatic data for 2010–2013 are presented in Table 1.

The study was implemented in a split-plot design with four replications. The main plot sizes were 60 m^2 and sub-plots of 20 m^2 (4 m × 5 m) based on the criteria proposed by EPPO (1986). All of the parcels were contaminated manually (1 g seed m^{-1}) with smoothseed alfalfa dodder (*C. approximata* Bab.) before sowing in October 2009, and all of the applications were compared with control parcels without dodder. Alfalfa was planted in rows 20 cm apart at a rate of 15 kg seeds ha^{-1} on 16 April 2010. Seedlings emerged uniformly in 17–24 days after sowing. In the study, the efficacies of two herbicides, imazethaphyr [Sansuit (100 g active ingredient (ai) L^{-1}): Agrobest], and applied three times (pre-sowing, pre-emergence, and post-emergence) at a rate of 50 g ai ha^{-1}, and imazamox [RAPTOR 129 SL (120 g a.i. L^{-1}): BASF] applied twice (pre-emergence and post-emergence) at a rate of 48 g ai ha^{-1}, were investigated for the control of dodder. The three different cutting heights (5, 10 and 15 cm) were also included (Wiersma, 2000). Herbicide treatments were applied with a battery-powered backpack sprayer [Matabi (0.2–0.4 MPa), Spain] using a boom with flat-fan nozzles (Teejet TIM 03–F 110; Timsan, Turkey). Pre-sowing applications were carried out by soil incorporation on 6 April 2010, pre-emergence applications on 20 April 2010, and post-emergence applications on 3 June 2010.

The trial was regularly irrigated using a sprinkler system and areas between parcels were weeded. Dodder seedlings attached to alfalfa stems were monitored at 10 day intervals for two months after emergence (Sarpe *et al.*, 1992; Lanini and Kogan, 2005; Mishra *et al.*, 2007). Alfalfa was cut for clean up in the first year and no data obtained, during 1st year of establishment and obtained in the following years.

Alfalfa was cut three times a year and data related to dodder infestation alfalfa plant height, hay yield, and the raw protein ratio were obtained (Table 2). The amount of the dodder dry-biomass, number of attached alfalfa stems, and parasitizing level of dodder were measured to understand the dodder infestation. Measurements related to the plant height, number of attached stems and parasitizing level of dodder were carried out in the field. A scale of 1–5 was used to determine the parasitizing level of dodder (Rao, 2000). Samples were brought to the laboratory for the hay yield and raw protein ratio measurements of alfalfa and measurements of the dodder dry-biomass (Bulgurlu and Ergul, 1978; Mansoor *et al.*, 2004). Measurements were carried out in three cutting periods, and performed analyses using averages of the data from the three cutting periods in each year.

The effect of treatments on dodder, yield and yield components of alfalfa, as well as the differences among treatments, were analysed using analysis of variance procedures for a split-plot design with the SAS (2015) statistical package. The *F*-value of the analysis of variance was considered significant at $P > 0.05$ level of probability. The means related to the effects of applications and cutting heights on the investigated criteria were evaluated using Tukey's range test.

Results

The effects of herbicide applications on the numbers of attached alfalfa stems, dodder dry-biomass, and scale values of dodder infestation were statistically significant in all three years. In the second year (2011), the number of attached alfalfa stems, dodder dry-biomass, and scale value of dodder infestation were high with pre-sowing and post-emergence applications of imazethaphyr. These values were lowest in the control parcels without dodder; and the closest values to those of the dodder-free control were obtained with pre-emergence applications of imazethaphyr, pre-sowing and post-emergence applications of imazamox. During 3rd year (2012), similar trend was observed as for during 2011 i.e. the lowest level of infestation in the dodder-free control parcels and the closest with pre-emergence imazethaphyr and post-emergence applications of imazamox.

Table 1: Some climatic data for the growing seasons of 2010–2013, and long-term averages (LTA, 1949–2013) in Van, Turkey (TSMS, 2014)

Months	Precipitation (mm)					Average temperature (°C)				
	2010	2011	2012	2013	LTA	2010	2011	2012	2013	LTA
January	51.6	14.2	54.5	64.9	31.8	0.1	-4.2	-3.6	-1.6	-3.5
February	71.1	26.6	43.6	40.5	33.0	1.4	-3.0	-6.1	-0.1	-2.9
March	38.3	30.7	77.6	39.3	45.6	5.8	0.1	-4.1	3.1	1.5
April	46.3	133.7	41.6	36.0	57.2	8.4	7.4	8.3	9.8	7.7
May	69.8	62.8	38.3	48.0	46.6	13.3	12.4	14.6	13.9	13.1
June	41.0	28.1	8.7	8.6	18.8	19.8	18.9	19.4	18.9	18.2
July	0.0	11.0	3.0	0.0	5.1	24.0	22.2	21.2	23.3	22.3
August	1.0	0.0	1.5	0.0	3.4	23.2	21.7	22.0	22.1	21.9
September	3.8	53.0	10.7	8.7	13.0	9.5	15.8	16.1	17.4	17.4
October	45.8	125.9	40.0	21.4	48.7	12.6	7.2	10.0	9.6	10.5
November	0.0	18.8	26.0	35.9	51.5	4.3	-1.4	4.3	6.1	4.7
December	8.7	12.1	60.3	33.6	42.0	2.0	-3.2	-0.4	-3.5	-0.7
Total	377.4	516.9	405.8	336.9	396.7					
Average						10.37	9.48	8.48	9.92	9.18

Table 2: Cutting dates of alfalfa

Year	First cutting	Second cutting	Third cutting
2011	16 June 2011	4 August 2011	4 October 2011
2012	13 June 2012	1 August 2012	16 September 2012
2013	12 July 2013	3 September 2013	25 September 2013

Table 3: Effect of herbicide applications on infestation of alfalfa dodder

Applications	Number of attached alfalfa (stem m^{-1})			Dry biomass of dodder (g m^{-1})			Scale value of infestation		
	2011	2012	2013	2011	2012	2013	2011	2012	2013
Imazethaphyr (Pre-sowing)	37.50 a	66.50 b	55.80 a	12.40 ab	39.40 a	68.60 a	1.90 a	2.00 ab	3.70 a
Imazethaphyr (Pre-emergence)	0.80 b	36.10 bc	49.80 a	0.30 b	22.00 ab	39.80 a	1.10 b	1.60 bc	3.30 a
Imazethaphyr (Post-emergence)	41.10 a	54.90 b	56.70 a	24.80 a	26.20 a	46.00 a	1.80 a	1.80 ab	3.60 a
Imazamox (Pre-emergence)	2.50 b	50.10 b	62.30 a	0.50 b	27.70 a	58.50 a	1.10 b	2.00 ab	3.50 a
Imazamox (Post-emergence)	8.60 b	40.10 bc	77.80 a	8.80 b	21.80 ab	60.00 a	1.20 b	1.70 b	3.50 a
Control (Infested with dodder)	22.70 ab	111.70 a	69.40 a	10.80 ab	43.50 a	57.00 a	1.10 b	2.40 a	4.30 a
Control (Without dodder)	00.0 b	00.0 c	0.80 b	00.0 b	00.0 b	0.30 b	1.00 b	1.00 c	1.60 b
Mean Square	11161.0	41310.0	20382.0	2934.0	7199.0	16495.0	4.5	6.4	22.7
F Value	10.53**	10.62**	5.56**	6.21**	6.02**	3.74**	16.44**	7.80**	41.14**

Scale: 1 = No infestation, 2 = Very low infestation, 3 = Moderate infestation, 4 = High infestation, 5 = Full infestation (deaths in alfalfa); ** P > 0.01

The highest infestation occurred in the control parcels infested with dodder. In fourth year (2013), the data on the numbers of attached alfalfa stems, dry-biomass measurements of dodder, and scale values of dodder infestations were low only in the dodder-free control parcels. All of the other applications were grouped together, and an intensive dodder infestation occurred (Table 3).

The effects of herbicide applications on alfalfa plant height and hay yield were statistically significant in 2011 and 2013 only but not in 2012, while effects on raw protein ratio were significant only in 2013. The highest plant height, hay yield, and raw protein ratio values were obtained from the dodder-free control. However, the other applications remained in the same group. In the fourth year, the lowest values for the three alfalfa measurements were obtained from control parcels infested with dodder (Table 4).

The effects of cutting heights on the number of attached alfalfa stems, dry-biomass of dodder, and scale value of dodder infestation were not statistically significant in any year of study. The effect of cutting heights on the scale value of dodder infestation was only significant in the second year and the lowest infestation was observed at 15 cm and highest at a 10-cm cutting height (Table 5). The effects of cutting heights on the alfalfa plant's height and hay yield were also not statistically significant in any year. However, the effects of cutting heights on the raw protein ratio were only significant in 2011. The highest raw protein ratio was obtained at the 15 cm cutting height, and the lowest at the 10 cm cutting height (Table 6).

Discussion

In field experiments, the effects of herbicides on dodder infestations and alfalfa yield parameters were significant in 2011, decreased in 2012 and were ineffective by 2013. Thus, it may be necessary to use herbicides every year to keep dodder out of fields during alfalfa cultivation. In terms of dodder infestations and alfalfa yield parameters, a pre-emergence application of imazethaphyr, pre- and post-emergence applications of imazamox, were the most effective during the first two years after applications.

Table 4: Effect of herbicide applications on plant height, hay yield, and raw protein ratio of alfalfa dodder

Applications	Plant height (cm)			Hay yield (kg ha^{-1})			Raw protein ratio (%)		
	2011	2012	2013	2011	2012	2013	2011	2012	2013
Imazethaphyr (Pre-sowing)	83.40 b	86.30	62.30 b	5245 ab	4813	2289 b	20.30	18.10	15.00 b
Imazethaphyr (Pre-emergence)	88.10 ab	89.60	63.80 b	5914 ab	5204	2882 b	20.10	18.50	14.90 b
Imazethaphyr (Post-emergence)	87.30 ab	87.10	49.70 b	5568 ab	5336	2172 b	20.20	18.20	12.40 b
Imazamox (Pre-emergence)	88.90 ab	88.30	48.70 b	5079 b	5063	1920 b	20.20	18.80	13.00 b
Imazamox (Post-emergence)	87.90 ab	86.00	58.90 b	5721 ab	5313	2256 b	20.40	18.60	15.00 b
Control (Infested with dodder)	89.30 ab	85.40	42.60 b	5699 ab	4931	1698 b	20.80	17.80	10.80 b
Control (Without dodder)	90.10 a	88.80	85.30 a	6139 a	5581	4705 a	20.00	18.40	17.30 a
Mean Square	171.5	87.5	6070.0	48559.0	248856.0	332894.0	2.0	3.8	13.1
F Value	2.00*	1.32	8.55**	2.52*	20.76**	20.76**	1.50	1.33	3.83**

* P > 0.05; ** P > 0.01

Table 5: Effect of cutting heights on infestation of alfalfa dodder

Cutting heights(cm)	Number of attached alfalfa (stem m^{-1})			Dry biomass of dodder (g m^{-1})			Scale value of infestation		
	2011	2012	2013	2011	2012	2013	2011	2012	2013
5	15.90	55.30	51.60	6.80	27.80	48.30	1.30 ab	1.80	3.30
10	20.80	41.70	50.40	10.90	20.90	43.30	1.50 a	1.60	3.20
15	11.80	57.00	48.40	7.00	28.70	40.50	1.20 b	1.90	3.30
Mean Square	1710.0	5883.0	203.0	44.3	1518.0	1060.0	1.0	1.7	0.01
F Value	1.60	1.51	0.06	0.90	0.30	0.20	3.50 *	1.90	0.02

Scale: 1 = No infestation, 2 = Very low infestation, 3 = Moderate infestation, 4 = High infestation, 5 = Full infestation (deaths in alfalfa); * P > 0.05

Table 6: Effect of cutting heights on plant height, hay yield and raw protein ratio of alfalfa dodder

Cutting heights (cm)	Plant height (cm)			Hay yield (kg ha^{-1})			Raw protein ratio (%)		
	2011	2012	2013	2011	2012	2013	2011	2012	2013
5	88.70	87.90	60.60	5740	5195	2552	20.30 ab	18.60	14.50
10	87.30	86.60	60.10	5617	5465	2716	20.00 b	18.30	14.00
15	87.40	87.50	60.90	5514	4872	2841	20.60 a	18.20	14.40
Mean Square	45.9	33.9	17.0	10792.0	74089.0	11951.0	6.5	3.8	4.0
F Value	0.50	0.50	0.02	0.60	2.70	0.75	3.90*	1.30	0.14

* P > 0.05

This corroborated that effects of pre- and post-emergence applications of imazethaphyr and imazamox, independently, on the control of dodder in alfalfa (Nice *et al.*, 2006; Mahadevappa and Bhanumurthy, 2007; Sarić-Krsmanović *et al.*, 2015). In 2013, a significant yield loss was observed in comparison with the values in 2011 and 2012. In fourth year, the hay yield varied between 1823 and 3007 kg/ha in dodder-free control parcels and the dodder-infested parcels, respectively. The values for the raw protein ratio in 2011 and 2012 were within the expected range (15.6–23.3%) and close to each other (Kehr *et al.*, 1979; Acikgoz, 2001). The raw protein ratio was below the expected level for each of the applications in the fourth year of the study, except for the control parcels without dodder. These results indicated that dodder suppression had completely limited alfalfa growth by the fourth year. Thus, we concluded that dodder uses proteins, as well as the metabolites, transported through the alfalfa symplastic system, and substantially decreases the plant's protein content (Haupt *et al.*, 2001; Jiang *et al.*, 2013).

In this study, dodder infestations and alfalfa yield parameters were not affected by different cutting heights, eliminating it as a convenient way to control dodder (Anter and Kassim, 2011).

Conclusion

Pre-emergence application of imazethaphyr, pre- and post-emergence applications of imazamox, substantially suppressed dodder during the first year after application in alfalfa. In the fourth year, the herbicides lost effectiveness, the dodder infestation increased, and the alfalfa yield significantly decreased. Unfortunately, different cutting heights of alfalfa were ineffective in controlling dodder as a conventional weed control method. According to these results, it is suggested that imazethaphyr or imazamox should be applied as pre-emergence applications in first year and in following years; imazamox should be applied each year for controlling dodder in alfalfa.

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