

Valerian (*Valeriana officinalis* L.) tolerance to some post-emergence herbicides

Nadia Monjezi, Jamshid Razmjo, Hassan Karimmojeni*

Department of Agronomy and Plant Breeding, College of Agriculture, Isfahan University of Technology, Isfahan, 84156-83111, Iran

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Abstract: Valerian (*Valeriana officinalis* L.) is a medicinal plant, but its cultivation is restricted by weed competition. Therefore, three rates (0.75X, 1X, and 1.25X, where X is equal to the recommended dose of haloxyfop-R (methyl ester), sethoxydim, oxadiargyl, bentazon, oxadiazon, and oxyfluorfen) were applied at the 3–4 leaf stages to valerian plants. This application was done to select the herbicide type and rate for post-controlling broadleaf and grasses weeds in this species. Herbicide injury, Soil-Plant Analyses Development (SPAD) reading, number of leaves per plant, stem diameter, and fresh and dry weights were determined 10, 20, and 30 days after herbicide application. Oxyfluorfen application caused the most herbicide injury followed by bentazon. Injury increased as the rate and the days after application increased. Oxadiazon only caused significant damage 30 days after application under all three rates. Other treatments showed no marked injuries under any rate or date after application, as compared with the control. Effects on other measured traits depended on the trait, herbicide, and herbicide rate. The highest SPAD, leaf number, shoot diameter, fresh weight and dry weight, was recorded under application of 30 mg a.i. · kg⁻¹ soil oxadiargyl and 90 mg a.i. · kg⁻¹ soil oxadiazon, 81 mg a.i. · kg⁻¹ soil haloxyfop-R, 37.5 mg a.i. · kg⁻¹ soil oxadiargyl, 22.5 mg a.i. · kg⁻¹ soil oxadiargyl, 81 mg a.i. · kg⁻¹ soil haloxyfop-R, and 81 mg a.i. · kg⁻¹ soil haloxyfop-R, respectively. To sum up, the results showed that sethoxydim, oxadiargyl, and haloxyfop-R produced no significant symptoms of phytotoxicity or reduction of measured traits. This means that oxadiargyl, haloxyfop-R, and sethoxydim may be used safely for weed control of valerian at the rates used in this experiment under similar conditions.

Key words: accase inhibitor herbicide, medicinal plant, photosynthesis inhibitor herbicides, selective weed control

Introduction

Valerian (*Valeriana officinalis* L.) (Valerianaceae) is a perennial flowering medicinal plant native to Europe and Asia. Weed control of valerian is one of the main limiting factors for production of this species especially when valerian is being established. Use of herbicides in weed control of some medicinal plants has been reported. Qasem and Foy (2006) applied pre-planting and post-planting of oxyfluorfen and oxadiazon to *Origanum syriacum* L., and reported that both herbicides were highly selective and effective for weed control of this species. Zheljzako *et al.* (2006) showed that application of pendimethalin and metribuzin alone or in combination were safe for weed control of *Silybum marianum* L. Forcella *et al.* (2012) tested the tolerance of *Calendula officinalis* L. to desmedipham and phenmedipham postemergence herbicides, and showed that this species was tolerant to both herbicides. Yousefi and Rahimi (2014) applied trifluralin and pendimethalin herbicides to *Foeniculum vulgare* Mill. and reported that both herbicides provided some weed control without any injury to the crop, but pendimethalin was more effective. Yet, there is little information on the tolerance of valerian to the application of herbicides. There have been a number of herbicides found in Poland that could be used for weed control of valerian, after sowing valerian seedlings

and after growing weed species. Studies showed that foliar application of the haloxyfop-R herbicide after using pendimethalin herbicide and by using mechanical implement methods, caused maximum root and rhizome productivity. It also caused a low degree of damage to this herbal plant. Also, mechanical implement methods decreased the amount of weeds in crops (Kwiatkowski 2010). Pank *et al.* (1980) state that the application of chlorpropham controlled weeds in valerian and that this herbicide did not have an effect on the quality and quantity of valerian.

Thus, information regarding both crop tolerance and sensitivity to an herbicide are required for chemical weed control in this species. If tolerance is adequate, herbicides will then provide management of grass and broadleaf weeds. The objective of this study was to determine the tolerance of valerian to the post-planting application of bentazon (Basagran), oxadiargyl (Top Star), oxadiazon (Ronstar), oxyfluorfen (Goal), haloxyfop-R (methyl ester) (Gallant Duper), and sethoxydim (Nabo-S).

Materials and Methods

This experiment was conducted in the research greenhouse, located in the Faculty of Agriculture, Isfahan

*Corresponding address:
kmojeni@cc.iut.ac.ir

University of Technology, in 2014. The experiment was performed as factorial, based on the randomised complete block design with 19 treatments and 3 replications. Valerian plant seedlings, in compound media (50% cocopeat and 10% perlite and 40% soil) were prepared and the seedlings at the 2–3 leaf stages were transferred to pots. The pots had a 15 cm diameter and 20 cm height. Pots were irrigated every 3 days, during the growing season, and the plants before application of herbicides, were thinned to 3 plants per pot.

In this experiment, haloxyfop-R (methyl ester) (Galant Super) and sethoxydim (Nabo-S) as graminicide and broadleaf herbicides including oxadiargyl (Top Star), bentazon (Basagran), oxadiazon (Ronstar), and oxyfluorfen (Goal), as well as a control (not-sprayed), were used.

The herbicides were applied at three different doses: 0.75X (D1), 1X (D2), and 1.25X (D3) where X is equal to the recommended dose. Recommended dosages included: haloxyfop-R (methyl ester) – 108 mg a.i. · kg⁻¹ soil (10.8% EC), sethoxydim – 125 mg a.i. · kg⁻¹ soil (12.5% OEC), oxadiargyl – 30 mg a.i. · kg⁻¹ soil (3% EC), bentazon – 480 mg a.i. · kg⁻¹ soil (48% SL), oxadiazon – 120 mg a.i. · kg⁻¹ soil (12% EC), and oxyfluorfen 240 mg a.i. · kg⁻¹ soil (24% EC) (Table 1).

Herbicides were applied, at the beginning growth stage of the crop, at the 3–4 leaf stages of the valerian plant. After the herbicide treatment, plants were checked for symptoms (Stunting, chlorosis, and necrosis). Plants were compared to the control by visual ratings provided by the Weed Research Society of Europe (EWRS). Damage to the product was defined in three steps: 10, 20, and 30 days after treatment (DAT) using a scale of 0–100%, where a score of 0 means no visible damage to the plant, and a score of 100 means plant death. The leaf Soil-Plant

Analyses Development (SPAD) index, using a chlorophyll meter for the same leaves, was measured and its average was recorded for each pot. In the 6–7 leaf stages, the fresh and dry weight of the shoots were measured. To do the measuring, plants were harvested from the above soil level, and dry matter accumulation was determined by drying the plant shoot material for 48 h in an oven at 75°C. Other traits measured included stem diameter (cm), and number of leaves per plant.

Statistical analysis

Before analysing the data, the assumption of a homogeneous variation was tested. Finally, the analysis of variance for the data collected for each attribute, using the statistical program (Statistics) was done. If the analysis of variance indicated statistically significant differences, the means were compared using Fisher's protected least significant difference test ($p > 0.05$).

Results and Discussion

Crop injury

Herbicide application had a significant effect on crop injury 10, 20, and 30 days after the application of the herbicide (Table 2). Oxyfluorfen caused the highest injury followed by bentazon under D1, D2, and D3 (D1 = 0.75X, D2 = X, and D3 = 1.25X). Injury appeared over time and became more pronounced (Table 3). Oxadiazon application under D3 only, showed significant injury. The other herbicides had no marked injury under any date after application.

Table 1. Herbicides and herbicide rates used in the experiment

Herbicides	Proportion of labeled use rate		
	0.75X (D1)	1X (D2)	1.25X (D3)
Bentazon	360.0	480	600.0
Oxadiargyl	22.5	30	37.5
Oxadiazon	90.0	120	150.0
Oxyfluorfen	180.0	240	300.0
Haloxyfop-R (methyl ester)	81.0	108	135.0
Sethoxydim	93.75	125	156.25

Rates of herbicides are presented in (mg a.i. · kg⁻¹ soil)

Table 2. Analysis of variance results for injury score of *Valeriana officinalis* 10, 20, and 30 days after treatment (DAT) with herbicides

Source of variation	df	% Injury ^a (DAT)		
		10 DAT	20 DAT	30 DAT
Herbicide	6	5,131.26***	6,388.42***	7,574.24***
Dose	2	153.16***	63.58**	90.21*
Herbicide × dose	12	101.86***	33.5***	16.84
Error	40	2.34	8.83	19.27
CV	–	14.21	21.08	27.30

*significant at $p \leq 0.05$; **significant at $p \leq 0.01$; ***significant at $p \leq 0.001$

^acrop injury score using a scale of 0–100 (%) where 0% represents no injury to plants and 100% means dead plants

df – degrees of freedom; CV – coefficient of variation

Table 3. Effect of herbicide on the injury score of *Valeriana officinalis* 10, 20, and 30 days after treatment (DAT)

Herbicides	% Injury ^a (DAT)		
	10 DAT	20 DAT	30 DAT
The control	1.0 c	1.0 c	1.0 d
Bentazon	3.1 b	16.4 b	14.3 b
Oxadiargyl	2.4 bc	2.0 c	1.9 d
Oxadiazon	1.5 c	2.5 c	8.3 c
Oxyfluorfen	64.8 a	73.2 a	81.0 a
Haloxyfop-R (methyl ester)	1.0 c	2.1 c	2.0 d
Sethoxydim	1.3 c	1.3 c	4.0 d
LSD	1.45	2.83	4.2

Treatments with the same letter in a column are not significantly different at $p < 0.05$ as assessed by the LSD test

^acrop injury score using a scale of 0–100 (%) where 0% represents no injury plants and 100% means dead plants

Table 4. Effect of various doses of herbicide on the injury score of *Valeriana officinalis* 10, 20, and 30 days after treatment (DAT)

Rate of application [mg a.i · kg ⁻¹]	% Injury ^a (DAT)		
	10 DAT	20 DAT	30 DAT
0.75X (D1)	8.2 c	12.4 b	14.0 b
1X (D2)	10.5 b	14.0 b	16.0 ab
1.25X (D3)	13.6 a	15.9 a	18.2 a
LSD	0.95	1.85	2.7

Treatments with the same letter in a column are not significantly different at $p < 0.05$ as assessed by the LSD test

^acrop injury score using a scale of 0–100 (%) where 0% represents no injury plants and 100% means dead plants

The percent injury increased with an increased rate and became more pronounced over time (Table 4). Oxyfluorfen herbicide injury increased the most, followed by bentazon as the rate increased, and became more pronounced over time (Table 5). Marked injury was noted after bentazon had been applied at 20 and 30 days, under all three rates. Oxadiazon only caused significant damage at 30 days after application under all three rates. The other treatments showed no marked injuries under any rate or date after application, as compared with the control (Table 5).

Bentazon is a benzothiadiazole post-emergence herbicide that can control several broadleaf weed species (Vencill 2002). Bentazon inhibits the electron flow in the photosynthesis pathway (Mine *et al.* 1975). Soltani *et al.* (2008) reported that application of 840 g · ha⁻¹ bentazon once caused no injury in pinto bean, but when it was applied twice, it caused a 4% injury after 7 days, but no significant injury at 14 or 28 DAT. Beuer *et al.* (1995) also reported no marked injury in pinto bean 7 and 14 days after treatment with 420, 840, and 1,680 g · ha⁻¹ bentazon. In contrast, Soltani *et al.* (2006) reported 30 to 33% plant injury in Adzuki bean when bentazon was applied at 1,080 g · ha⁻¹. The visual injury range to sweet corn from the bentazon application was 0 to 69%, depending on the cultivar and rate of application (Diebold *et al.* 2004). Contrasting results could be due to genotype, application rate, plant growth, and the amount of DAT of the herbicide.

Sethoxydim and haloxyfop inhibit acetyl-CoA (ACC) carboxylase that is needed for fatty acid synthesis and the subsequent production of phospholipids which is needed for plant cell membrane. These herbicides control an-

nual and perennial grass species (Vencill 2002; Pavlivna *et al.* 2014). Their selectivity for grass control depends on ACCase susceptibility in the grass species. Normally, dicotyledonous contains an ACCase less susceptible to inhibition by such herbicides, when compared to the enzyme in grasses (Stoltenberg *et al.* 1989). Sethoxydim and haloxyfop-R injuries were not significantly different from that of the control under any rate or day after application. These results were expected since both herbicides are used for controlling grass weeds in dicotyledonous crops. In line with our results, Soltani *et al.* (2006) reported that sethoxydim at 500 and 1,000 g · ha⁻¹ caused between 0 to only 4% injury at lower rates of use and 1 to 5% at higher rates, in Adzuki bean. Visual injury increased with an increasing rate of herbicide use but the rate was reduced over time. In contrast, Al-Khatib *et al.* (2003) reported that injury to grain sorghum depend on the cultivar, year, and rate of sethoxydim application, and the injury was from 0 to 99%.

Oxadiazon inhibits the Hill reaction of photosynthesis. Oxadiazon is a selective pre-emergence herbicide for controlling annual grasses and broad leaf weeds. Our results showed that oxadiazon application had no marked injuries to valerian 10 to 20 DAT at any rate, but caused 6.1 to 11.3% injury, 30 DAT (Table 5). That was expected since this herbicide is a long-lasting performance herbicide. Brecke *et al.* (2010) applied oxadiazon to turfgrass species prior to or following turfgrass sprigging. The herbicide did not cause any injury, but caused serious injury at sprigging. Lamont and Spohr (1988) showed that application of oxadiazon produced no injury on ornamental plants.

Table 5. Interactions of herbicides and their doses on the injury score of *Valeriana officinalis* 10, 20, and 30 days after treatment (DAT)

Treatment	Rate of application [mg a.i. · kg ⁻¹]	% Injury (DAT)		
		10 DAT	20 DAT	30 DAT
The control	–	1.0 e	1.0 e	1.0 e
Bentazon	D1	1.0 e	14.8 d	10.8 cd
	D2	3.1 de	16.6 d	15.3 c
	D3	3.5 de	17.8 d	16.6 c
Oxadiargyl	D1	1.3 e	1.6 e	1.3 e
	D2	1.6 e	1.6 e	1.6 e
	D3	4.3 d	2.6 e	2.6 e
Oxadiazon	D1	1.0 e	2.3 e	6.1 de
	D2	1.0 e	2.3 e	7.5 de
	D3	2.6 de	3.0 e	11.3 cd
Oxyfluorfen	D1	49.3 c	64.3 a	74.3 b
	D2	64.3 b	72.6 b	81.0 ab
	D3	81.0 a	82.6 a	87.6 a
Haloxypop-R (methyl ester)	D1	1.0 e	1.6 e	1.3 e
	D2	1.0 e	2.3 e	2.3 e
	D3	1.0 e	2.3 e	2.3 e
Sethoxydim	D1	1.0 e	1.0 e	3.3 e
	D2	1.3 e	1.3 e	3.1 e
	D3	1.6 e	1.6 e	5.6 de
LSD	–	2.52	4.9	7.24

Treatments with the same letter in a column are not significantly different at $p < 0.05$ as assessed by the LSD test

Table 6. Analysis of variance results for chlorophyll content, leaf number, plant height, shoot diameter, shoot fresh and dry weight of *Valeriana officinalis* after treatment with herbicides

Source of variation	df	Chlorophyll content	Leaf number	Shoot diameter	Shoot fresh weight	Shoot dry weight
Herbicide	6	50.37***	1.72*	0.69**	78.65***	0.83***
Dose	2	20.45	0.12	0.33	40.62***	0.31***
Herbicide × dose	12	10.80	0.53	0.24	3.47***	0.02**
Error	40	6.6	0.62	0.18	0.55	0.007
CV	–	12.56	15.95	21.63	9.59	10.02

*significant at $p \leq 0.05$; **significant at $p \leq 0.01$; ***significant at $p \leq 0.001$

df – degrees of freedom; CV – coefficient of variation

Oxadiargyl herbicide inhibits the enzyme which is in the pigment synthesis pathway (Boger and Wakabayashi 1995). Application of oxadiargyl resulted in no marked injury to valerian at any rate or day after application. In contrast, at least in part, Alebrahim *et al.* (2012) reported that application of oxadiargyl caused 1 to 23% injury to potato, but three weeks after application the plants had recovered. Nethra and Jagannath (2011) showed that maize and sunflower injuries increased when oxadiargyl concentrations were increased.

Oxyfluorfen kills broadleaf plants by destroying cell membranes within the leaves and shoots. Oxyfluorfen 10, 20, and 30 DAT caused 49.3 to 81.0%, 64.3 to 82.6%, and 74.3 to 87.6% injuries to the crop, respectively (Table 5). Loken *et al.* (2010) reported that oxyfluorfen application injured onion about 15%. While Norsworthy *et al.* (2007), reported that application of oxyfluorfen (134 g · ha⁻¹) only caused 3% injury in green onion.

Chlorophyll content and morpho-physiological traits

Herbicide application had a significant effect on the SPAD reading, number of leaves per plant, stem diameter, and fresh and dry weights (Table 6). On average, all of the herbicides reduced fresh and dry weights, stem diameter, and the SPAD reading, but increased or did not affect leaf number (Table 7). The following were not affected: chlorophyll content, reduced fresh weight and dry weight, and leaf number and shoot diameter, when the herbicide rate was increased (Table 8). However, these effects depend on the measured trait. Herbicide and herbicide rate, and the highest chlorophyll content, leaf number, shoot diameter, fresh weight and dry weight were recorded under application of 30 mg a.i. · kg⁻¹ soil oxadiargyl and 90 mg a.i. · kg⁻¹ soil oxadiazon, 81 mg a.i. · kg⁻¹ soil haloxypop-R, 37.5 mg a.i. · kg⁻¹ soil oxadiargyl, 22.5 mg a.i. · kg⁻¹ soil oxadiargyl, 81 mg a.i. · kg⁻¹ soil haloxypop-R and 81 mg a.i. · kg⁻¹ soil haloxypop-R, respectively (Table 9).

Table 7. Effect of herbicides on chlorophyll content, leaf number, plant height, shoot diameter, shoot fresh and dry weight of *Valeriana officinalis*

Herbicides	Chlorophyll content [spad value]	Leaf number [plant]	Shoot diameter [cm]	Shoot fresh weight [g]	Shoot dry weight [g]
The control	23.5 a	4.5 bc	2.4 a	10.5 a	1.0 a
Bentazon	19.9 b	4.2 c	1.94 bcd	5.2 d	0.51 c
Oxadiargyl	21.2 ab	4.8 abc	2.10 ab	9.1 b	0.98 a
Oxadiazon	21.3 ab	5.2 a	1.75 cd	8.3 c	0.87 b
Oxyfluorfen	15.7 c	4.3 c	1.58 d	2.2 e	0.23 d
Haloxyfop-R (methyl ester)	21.1 ab	5.6 a	1.95 bcd	9.9 a	1.0 a
Sethoxydim	20.4 b	5.2 ab	1.96 bc	8.8 bc	0.9b
LSD	2.47	0.72	0.37	0.71	0.07

Treatments with the same letter in a column are not significantly different at $p < 0.05$ as assessed by the LSD test

Table 8. Effect of various doses of herbicide on chlorophyll content, leaf number, plant height, shoot diameter, shoot fresh and dry weight, of *Valeriana officinalis*

Rate of application [mg a.i. · kg ⁻¹]	Chlorophyll content [spad value]	Leaf number [cm]	Shoot diameter [cm]	Shoot fresh weight [g]	Shoot dry weight [g]
0.75X (D1)	21.2 a	5.0 a	2.0 a	9.3 a	0.93 a
1X (D2)	20.8 ab	4.9 a	1.9 a	7.3 b	0.76 b
1.25X (D3)	19.4 b	4.6 a	1.8 a	6.6 c	0.69 c
LSD	1.61	0.47	0.26	0.46	0.05

Treatments with the same letter in a column are not significantly different at $p < 0.05$ as assessed by the LSD test

Table 9. Interactions of herbicides and their doses on chlorophyll content, leaf number, height, shoot diameter, shoot fresh and dry weight of *Valeriana officinalis*

Treatment	Rate of application [mg a.i. · kg ⁻¹]	Chlorophyll content [spad value]	Leaf number [plant]	Shoot diameter [cm]	Shoot fresh weight [g]	Shoot dry weight [g]
The control	–	23.5 a	4.5 bcd	2.4 abc	10.6 bc	1.04 bcd
Bentazon	D1	20.6 abc	4.5 bcd	2.0 abc	7.8 f	0.73 gh
	D2	20.8 abc	4.2 cd	2.0 abc	4.2 h	0.43 i
	D3	18.3 bcd	4.0 d	1.7 c	3.7 h	0.38 i
Oxadiargyl	D1	21.9 ab	5.0 abcd	2.6 a	9.7 cd	1.08 bc
	D2	24.3 a	5.0 abcd	2.0 abc	9.3 de	0.97 cde
	D3	17.5 cde	4.6 bcd	1.9 bc	8.4 ef	0.91 ef
Oxadiazon	D1	24.0 a	5.3 abc	1.7 c	8.9 def	0.95 cdef
	D2	18.9 bcd	5.6 ab	1.8 bc	8.0 f	0.82 fgh
	D3	20.9 abc	4.8 abcd	1.8 bc	8.0 f	0.84 efg
Oxyfluorfen	D1	15.3 de	4.6 bcd	1.5 c	3.6 h	0.34 ij
	D2	18.3 bcd	4.5 bcd	1.6 c	2.0 i	0.21 jk
	D3	13.6 e	4.0 d	1.5 c	1.0 i	0.13 k
Haloxyfop-R (methyl ester)	D1	21.3 abc	6.0 a	1.8 bc	12.7 a	1.24 a
	D2	21.0 abc	5.6 ab	2.1 abc	8.9 def	0.94 def
	D3	21.2 abc	5.1 abcd	2.0 abc	8.1 ef	0.84 efg
Sethoxydim	D1	21.5 abc	5.1 abcd	2.0 abc	11.7 ab	1.12 ab
	D2	18.8 bcd	5.0 abc	1.8 bc	8.3 ef	0.90 ef
	D3	20.7 abc	5.5 ab	2.0 abc	6.5 g	0.7 h
LSD	–	4.28	1.26	0.65	1.22	0.13

Treatments with the same letter in a column are not significantly different at $p < 0.05$ as assessed by the LSD test

Soltani *et al.* (2008) reported that application of bentazon once or twice at 840 g · ha⁻¹ had no marked effect on the shoot weight of pinto bean. An increase in the above ground dry matter of winter wheat due to the good weed control from the sethoxydim application, was noted by Bidlack *et al.* (2006) as compared with the untreated control. Qasem and Foy (2006) reported that application of oxadiazon and oxyfluorfen increased shoot dry weight of *Syrian marjoram* L. Reduction in the chlorophyll content of sunflower, maize, and other plants by oxadiargyl application has been reported (Sandmann and Boger 1983; Nethra and Jagannath 2011).

Conclusion

The previous studies and ours suggest that the affects of the used herbicides depend on herbicide, herbicide rate, and plant species. The results of this experiment showed that oxadiargyl under all three rates, and that oxadiazon at the recommended dose and at 0.25% less than the recommended dose produced no significant symptoms of phytotoxicity or reduction of measured traits. The results also showed that haloxyfop-R and sethoxydim under all three rates, when used under similar conditions as those in this experiment, may be used safely for weed control in valerian.

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