

Effect of Trifluralin (Agreflan 48% ES) Applied Pre-Plant for Weed Control in Alfalfa

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ABSTRACT

A field experiment was conducted at Shambat Research Station Farm, Khartoum, Sudan, during the 2015/2016 and 2016/2017 winter seasons to evaluate the efficacy and crop selectivity of Agreflan 48% ES (trifluralin) for weed control in alfalfa (*Medicago sativa* L.). The herbicide was evaluated at three application rates (0.63, 0.84 and 1.05 L/fed) as a pre-plant soil treatment, applied 14 days prior to sowing and either mechanically incorporated or immediately irrigated into the soil. All herbicide treatments significantly reduced weed biomass. No phytotoxic symptoms were observed on the alfalfa crop. Uncontrolled weed competition reduced total alfalfa dry matter yield by 25% and 21% in the first and second seasons, respectively. Fresh and dry matter yields were consistently higher in herbicide-treated plots than in the unweeded control, attributable to effective weed suppression. No residues of Agreflan 48% ES were detected in any alfalfa samples.

Key words: Weed Control; Alfalfa and Agreflan

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INTRODUCTION

Alfalfa (*Medicago sativa* L.) is one of the most important perennial forage legumes cultivated worldwide because of its high biomass production, excellent nutritional quality, and ability to fix atmospheric nitrogen. In Sudan, the cultivated area of alfalfa has expanded considerably during the last decade to meet the increasing demand for livestock feed and export markets. However, successful establishment and productivity of alfalfa are frequently constrained by severe weed infestation, particularly during the early stages of crop growth. Weeds compete aggressively with alfalfa for light, soil moisture, nutrients, and space, resulting in poor crop establishment, reduced forage yield, and deterioration of forage quality. Yield losses caused by weed competition depend on weed species, density, duration of interference, and environmental conditions. Previous studies reported that uncontrolled weed infestation may reduce alfalfa forage yield by more than 20–40%, especially during the establishment year (Mesbah and Miller, 2005; Wilson and Burgener, 2009). Chemical weed control has become an important component of integrated weed management programs because it provides effective, economical, and timely weed suppression compared with repeated hand weeding. Among pre-plant herbicides, trifluralin has been extensively used in many crops owing to its excellent activity against annual grasses and several small-seeded broadleaf weeds. Trifluralin belongs to the dinitroaniline group and inhibits root development and cell division of germinating weed seedlings. Because of its volatility and susceptibility to photodegradation, the herbicide should be incorporated into the soil shortly after application or followed immediately by irrigation to ensure optimum efficacy (Hassawy, 1970; Scursoni and Satorre, 1997). Several researchers have demonstrated the effectiveness of trifluralin for weed management in alfalfa and other field crops. McCordick *et al.*, (2008) reported that pre-plant incorporation of trifluralin significantly improved alfalfa establishment by reducing weed

competition. Likewise, Wilson and Burgener (2009) observed that effective early-season weed management resulted in greater forage yield and improved stand persistence. More recently, studies on integrated weed management have emphasized that combining appropriate herbicide application techniques with good agronomic practices improves weed control while minimizing crop injury and environmental risks. Although trifluralin has been widely evaluated in many countries, information regarding its efficacy and crop selectivity under Sudanese conditions remains limited. Climatic conditions, soil characteristics, and weed flora differ substantially from those reported elsewhere, making local evaluation essential before recommending the herbicide for commercial use. Therefore, the present study was conducted to evaluate the efficacy, crop selectivity, and residue status of Agreflan 48% ES (trifluralin) applied as a pre-plant soil treatment for weed control in alfalfa under Sudanese conditions.

Weed competition is considered one of the major constraints limiting successful establishment and productivity of alfalfa worldwide. During the establishment stage, weeds compete intensely with alfalfa seedlings for moisture, nutrients, light, and growing space, resulting in considerable reductions in forage yield and quality (Mesbah and Miller, 2005; Wilson and Burgener, 2009). Recent studies have further confirmed that effective early-season weed management remains essential for maximizing alfalfa biomass production and forage quality under different production systems (Jeong *et al.*, 2024). Recent advances in weed management have emphasized the integration of pre-plant herbicides with appropriate agronomic practices to improve weed suppression while minimizing crop injury and environmental risks. Extension recommendations published in recent years continue to identify pre-plant incorporated herbicides, including trifluralin, as an effective option for establishing new alfalfa stands (Adjewor and Montgomery, 2024).

Trifluralin is a dinitroaniline herbicide widely used for the control of annual grasses and certain broadleaf weeds in alfalfa and several field crops. It inhibits microtubule formation during cell division, thereby preventing root development in germinating weed seedlings. Because of its volatility, trifluralin should be incorporated into the soil immediately after application or followed by irrigation to achieve optimum efficacy (Adjesiwor & Montgomery, 2024; Cornell CALS, 2025). Therefore, this study was set to evaluate the activity and selectivity of the herbicide Agreflan 48% ES (trifluralin) for weed control in alfalfa.

MATERIALS AND METHODS

Field trials were conducted during the 2015/2016 and 2016/2017 winter seasons at Shambat Research Station Farm, Khartoum, Sudan. The experimental sites were disc-sloughed, levelled and divided into small plots measuring 2×4 m. Alfalfa (cv. Hijazi) was sown on flat ground at a seed rate of 10 kg/fed, in rows spaced 20 cm apart. In both seasons, sowing was carried out in the first week of December. Agreflan 48% ES (trifluralin) was evaluated at three application rates (0.63, 0.84 and 1.05 L/fed) as a pre-plant soil treatment.

The herbicide Trifluralin (Agreflan) was applied 14 days before sowing and mechanically incorporated into the soil before planting. In the second method, herbicide treatments were applied at different rates immediately after sowing, followed by irrigation to ensure proper herbicide activation in the soil. All herbicide treatments were applied using a knapsack sprayer calibrated to deliver 120 L/fed. Weeded and unweeded control treatments were included for comparison. In the weeded plots, weeds were removed manually at 15, 30, and 45 days after sowing, whereas the unweeded plots were left untreated throughout the experimental period.

Since trifluralin is a soil-applied herbicide with low mobility and high susceptibility to volatilization and photo decomposition, incorporation into the soil immediately after application is essential to improve herbicide availability within

the weed seed germination zone and maximize weed control efficacy.

Treatments were arranged in a randomized complete block design (RCBD) with three replicates. Visual assessments of herbicide phytotoxicity to the alfalfa crop were conducted periodically throughout the growing season.

The effects of herbicide treatments on the weed community were assessed by counting total weed numbers and individual weed species at 4 and 8 weeks after herbicide application (WAA). Counts were made using a 1 × 1 m quadrat placed randomly in each plot. Weeds within the quadrat were identified and total and species-specific weed counts were recorded. Percentage weed control for grassy and broad-leaved species was calculated relative to the unweeded treatment. Weed samples from 1 m² per plot were harvested at 80 days after sowing (DAS), air-dried and weighed to determine weed biomass.

Alfalfa was first harvested at the initial bloom stage in late February, with subsequent second and third harvests conducted at 28-day intervals. Fresh weight was recorded from the whole plot. Forage sub-samples of 0.5 kg per plot were oven-dried and weighed to determine dry matter yield.

Data were subjected to analysis of variance (ANOVA), and treatment means were separated using Duncan's Multiple Range Test (DMRT) at $P \leq 0.05$. For residue analysis, alfalfa samples from plots treated with the highest herbicide rate were collected at the first harvest. A standard extraction and clean-up procedure was applied to both treated and control samples. Analysis was performed by Thin Layer Chromatography (TLC) using plastic-backed silica gel GF₂₅₄ plates with a thickness of 0.25 mm (Merck).

RESULTS AND DISCUSSION

Effect on Weeds

The weed flora associated with the alfalfa crop consisted of both grassy and broad-leaved weed species. The dominant grassy weeds were *Echinochloa colona* (jungle rice) and *Cyperus rotundus* (purple nutsedge), while *Tribulus terrestris* (puncture vine) was the predominant broad-leaved weed recorded throughout the experimental period. Total weed densities in the weedy check were 68 and 57 plants m⁻² at 4 WAA in the 2015/2016 and 2016/2017 seasons, respectively

Grassy weeds constituted approximately 63% of the total weed population during the first season and 85% during the second season, whereas broad-leaved weeds represented the remaining proportion.

The predominance of grassy weeds during both seasons explains the high efficacy of Agreflan 48% ES (trifluralin), since trifluralin is particularly effective against germinating annual grasses and some small-seeded broad-leaved weeds. Weed species composition remained generally similar throughout the study, although their densities varied between seasons owing to environmental conditions and weed emergence patterns. In both seasons, Agreflan applied at 0.63–1.05 L/fed provided satisfactory to good control of grassy weeds (60–79%) and moderate to good control of broad-leaved weeds (50–79%) at 4 and 8 WAA (Tables 1 and 2). All herbicide treatments significantly reduced the air-dry weight of weeds compared to the unweeded check (Tables 1 and 2). These efficacy levels are consistent with previous reports on trifluralin as a pre-plant incorporated herbicide in field crops (Scursoni and Satorre, 1997).

Effect on Crop Yield and Phytotoxicity

No visual phytotoxic symptoms were observed on alfalfa plants in any herbicide-treated plot throughout either season, confirming the selectivity of trifluralin at all rates tested. Uncontrolled weed growth reduced total fresh matter yield of alfalfa by 26% and 21% in the first and second seasons, respectively, relative to the hand-

weeded control (Tables 3 and 4). Correspondingly, total dry matter yield was reduced by 25% and 22% in the two seasons (Tables 5 and 6). These yield losses underscore the economic importance of weed control in alfalfa and are consistent with competitive effects reported for weeds in legume forage crops (Mesbah and Miller, 2005).

All herbicide treatments increased total fresh and dry matter yields of alfalfa compared to the unweeded control (Tables 3–6). The absence of significant differences between most herbicide treatments and the hand-weeded control indicates that chemical weed management using trifluralin is a viable substitute for manual weeding under these conditions.

Residue Analysis

The Rf value of the trifluralin reference standard was 0.62. The minimum detectable quantity was 0.08 µg and the method recovery was 88%. No residues of Agreflan 48% ES were detected in any of the alfalfa samples collected at the first harvest. Given that the established maximum residue limit (MRL) for trifluralin in alfalfa is 1 ppm, these findings confirm that the use of Agreflan (*trifluralin*) at the tested rates does not constitute a food safety risk.

The absence of detectable trifluralin residues in alfalfa samples at the first harvest can be attributed to the physicochemical properties of trifluralin. Trifluralin is a pre-emergence dinitroaniline herbicide characterized by very low water solubility, high adsorption to soil organic matter, and limited systemic movement within plants. After soil incorporation, the herbicide remains largely confined to the upper soil layer, where it inhibits root development of germinating weed seedlings with minimal translocation into established crop tissues. Consequently, residue accumulation in harvested forage is generally very low or below detectable limits when the herbicide is applied according to recommended agricultural practices (Shaner, 2014; PPDB, 2025). Furthermore, trifluralin is susceptible to degradation in soil through microbial activity, photodegradation on exposed soil surfaces, and

volatilization, resulting in a gradual decline in its concentration over time. The interval between herbicide application and the first alfalfa harvest provides additional time for dissipation, thereby reducing the likelihood of detectable residues in harvested forage (PPDB, 2025; USEPA, 2023). The present findings are consistent with previous studies reporting that trifluralin residues in forage and field crops are frequently below analytical detection limits when the herbicide is incorporated into the soil before planting and applied at recommended rates. Similar observations have been reported in several residue monitoring studies, which concluded that proper application of trifluralin does not result in significant residue accumulation in harvested crops (FAO and WHO, 2023; USEPA, 2023)

RECOMMENDATION

Based on the results of this study, it is recommended that the National Pests and Diseases Committee approve the use of Agreflan 48% ES (*trifluralin*) at a rate of 0.84 L/fed (equivalent to 0.4 kg a.i./fed), applied as a pre-plant soil treatment 14 days prior to sowing and mechanically incorporated into the soil, for weed control in alfalfa. Agreflan 48% ES is manufactured by Jiangsu Fengshan Group Co., Ltd. and is imported into Sudan by Oranos Trading and Investment Co. Ltd.

Table 1. Effect of *Trifluralin* herbicide treatments on weed control (%) and air-dry weight of weeds (season 2015/2016)

Treatments	Herbicide rate (L./fed)	Weed control (%)				Air dry weight of weeds at 80 DAS (g/m ²)
			Broadleaved	Grasses		
		4WAA	8WAA	4WAA	8WAA	
Agreflan*	0.63	71	65	59	50	54.7 b
Agreflan*	0.84	73	60	70	53	30.5 bc
Agreflan*	1.05	70	70	73	58	38.8 bc
Agreflan**	0.63	68	68	69	48	53.0 bc
Agreflan**	0.84	70	60	75	55	35.2 bc
Agreflan**	1.05	79	70	75	50	43.8 bc
Weeded control	—	100	100	100	100	17.6 c
Weedy check	—	0	0	0	0	87.7 a
SE ±						10.57
CV%						40.5

WAA = Weeks after herbicide application; DAS = Days after sowing.

* = Applied 14 days prior to sowing and mechanically incorporated into the soil.

** = Applied 14 days prior to sowing and immediately irrigated.

Means within a column followed by the same letter are not significantly different at $P \leq 0.05$ (DMRT).

Table 2. Effect of *Trifluralin* herbicide treatments on weed control (%) and air-dry weight of weeds (season 2016/2017)

Treatments	Herbicide rate (L./fed)	Weed control (%)				Air dry weight of weeds at 80 DAS (g/m ²)
			Broadleaved	Grasses		
		4WAA	8WAA	4WAA	8WAA	
Agreflan*	0.63	74	69	75	60	58.5 b
Agreflan*	0.84	70	64	70	51	38.27 b
Agreflan*	1.05	79	70	79	70	46.5 b
Agreflan**	0.63	73	69	50	58	60.6 b
Agreflan**	0.84	69	70	54	70	66.2 b

Agreflan**	1.05	70	78	65	59	51.2 b
Weeded control	—	100	100	100	100	32.3 b
Weedy check	—	0	0	0	—	107.1 a
SE ±						32.49

WAA = Weeks after herbicide application; DAS = Days after sowing.

* = Applied 14 days prior to sowing and mechanically incorporated into the soil.

** = Applied 14 days prior to sowing and immediately irrigated.

Means within a column followed by the same letter are not significantly different at $P \leq 0.05$ (DMRT).

Table 3. Effect of herbicide treatments on fresh matter yield of alfalfa (ton/fed) — season 2015/2016

Treatments	Herbicide rate (L/fed)	Fresh yield (ton/fed)			Total fresh yield (ton/fed)
		1st cut	2nd cut	3rd cut	
Agreflan*	0.63	7.1 ab	7.5 ab	6.7	21.3 ab
Agreflan*	0.84	6.9 ab	7.2 b	5.8	19.9 ab
Agreflan*	1.05	6.4 abc	6.8 bc	5.5	18.7 bc
Agreflan**	0.63	6.1 bc	7.8 ab	6.7	20.6 ab
Agreflan**	0.84	6.9 ab	8.6 a	7.1	22.6 a
Agreflan**	1.05	6.6 ab	7.4 ab	6.0	20.0 ab
Weeded control	—	7.3 a	7.9 ab	6.5	21.7 ab
Weedy check	—	5.3 c	5.6 c	5.3	16.1 c
SE ±		0.347	0.418	NS	1.120
CV%		9.15	9.87	18.51	9.65

NS = non-significant. * and ** as defined in Tables 1 and 2.

Means within a column followed by the same letter are not significantly different at $P \leq 0.05$ (DMRT).

Table 4. Effect of *Trifluralin* herbicide treatments on fresh matter yield of alfalfa (ton/fed) season 2016/2017

Treatments	Herbicide rate (L/fed)	Fresh yield (ton/fed)			Total fresh yield (ton/fed)
		1st cut	2nd cut	3rd cut	
Agreflan*	0.63	6.0 a	7.4 ab	7.2 a	20.5 ab
Agreflan*	0.84	6.1 a	7.4 ab	5.5 a	19.0 ab
Agreflan*	1.05	5.7 a	8.2 ab	6.95 a	20.8 a
Agreflan**	0.63	5.9 a	8.2 ab	6.5 a	20.5 ab
Agreflan**	0.84	5.8 a	7.6 ab	6.95 a	20.3 ab
Agreflan**	1.05	6.1 a	9.0 a	6.6 a	21.6 a
Weeded control	—	5.6 a	7.5 ab	7.46 a	20.5 ab
Weedy check	—	4.1 b	6.2 b	5.9 b	16.2 b
SE ±		0.384	0.617	0.591	1.306
CV%		—	13.93	15.49	11.36

NS = non-significant. * and ** as defined in Tables 1 and 2.

Means within a column followed by the same letter are not significantly different at $P \leq 0.05$ (DMRT).

Table 5. Effect of *Trifluralin* herbicide treatments on dry matter yield of alfalfa (ton/fed) season 2015/2016

Treatments	Herbicide rate (L/fed)	Dry matter yield (ton/fed)			Total dry matter yield (ton/fed)
		1st cut	2nd cut	3rd cut	
Agreflan*	0.63	1.8 ab	2.1 abc	1.9 b	5.8 abc
Agreflan*	0.84	2.1 a	2.1 abc	2.0 b	6.3 ab
Agreflan*	1.05	2.2 a	1.9 abc	2.2 ab	6.0 ab
Agreflan**	0.63	1.9 ab	2.3 ab	2.0 b	6.2 ab
Agreflan**	0.84	2.0 a	2.4 a	2.6 a	7.0 a
Agreflan**	1.05	1.9 ab	1.5 bc	2.3 ab	5.6 bc
Weeded control	—	1.9 ab	2.3 a	2.2 ab	6.4 ab
Weedy check	—	1.4 b	1.5 c	1.9 b	4.8 c
SE ±		0.161	0.219	0.150	0.351
CV%		14.79	18.94	12.26	10.13

NS = non-significant * and ** as defined in Tables 1 and 2.

Means within a column followed by the same letter are not significantly different at $P \leq 0.05$ (DMRT).

Table 6. Effect of *Trifluralin* herbicide treatments on dry matter yield of alfalfa (ton/fed) season 2016/2017

Treatments	Herbicide rate (L/fed)	Dry matter yield (ton/fed)			Total dry matter yield (ton/fed)
		1st cut	2nd cut	3rd cut	
Agreflan*	0.63	1.6 a	2.1 ab	2.3	5.9 a
Agreflan*	0.84	1.7 a	2.2 ab	1.8	5.8 a
Agreflan*	1.05	1.4 a	2.3 ab	2.2	5.9 a
Agreflan**	0.63	1.5 a	2.3 ab	2.0	5.8 a
Agreflan**	0.84	1.6 a	2.3 ab	2.2	6.0 a
Agreflan**	1.05	1.7 a	2.5 a	2.0	6.1 a
Weeded control	—	1.5 a	1.9 ab	2.5	5.9 a
Weedy check	—	1.1 b	1.8 b	1.7	4.6 b
SE ±		0.107	0.170	NS	0.360
CV%		12.44	13.53	18.04	10.84

NS = non-significant. * and ** as defined in Tables 1 and 2.

Means within a column followed by the same letter are not significantly different at $P \leq 0.05$ (DMRT).

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تأثير تريفلورالين (أغريفلان 48%) عند تطبيقه قبل الزراعة لمكافحة الحشائش في البرسيم

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الملخص

أُجريت تجربة حقلية في مزرعة محطة شمبات للأبحاث، الخرطوم، السودان، خلال موسمي الشتاء 2015/2016 و 2016/2017 لتقييم فعالية مبيد الأعشاب أغريفلان 48% إي إس (تريفلورالين) وانتقائيته للمحصول في مكافحة الأعشاب الضارة في البرسيم (*Medicago sativa* L.). تم تقييم المبيد بثلاثة معدلات استخدام (0.63، 0.84، و 1.05 لتر/فدان) كمعالجة للتربة قبل الزراعة، حيث تم تطبيقه قبل 14 يومًا من البذر، إما بخلطه ميكانيكيًا مع التربة أو بريه مباشرة. أدت جميع المعالجات بالمبيد إلى انخفاض ملحوظ في الكتلة الحيوية للأعشاب الضارة. لم تلاحظ أي أعراض تسمم نباتي على محصول البرسيم. أدى التنافس غير المسيطر عليه مع الأعشاب الضارة إلى انخفاض إجمالي محصول المادة الجافة للبرسيم بنسبة 25% و 21% في الموسمين الأول والثاني على التوالي. كانت غلة المادة الطازجة والجافة أعلى باستمرار في الحقول المعالجة بالمبيد مقارنةً بالحقول غير المعالجة، ويعزى ذلك إلى فعالية كبح نمو الأعشاب الضارة. لم يتم الكشف عن أي بقايا من مادة Agreflan 48% ES في أي من عينات البرسيم.

الكلمات المفتاحية: مكافحة الحشائش؛ البرسيم و أغريفلان

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