

# An Approach to The Biological Performance of Halosulfuron Against *Cyperus Rotundus* L. in Relation to its Effect on Maize Chemical Analysis and Yield

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## ABSTRACT

Two field experiments were conducted at Agricultural Experimental Research station, Faculty of Agriculture at Giza, during 2006 and 2007 seasons. The biological performance of Halosulfuron against *Cyperus rotundus* L. and some other broad leaves were estimated. Furthermore the chemical analysis of the total chlorophyll, amino acids and protein contents as well as the maize yield were considered.

The fluroxypyr was gave an efficient results against Broad leaved weeds. Furthermore Halosulfuron at 30 g/fed. was more effective than hand weeding treatment in controlling weeds. The efficiency effects against *Cyperus rotundus* L. were 80.83 and 72% during 2006 season, and 84.62 and 70.90% during 2007 season at of 40 and 70 days after sowing (DAS), respectively. The highest increase in the maize grain yield was obtained by fluroxypyr at 20 g/fed. (2858 and 3266 kg/fed.), followed by halosulfuron a 30g/fed. (2741 and 3199 kg/fed.) in both seasons, respectively. However, there were no significant differences between the tested two rates of both herbicides. All treatments caused significant increases in the total chlorophyll contents in maize leaves determined at 40 and 70 DAS during both seasons. In addition, significant increases in the total amino acids and protein content in maize grains were recorded as compared with those results of the untreated check treatment during both seasons.

## INTRODUCTION

Maize (*Zea mays* L.) is the third most important cereal worldwide after the wheat and the rice. Today the crop is one of the most important sources for the world's food supply (FAO, 2004). Weeds causes great losses in yield, i.e. Purple nutsedge (*Cyperus rotundus* L.) which has gained the reputation as the world's weed from its global distribution, competitiveness and its high vegetatives. The purple nutsedge tubers had a half- life of 16 months and longevity of (99% mortalities) 42 months (Neeser *et al.* 1997). The sulfonylurea herbicides are the chemical group of the high level of herbicide activities that are applied at very low doses (Moberg and Cross, 1990). Halosulfuron is a sulfonylurea herbicide that can be applied as pre plant incorporated, pre-emergence and post-emergence herbicide in maize (Sikkema *et al.*, 2007). On the other hand

Halosulfuron is active at low doses, with low mammalian toxicity and has low potential to contaminate both ground water and the environment (Vencill, 2002).

Using the halosulfuron would provide maize with a new herbicide that controls some weeds including *Chenopodium album* L. (Common Lambsquarters) *Amaranthus retroflexus* L. (Redroot pigweed) and *Cyperus* sp. (nutsedge) including triazine – resistant biotypes (Vencill, 2002).

The objective of this study to through a light on halosulfuron for controlling of *Cyperus rotundus* and some broad leaved weeds. Meanwhile to determine the chemical analysis of the total chlorophyll, protein and Amino acid contents and maize yield

## MATERIALS AND METHODS

Two field experiments were conducted at Agricultural Experiment and Research Station, Faculty of Agriculture at Giza, during the two successive summer seasons of 2006 and 2007 in order to study:

- a) The effect of different rates of halosulfuron 70% WG on purple nutsedge (*Cyperus rotundus* L.) and some broad leaved weeds control
- b) Maize yield and chemical analyses of total chlorophyll, amino acids and protein content in response to applied treatments.

Maize hybrid c.v. single cross 10 (S.C.10) grains were sown on May 29<sup>th</sup> and 17<sup>th</sup> in 2006 and 2007 seasons, respectively. The experimental area was divided according to the complete randomized block design including four replicates for each treatment. Each replicate was a plot of 6 x 7 m<sup>2</sup> (1/100 feddan). Halosulfuron was applied at the rates of 15, 20, 25 and 30g a.i / fed. A standard or reference herbicide , fluroxypyr 20% EC at the recommended rate 200 ml/ fed. Was also used. Hand weeding was conducted twice, the first one was done after two weeks of sowing and the second after two weeks of the first hand weeding, unweeded check (control) was included in the test.

**Identification of weeds:** Weeds in each treatment were identified at 40 and 70 days after sowing (DAS) as they recorded in Table (1).

Percent control of weeds per each treatment was calculated according to the following formula : % control = (C-T/C) x 100

C= Weight of weeds in control

T = Weight of weeds in treatment

**Maize yield:** At harvest, grain yield (kg/fed.) was shelled and weighted compared with the yield in unweeded check.

### **Chemical analysis:**

**Chlorophyll content:** Total chlorophyll was estimated in representative samples of fresh leaves at 40 and 70 DAS. Total chlorophyll (mg/g fresh weight) was determined according to the method described by Hiscox and Israelstam (1979).

**Chlorophyll extraction:** leaves were harvested and washed for several times with tap water. Leaves were plotted on paper towels and left at room temperature until they were dry. Then, leaves were cut into very small pieces and mixed well. Thereafter, 100 milligrams sample was weighted and placed in a vial containing 7 ml dimethyl sulfoxide (DMS). Samples were left in an incubator at 65°C for 24 hours for chlorophyll extraction. The extract was transferred to a graduated tube and raised to a total final volume of 10 ml by dimethyl sulfoxide and stored at a temperature ranged between 0-4°C.

**Determination of chlorophyll content:** A 3 ml sample of chlorophyll extract was transferred to a cuvette and the optical density (OD) values at 645 and 663 nm were read in Shemadzue UV -150-02 spectrophotometer. Total chlorophyll values were calculated according to Arnon equations (1949), as follows: Total chlorophyll =  $20.0 A_{645} - 8.02 A_{663}$  where "A" is the absorbance (optical density) of chlorophyll extract.

### **Analysis of protein and amino acids :**

**Sample preparation:** At harvest, samples of air dried grains (250g) were ground into fine powder and stored in a deep freezer at -4°C for further analyses of proteins and amino acids.

**Protein determination:** According to Lowry *et al.* (1951), a weighed sample of the dried plant powder (0.01g) was added to glass tubes.

Sodium carbonate 2% (5 ml) was added to the sample and kept in shaking water bath at 37°C for 24 hours. Then, the extract was filtered through whatman No. 1 filter paper and brought to 10 ml volume with sodium carbonate 2%. From that extract, a sample of 0.5 ml was pipetted into test tube and completed to 1.0 ml with distilled water. Four milliliters of Na<sub>2</sub>CO<sub>3</sub> (2%) were added to that extract. Folin reagent (0.5 ml diluted to 1:

2 ratio with distilled water) was added to the mixture. After about 10min, when the color completed, absorbance was measured at 600 nm using spectronic 20 Lamb  $\alpha$  Bouch. Two hundred milligrams of Bovine Serum Albumin Bovine (98 -99% albumin remainder mostly globulins) were dissolved and diluted with 100 ml distilled water and kept as a stock solution.

Series of various concentrations i.e. 200 to 2000 mg were used for the standard calibration and standard curve.

**Extraction of amino acids:** Extraction of amino acids was done as described by Lee and Takahashi (1966). Weighed samples (1.0 g) were mixed with 25 ml methanol 80% in tubes and left in water bath at 70 ° for 5 hrs. or for 24 hours for full extraction at room temperature. Then, extracts were filtered through whatman No. 1 filter Paper. Filtrates were evaporated at 40-45°C under vacuum in rotary evaporator to remove methanol. The aqueous phase was transferred to 250 ml separatory funnel. A portion of saturated sodium chloride solution was added to facilitate the separation of the suspension. An equal volume of chloroform was added and shaken carefully for one minute. The procedure was repeated at least 3 times. Epiphase layer containing amino acids, was received in conical stoppered flasks. The combined epiphase was evaporated neare dryness under reduced pressure. Residues within flasks were extracted several times using 10% isopropyl alcohol to reach 10 ml in measuring flask.

### **Determiration of soluble amino acids:**

preparation of mixture ninhydrin :

- 1- Citric Buffer: A- 2.1014 g from citric acid in 100 ml distilled water  
B- 2.941 g from sodium citrate in 100 ml distilled water C- 25.5 ml of A+ 74.5 ml of B ( pH=5.4)
- 2- Ninhydrin 1%: one gram of ninhydrin was dissolved in 100 ml citric buffer.
- 3- Mixture ninhydrin: A ninhydrin mixture of citric buffer (1.0ml) + ninhydrin 1% (2.0 ml) + glycerol (6.0ml) was used.

A slight modification of Lee and Takahashi method (1966) was followed. From that ninhydrin mixture, one ml was added to a test tube containing 0.1 ml from sample extract. Then, total volume was completed to 2 ml with distilled water. The test tube was then kept in boiling water bath for 12 minutes till color development.

The colored solution was left to cool at room temperature and completed to 5 ml with distilled water and measured at 570 nm using spectronic 20 lamb and Bouch. One hundred milligrams of phenylalanine were dissolved and diluted with distilled water of final volume of 100 ml as stock solution. Series of 100, 200, 300, 400 and 500 mg phenylalanine concentrations were used for standard curve and calibration.

**Mathematical calculation:** Soluble protein and total amino acids were mathematically calculated according to Beer – Lambert Law (1989) as follows:

$$\frac{C1}{OD1} = \frac{C2}{OD2} = \frac{C3}{OD3} = K$$

Therefore,  $\frac{C}{OD}$  for each concentration of the standards were estimated.

The mean of Ks were determined which is denoted as k, the concentration of the unknowns were calculated as follows:

$$\frac{C}{OD} K = C = OD \times K \text{ amount mg / g sample} = \frac{OD \times K \times \text{dillution factor}}{\text{weight of sample (g)}}$$

**Statistical Analysis:** The data were subjected to statistical analysis of variance and the significant difference among the means were compared using Duncan multiple range test. (Duncan, 1955).

## RESULTS AND DISCUSSION

**Identification of weeds:** The dominant weeds in maize experimental plots at 40 and 70 DAS during 2006 and 2007 seasons are listed in Table (1).

**Table (1): Dominant weed species related with maize at the experimental site during 2006 and 2007 seasons.**

Species	Scientific name	Common name	Family	Arabic name
Annual	<i>Xanthium stramarium</i> L.	Common cocklebur	Compositae	Shobat (Shabka)
Annual	<i>Portulaca oleracea</i> L.	Common Purslane	Portulacaceae	Regla
Annual	<i>Corchorus olitorius</i> L.	Malta Jute Jew's mallow	Tilaceae	Elmolkia
Annual	<i>Amaranthus viridis</i> L.	Slender amaranthus	Amaranthaceae	Zorbaih el-agter
Annual	<i>Echinochloa colonum</i> L.	Junglerice	Gramineae	Abo-rokba
Perennial	<i>Convolvulus arvensis</i> L.	Field bindweed	Convolvulacea	Olleiq
Perennial	<i>Cyperus rotundus</i> L.	Purple nutsedge	Cyperaceae	Saad

### Effect of halosulfuron rates on purple nutsedge (*Cyperus rotundus* L.):

The effect of the two tested herbicides (halosulfuron and fluoxypyr) and hand weeding treatments on *Cyperus rotundus* L. at 40 and 70 DAS during 2006 and 2007 seasons are shown in Tables 2 and 3. All halosulfuron rates and hand weeding treatments significantly decreased the dry weights of *Cyperus rotundus* as compared with unweeded check.

During the season of 2006, halosulfuron at 30g/fed. gave the best weed control measurements. Its efficiency values against *Cyperus rotundus* were 80.83 and 72.00% at 40 and 70 DAS, respectively.

Meanwhile, fluoxypyr at 20 g/fed. caused an increase in *Cyperus rotundus* L. dry weights by 230.6 and 316 g/m<sup>2</sup> as compared with unweeded check (218.7 and 306.5g/m<sup>2</sup>) at 40 and 70 DAS, respectively.

During 2007, the most effective weed control against *Cyperus rotundus* was obtained by halosulfuron at 30 g/fed., which caused reduction in *Cyperus rotundus* dry weights (42.6 and 101.7 g/m<sup>2</sup>) as compared with unweeded check (277.0 and 349.5 g/m<sup>2</sup>) at 40 and 70 DAS , respectively. However, fluroxypyr at 20 g/fed. gave poor effectiveness against *Cyperus rotundus* (3 and zero%) at 40 and 70 DAS , respectively.

In this respect, Molin *et al.* (1999) reported that halosulfuron-methyl gave excellent control with greater than 80%, dry weight reduction in *Cyperus rotundus* L. weeds at all their tested rates of 36, 54 and 72 g/ha.

Halosulfuron reduced purple nutsedge tuber population by 35 to 50% when it was applied in consecutive years (Webster and Coble, 1997).

In maize crops, the most effective herbicide halosulfuron reduced the population and ground cover of purple nutsedge at rates between 32.5 and 130 g/ha. Halosulfuron has now been registered in Newzeland for control of purple nutsedge in maize (Rahman and James, 2000). Furthermore, (Barry *et al.* 2005) they found that halosulfuron provided at least 80% control of purple nutsedge shoots.

#### **Effect of halosulfuron rates on broad leaved weeds:**

All herbicides and hand weeding treatments significantly decreased dry weights of broad leaved weeds as compared with the unweeded check (Tables 2 and 3).

Generally, halosulfuron was effective on *Cyperus rotundus* and also broad leaved weeds. In contrast, fluroxypyr as a standard herbicide was only effective against broad leaved weeds.

**Table (2): Dry weights (g/m<sup>2</sup>) of *Cyperus rotundus* L. and broad leaved weeds as affected by halosulfuron rates at 40 and 70 DAS\* during 2006 season.**

Treatments	Rate (g a.i./fed.)	40 DAS				70 DAS			
		<i>Cyperus rotundus</i> (g/m <sup>2</sup> )	% control	Broad leaved weeds (g/m <sup>2</sup> )	% control	<i>Cyperus rotundus</i> (g/m <sup>2</sup> )	% control	Broad leaved weeds (g/m <sup>2</sup> )	% control
Halosulfuron	15	84.57c*	61.33	77.03b	69.53	142.2b	53.61	216.5b	48.47
	20	72.03cd	67.06	65.93bc	73.92	115.4c	62.35	179.6b	57.42
	25	55.20d	74.76	58.70bc	76.15	103.6cd	66.20	131.7c	68.72
	30	41.93d	80.83	48.70cd	80.74	85.83d	72.00	91.17d	78.38
Fluroxypyr	20	230.6a	00.00	33.30d	86.83	316.0a	00.00	75.47d	82.10
Handweeding	-	55.47d	74.64	54.50cd	78.44	113.0c	63.13	122.6c	70.93
Unweeded check	-	218.7b	00.00	252.8a	00.00	306.5a	00.00	421.7a	00.00

\*Means followed with the same letter (s) are not significantly different.

\*\* DAS = Days after sowing

**Table (3): Dry weights (g/m<sup>2</sup>) of *Cyperus rotundus* L. and broad leaved weeds as affected by halosulfuron rates at 40 and 70 DAS during 2007 season.**

Treatments	Rate (g a.i./fed)	40 DAS				70 DAS			
		<i>Cyperus rotundus</i> (g/m <sup>2</sup> )	% control	Broad leaved weeds (g/m <sup>2</sup> )	% control	<i>Cyperus rotundus</i> (g/m <sup>2</sup> )	% control	Broad leaved weeds (g/m <sup>2</sup> )	% control
Halosulfuron	15	113.6b	58.99	142.9b	55.00	153.6b	56.05	218.8b	42.75
	20	95.30bc	65.60	123.3b	61.01	133.1bc	61.92	190.8bc	50.08
	25	65.37d	76.40	77.13c	75.71	123.0bc	64.78	136.6d	64.26
	30	42.60e	84.62	53.70c	83.09	101.7d	70.90	97.83e	74.46
Fluroxypyr	20	268.6a	3.00	22.63d	92.87	356.8a	00.00	71.27e	81.35
Handweeding	-	83.87cd	69.73	67.07c	78.88	126.0bc	63.95	148.8d	61.07
Unweeded check	-	277.0a	00.00	317.5a	00.00	349.5a	00.00	382.2a	00.00

\*Means followed with the same letter (s) are not significantly different.

Halosulfuron at 30 g/fed. was more effective than hand weeding treatment in controlling weeds in maize field after 40 and 70 DAS during both seasons.



During 2006, highest weed control efficiencies were achieved by fluroxypyr at 20 g/fed. (86.83 and 82.1%) followed by halosulfuron at 30 g/fed. (80.74 and 78.38%) at 40 and 70 DAS, respectively. In contrast, lowest weed control efficiencies were noticed for halosulfuron at 15 g/fed. (69.53 and 48.47%) at 40 and 70 DAS respectively.

During 2007, the same trend was noticed for the weed control treatments. Fluroxypyr at 20g/fed. showed excellent control of the broad leaved weeds (92.87 and 81.35%) at 40 and 70 DAS, respectively, followed by halosulfuron at 30 g/fed.(83.1 and 74.46%), respectively. Whereas, halosulfuron at 15 g/fed. gave the lowest values (55.0 and 42.75%) at 40 and 70 DAS, respectively.

These results are in accordance with those reported by El-Metwally (2002) who showed that fluroxypyr at 0.2 l/fed. gave best control of broad leaved weeds (98.3 and 98.1%) at 50 DAS in 2000 and 2001 seasons, respectively. The registration of halosulfuron would provide maize producers with a new herbicide that controls troublesome weeds including *Cyperus* sp. (nutsedge) (Vencill 2002).

#### **Total chlorophyll content :**

The effect of herbicides and hand weeding treatments on total chlorophyll content in maize leaves at 40 and 70 DAS during both seasons are presented in Table (4). It is evident that the highest total chlorophyll content (3.54 and 4.14 mg/g f.w.) was achieved by fluroxypyr at 20 g/fed. at 40 DAS in 2006 and 2007 seasons, respectively.

However, at the later sampling date (70 DAS), halosulfuron at 30g/fed. gave the highest value of 4.15 mg/g f.w in 2006 season. Whereas, the highest value (4.24 mg/g f.w) was obtained by fluroxypyr at 20g/fed. in 2007 season. In contrast, lowest total chlorophyll values (2.15, 2.34, 2.27 and 2.58 mg/g f.w.) were recorded by unweeded check at 40 and 70 DAS during both seasons, respectively. These results are in line with those obtained by Tollenaar *et al.* (1994) and Ahmed (1999). They reported that chlorophyll content was higher in weed infested area and that could be due to the presence of those weeds that put maize grown under high weed competition pressure. On the contrary, unweeded control recorded the lowest values.

**Table (4): Effect of halosulfuron rates on total chlorophyll content (mg/g f.w) in maize leaves at 40 and 70 DAS during 2006 and 2007 seasons.**

Treatments	Rate (g a.i./fed.)	40 DAS		70 DAS	
		2006	2007	2006	2007
Halosulfuron	15	2.75c	2.87d	3.10c	2.95de
	20	2.87c	3.15c	3.32bc	3.46bc
	25	3.16b	3.54b	3.59b	3.86ab
	30	3.27b	4.04a	4.15a	4.21a
Fluroxypyr	20	3.54a	4.14a	4.11a	4.24a
Handweeding	-	3.14b	3.19c	3.45b	3.32cd
Unweeded check	-	2.15d	2.34e	2.27d	2.58e

\*Means followed with the same letter (s) are not significantly different.

### Grain yield:

Grain yield was significantly affected by herbicides and hand weeding treatments as compared with unweeded check in both seasons (Table 5) Highest increases in maize grain yield were obtained with fluroxypyr at 20 fed. (2858 and 3266 kg/fed.), followed by halosulfuron at 30g/fed. (2741 and 3119 kg/fed.) in 2006 and 2007 seasons, respectively However, there were no significant differences between halosulfuron at 30 g/fed. and fluroxypyr at 20 g / fed. in both seasons. Halosulfuron at 30g/fed. gave maize grain yield of 2741 and 3119 kg/fed. and yield was more than that of hand weeding (2593 and 2895 kg/fed) in both seasons, respectively. These findings are in accordance with those obtained by Yehia *et al.* (1992) and El-Metwally *et al.* (2001) who found that fluroxypyr caused significant increase in maize grain yield. The superiority of fluroxypyr in producing high grain yield may be due to its high efficiency in controlling broad spectrum of weeds which reduced their competition with maize (El-Metwally, 2002). Increased maize grain yield as affected by halosulfuron was reported by Justin *et al.* (2007) who found that halosulfuron treatment at an application rate of 53 g/ha. increased yield relative to weed control. Mahadi *et al.* (2007) recorded that the grain yield of maize was significantly increased by all treatments except cinosulfuron and the weedy check in both seasons.

**Table (5): Grain yield (Kg/fed.), total amino acids (mg/g d.w) and protein (%) contents in maize grains as affected by halosulfuron rates during 2006 and 2007 seasons.**

Treatments	Rate (g.a./fed.)	Grain yield (kg/fed)		Amino acid (mg/g.dw)		Protein (%)	
		2006	2007	2006	2007	2006	2007
Halosulfuron	15	2054c	2160e	2.75b	3.19cd	9.06d	9.45e
	20	2289bc	2538d	3.02b	3.67abc	9.69cd	9.85d
	25	2554ab	2823c	3.13b	3.47bc	10.20bc	10.91b
	30	2741a	3119ab	3.66a	3.87ab	10.82ab	11.06ab
Fluroxypyr	20	2858a	3266a	3.78a	4.03a	11.08a	11.59a
Handweeding	-	2593ab	2895bc	2.98b	3.44bcd	9.97c	10.49c
Unweeded check	-	1695d	1865f	2.20c	2.97d	8.14e	8.95f

\*Means followed with the same letter (s) are not significantly different.

### Amino acids content:

It is worthy to note that all treatments caused significant increases in total amino acids in maize grains as compared with unweeded check through both seasons (Table 5). Highest amino acids values (3.78 and 4.03 mg/g d.w) were given by fluroxypyr at 20 g/fed., followed by halosulfuron at 30 g/ fed. (3.66 and 3.87 mg/g d.w) in 2006 and 2007 seasons, respectively. On the other hand, lowest values in total amino acids (2.20 and 2.97 mg/g d.w.) were obtained by unweeded check during both seasons, respectively.

### Protein content:

The obtained results (Table 5) showed significant increases in protein content in maize grains as affected by different treatments as compared with unweeded check during both seasons.

Highest values of protein percentage (11.08 and 11.59%) were given by fluroxypyr at 20 g/fed., followed by halosulfuron at 30 g/fed. (10.82 and 11.06%) and halosulfuron at 25 g/fed. (10.20 and 10.91%) in 2006 and 2007 seasons, respectively. On the contrary, unweeded check recorded the lowest values (8.14 and 8.95%) in both seasons, respectively. These results are generally in agreement with those obtained by Ahmed (1999) and El-Metwally *et al.* (2001) reported that protein percentage in maize grains significantly increased by different weed control treatments compared to unweeded treatment.

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## المخلص العربي

التأثير البيولوجي الفعال لمبيد الهالوسلفيرون ضد حشائش السعد وعلاقته بالتأثير على تحليل نبات الذرة والمحصول

ناجح عبد النور خليل

المعمل المركزي للمبيدات - مركز البحوث الزراعية - الدقي - الجيزة - مصر

أجريت تجربتين حقليتين بمحطة البحوث والتجارب الزراعية بكلية الزراعة - الجيزة في موسمي 2006 ، 2007 لدراسة : تأثير المعدلات المختلفة (15، 20 ، 25 ، 30 جم/فدان) من مبيد هالوسلفيرون 70% WG. في مكافحة حشيشة السعد وبعض الحشائش عريضة الأوراق ، وإنتاجية محصول الذرة الشامية والمحتوى الكلي للكلورفيل في الأوراق ومحتوي الحبوب من الأحماض الأمينية الكلية والبروتين. وقد أوضحت النتائج أن مبيد هالوسلفيرون كان له تأثير فعال في مكافحة حشائش السعد والحشائش عريضة الأوراق أيضاً ولكن مبيد فلوكسيبير 20% EC ( المستخدم كمبيد قياسي ) كان له

تأثير فعال في مكافحة الحشائش عريضة الأوراق فقط . وكانت المعاملة بمبيد هالوسلفيرون بمعدل 30 جم/ فدان أكثر كفاءة في مكافحة الحشائش بالمقارنة مع معاملة النقاوة اليدوية وكانت نسبة المكافحة لحشيشة السعد هي 80.83 و 72% في موسم 2006 ، 84.62 و 70.90% في موسم 2007 وذلك بعد 40 ، 70 يوم من الزراعة على التوالي. أما المعاملة بمبيد فلوكسبير بمعدل 20 جم/فدان أعطت أعلى النتائج في محصول حبوب الذرة (2858 و 3266 كجم/فدان) يليها المعاملة بمبيد هالوسلفيرون بمعدل 30جم/فدان (2741 و 3199 كجم/فدان) في كلا الموسمين على التوالي. بالرغم من هذا فإنه لا توجد فروق معنوية في محصول الحبوب لهاتين المعاملتين. أعطت جميع المعاملات زيادة معنوية في محتوى الأوراق من الكلورفيل الكلي بعد 40 ، 70 يوم من الزراعة في كلا الموسمين . كما أدت أيضا جميع المعاملات إلى حدوث زيادة معنوية في محتوى الحبوب من الأحماض الأمينية الكلية والبروتين بالمقارنة مع معاملة الكنترول في كلا الموسمين.