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Effect of water stress, chemical and organic fertilizers on yield and yield components of rosemary (*Rosmarinus officinalis* L.)

Mojtaba HASSANZADEHDELOUEI (✉), Ahad MADANI, Ali MOGHADASFAR

Department of Agronomy, Gonabad Branch, Islamic Azad University, 9691655675, University St., Basij Square, Gonabad, Khorasan- E- Razavi, Iran

✉ Corresponding author: hassanzadehdluie@gmail.com

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ABSTRACT

To improve crop productivity under water stress conditions, understanding the interaction between fertilizers and water stress is very important for the proper management of fertilizers consumption. The present study was aimed at investigating the impact of water stress, and chemical and organic fertilizers on yield and yield components of rosemary (*Rosmarinus officinalis* L.). The experiment was carried out in Ferdows, Iran in 2015 in the form of split plots based on a randomized complete block design. 50% and 80% of water requirements were allocated to the main plots. Five kinds of fertilizers including organic fertilizer before planting (F_1), foliar application of Macromix Gold organic fertilizer (F_2), NPK chemical fertilizer before planting (F_3), and foliar application of NPK chemical fertilizer after planting (F_4), and no fertilizer (control, F_5) were allocated to subplots. Results indicate that in conditions of 80% water requirement, fertilizer treatments of F_2 and F_4, and in conditions of 50% water requirement, fertilizer treatment F_4 had the highest percentage of essential oil (more than 2%). Concerning dry matter, in both conditions of 50% and 80% water requirement, all fertilizer treatments had more dry matter than the control. Results demonstrated that in terms of achieving the maximum percentage of essential oil in conditions of 80% of water requirement, only chemical fertilizer as a foliar application (F_4) is recommended, but in the case of 50% of water requirement, foliar application of Macromix Gold organic fertilizer (F_2) can be a suitable alternative to chemical fertilizer (F_4). Considering the achievement of maximum dry matter in both 80% and 50% treatments, organic fertilizer before planting (F_1) is an economically and environmentally appropriate treatment.

Keywords: rosemary, water stress, chemical, and organic fertilizers, irrigation

INTRODUCTION

Rosemary (*Rosmarinus officinalis* L.) is a perennial plant of the Labiate (Mint) family (Dehghani and Bidgoli, 2018). It is a medicinal and aromatic plant that can also be used as a large evergreen shrub in urban gardening. Like other crops and medicinal plants, the most important factor limiting the yield of this plant is water deficiency (Wang et al., 2021). One of the most effective ways to reduce the damage caused by water stress is to add organic fertilizers to the soil. In recent decades, chemical inputs in agricultural lands have caused many environmental problems, including pollution of water resources, a decline in the quality of agricultural products, and a reduction in soil fertility. As a result, the use of biological fertilizers such as vermicompost, organic fertilizers made from cow manure and poultry manure with some organic matter, and also organic fertilizers based on amino acids extracted from brown algae have been considered (Mohammadpour et al., 2019). Organic fertilizers have high porosity, high absorption, and storage capacity of water and nutrients, proper ventilation and drainage, and high water storage capacity and its use in sustainable agriculture, in addition to increasing the population and activity of beneficial soil microorganisms (such as mycorrhiza fungi) and phosphate-solubilizing microorganisms), provide the nutrients needed by the plant, such as nitrogen, phosphorus, and soluble potassium, and improve the growth and yield of crops (Mohammadzadeh and Pirzad, 2021).

Therefore, organic fertilizers provide nutrients for plants in water stress conditions by increasing the amount of soil humus, in addition, they increase the storage capacity of soil water (García-Caparrós et al., 2019), while chemical fertilizers, not only do not have this capability but their consumption under water stress conditions should be reduced because they intensify the effect of stress on the plant (Tamadon and Riasat, 2021). Therefore, the purpose of this study is to investigate the possibility of replacing chemical fertilizers with organic fertilizers in rosemary cultivation. It is assumed that, at least under water stress, organic fertilizers can completely replace the use of chemical fertilizers.

MATERIAL AND METHODS

This experiment was carried out in 2015 in Iran, South Khorasan province, Ferdows city (Longitude: 58.1722075 Latitude: 34.0227698 Elevation: 1281m) in a field with loamy soil (pH=7.88, Ec=2.44 ds/m) and irrigation water EC = 8 ds/m. The altitude of the site was 1361 meters above sea level. The average rainfall was about 115.1 mm per year. The average annual maximum and minimum temperatures were 41.8 and -4.7 °C, respectively. The experiment was performed in the form of split plots based on a randomized complete block design (RCBD) with 3 replications and 10 treatments. Irrigation treatment at two levels of 50% and 80% water supply (FAO Penman-Monteith, 1990) allocated in the main plots and organic and chemical fertilizers including Organic fertilizer before planting (F_1): 30 % cow manure, 30% poultry manure, and 40% some plant organic matter. Macromix Gold commercial organic fertilizer as foliar application (F_2): 9% nitrogen, 6% phosphor, 8% potassium, 0.5% Iron, 0.5% Zinc, 0.2% Mn, 0.1% Cu, 0.2% Mg.NPK chemical fertilizer before planting (F_3): 40% nitrogen; 20% phosphorous; 20% potassium; NPK chemical fertilizer as foliar application after planting (F_4): 40% nitrogen; 20% phosphor; 20% potassium; Control without fertilizer (F_5), were allocated to subplots.

Organic manure is pelleted manure and a combination of cow manure, poultry manure, and, some plant organic matter. This fertilizer was applied before planting at a rate of 2.5 tons per hectare. Macromix Gold is an organic fertilizer based on amino acids extracted from brown algae, which in addition to having the essential amino acids required by the plant, contains the main elements nitrogen (9%), phosphorus (6%), and potassium (8%). This fertilizer was applied as a foliar spray at a rate of two liters per hectare one month after planting. Back spray was used for foliar application. NPK fertilizer contains the elements nitrogen (40%), phosphorus (20%) and potassium (20%) in equal proportions, and also contains a variety of trace elements such as iron (0.5%), zinc (0.5%), magnesium (0.2%), amino acids, barium and copper (0.1%). In this study, this fertilizer was used in two ways: before planting in soil and as a foliar spray at the first flowering stage. The rate of NPK fertilizer application was 100 kg/ha.

This study was conducted on land with an area of 600 meters with dimensions of 60 by 10 meters. To experiment, first, the land was plowed and after leveling, 30 plots with dimensions of 3 m in length and 2 m in width were prepared in three replications. The distance between the plots was one meter and the distance between the replicates was two meters. Irrigation frequency was based on total available soil water (TAW) and water soil depletion. Crop water requirement during the growing season was determined based on evapotranspiration (ETP). Reference ETP (ETP0) was measured using a class" A" evaporation pan. ETP0 was then multiplied by the water stress coefficient (Ks) and the crop coefficient (Kc) to calculate crop evapotranspiration (ETP crop). To create water stress according to 50%, and 80% water requirements, plots were irrigated after depletion of 50% and 20% of available soil water content (AWC). Soil water content (SWC) was measured daily by using four granular matrix sensors (Watermark Soil Moisture Sensors, Irrometer Co. Inc., Riverside, CA), installed in the soil at 20 cm intervals, up to 80 cm deep. The furrow-ridge method was used for planting. The width of the ridges was 40 cm, the width of the furrows was 30 cm, the depth of the furrow was 20 cm and the distance between the plants was 60 cm.

After the initial tillage, chemical fertilizer and livestock manure were added to each plot at the same time as the land was prepared. The first irrigation was performed before planting the cuttings on May 15, 2015. Then the cuttings were planted on May 25. The density of plants was 50,000 plants per hectare (30 plants per plot). Harvest was done when the plant was flowering. The first harvest was done three months after planting (August 25) and stem diameter (mm), canopy area (cm^2), leaf length (cm), number of sub-branches, fresh and dry weight of an average of 10 plants per plot (g) were measured. The Canopy area was measured by the methods described by Torne et al. (2002). To measure the amount of essential oil, the branch was harvested with a length of 30-35 cm, and after drying in the shade, it was extracted using a distillation device with water (Clevenger) and dehydrated using sodium sulfate without water.

In this device, the plant sample is placed in a balloon containing water and after reaching the boiling point, the steam from the plant is directed to the condenser after cooling, the steam is converted into liquid during condensation and entered into the collection container. Then, the essential oil of the sample was directed to the top of the tube due to its lower density and was separated by a syringe. The amount of branch used was 100 g and essential oil was collected within 4 hours.

Data were analyzed using SAS (Ver. 9.4) software (GLM Procedure) and the means were compared with Duncan's multiple range test (alpha=5%). To compare the means of interactions, the slicing method was used in SAS software and the graphs were drawn with Excel (2019) software.

RESULTS AND DISCUSSION

Leaf length in treatment of 80% and 50% of water requirement was significantly different (Table 1). In 80% water requirement treatment, no difference in leaf length was observed between fertilizer treatments, but in 50% water requirement treatment, F_1 fertilizer treatment compared to the control treatment, i.e. no fertilizer application (F_5) significantly increased leaf length (Figure 1). The first noticeable effect of water stress on plants can be detected by the smaller leaf size and height of plants. In addition, in stress conditions, nutrient uptake is reduced and therefore leaf growth and development are limited. Following a decrease in leaf area, light absorption and then the total photosynthetic capacity of the plant are reduced. By limiting photosynthetic products under water deficit conditions, plant growth and ultimately yield are reduced (Sarmoum et al., 2019).

Water stress decreased the amount of essential oil (Table 1). It seems that in more favorable moisture conditions, the plant has a better opportunity to absorb nutrients and convert them into aromatic and secondary metabolic substances, which leads to an increase in the amount of essential oil. In 80% water requirement, fertilizer treatments F_2 and F_4, and in 50% water requirement treatment, F_4 fertilizer treatment had the highest percentage of essential oil (more than 2%) (Figure 1). The use of organic fertilizer made from algae compared to chemical fertilizer increased the percentage of essential oil and leaf area of rosemary (Tawfeeq et al., 2016).

Consumption of 150 kg of nitrogen and 100 kg of phosphorus per hectare increased the yield of rosemary essential oil from 146 to 344 liters per hectare (Puttanna et al., 2010). A decrease in dry matter due to an increase in water stress was observed in all fertilizer treatments. If we consider the value of Kc = 1 in calculating the water required for rosemary, increasing the irrigation cycle from 4 to 8 days did not cause a difference in rosemary yield (Álvarez-Herrera et al., 2010). Reduction of essential oil due to increasing water stress was observed only in organic fertilizer treatments, i.e., fertilizer treatments F_1, and F_2. This result shows that lack of irrigation reduces the amount of dry matter more severely compared to the percentage of essential oil (Figure 1). Under drought stress, dry weight decreased from 34.49 to 21.38 (g/plant) and the essential oil percentage increased from 0.62 to 0.72 (La Bella et al., 2021). It seems that due to better root development in more favorable moisture conditions and subsequent absorption of more water and nutrients, the shoots develop better and with the increase of photosynthetic tissues, more nutrients are produced and stored, which directly causes an increase in dry weight. However, in the conditions of supplying 50% of the required water due to disturbance of root growth, all activities of the plant are affected and ultimately lead to less dry matter production. Shoot dry weight is reduced both by a decrease in vegetative growth rate and by a decrease in photosynthesis.

A decrease in vegetative growth rate and dry weight is due to osmotic processes due to a reduction in cell turgor in water deficit conditions (Mahewish et al., 2021). In terms of dry matter, in both stress treatments (80% and 50%), all fertilizer treatments showed a significant advantage over the control (F_5), with the difference that in 50% treatment, fertilizer treatments F_1, F_2, And F_4 as superior treatments did not differ significantly. These results suggest that in terms of achieving the maximum percentage of essential oil, in the conditions of supplying 80% of the water requirement, only chemical fertilizer treatment by foliar application (F_4) is recommended however, at 50% water requirement, Macromix Gold organic fertilizer by foliar application (F_2) can be a suitable alternative to chemical fertilizer (F_4).

In terms of achieving maximum dry matter yield, in 80% and 50% treatments, organic fertilizer before planting (F_1) is an economically and environmentally affordable treatment) Figure 1). Vermicompost was no different in terms of the growth of fennel shoots compared to chemical fertilizers containing manganese, iron, magnesium, nitrite, and ammonium, but the root growth rate decreased from 0.042 to 0.028 g / day.

Table 1. ANOVA and mean squares for the effect of irrigation regimes and fertilizer on some studied traits in rosemary

Essence Percentage %	Canopy Size cm^2	Dry matter g/m^2	Fresh Wight g/m^2	Leaf Length mm	Number of Branches	Stem Diameter mm	df	
NS	NS	NS	NS	*	NS	NS	2	Block
**	**	**	*	**	NS	NS	1	Irrigation (I)
**	**	**	**	*	**	**	4	Fertilizer (F)
**	*	*	*	**	**	**	1	I × F

*,** significance level at alpha=5% and alpha=1% respectively

Irrigation Regimes							
1.90 [a]	108.03 [a]	482.400 [a]	1226.00 [a]	32.52 [a]	7.22 [a]	14.13 [a]	0.8 ETP
1.64 [b]	86.55 [b]	357.133 [b]	907.33 [b]	32.36 [b]	6.94 [a]	13.43 [a]	0.5 ETP

0.5 ETP and 0.8 ETP: 50% and 80% of water requirement, respectively
Similar letters in each column indicate no significant differences between treatments

Fertilizers							
1.43 [d]	100.28 [ab]	443.33 [a]	1113.33 [a]	34.41 [a]	7.39 [a]	14.58 [a]	F_1
2.12 [b]	106.47 [a]	447.67 [a]	1136.67 [a]	32.09 [b]	7.13 [a]	14.12 [ab]	F_2
1.62 [c]	95.52 [b]	445.33 [a]	1140.01 [a]	31.74 [b]	7.31 [a]	13.65 [b]	F_3
2.21 [a]	100.94 [ab]	435.00 [a]	1121.67 [a]	32.75 [ab]	7.63 [a]	13.85 [ab]	F_4
1.46 [d]	83.24 [c]	326.5 [b]	821.67 [b]	31.22 [b]	5.95 [b]	12.70 [c]	F_5

Organic fertilizer before planting (F_1) – Macromix Gold organic fertilizer as a foliar application (F_2) - NPK chemical fertilizer before planting (F_3) - NPK chemical fertilizer as a Foliar application after planting (F_4) and control without fertilizer (F_5)

[a,b,c] Different letters in each column indicate significant differences between treatments

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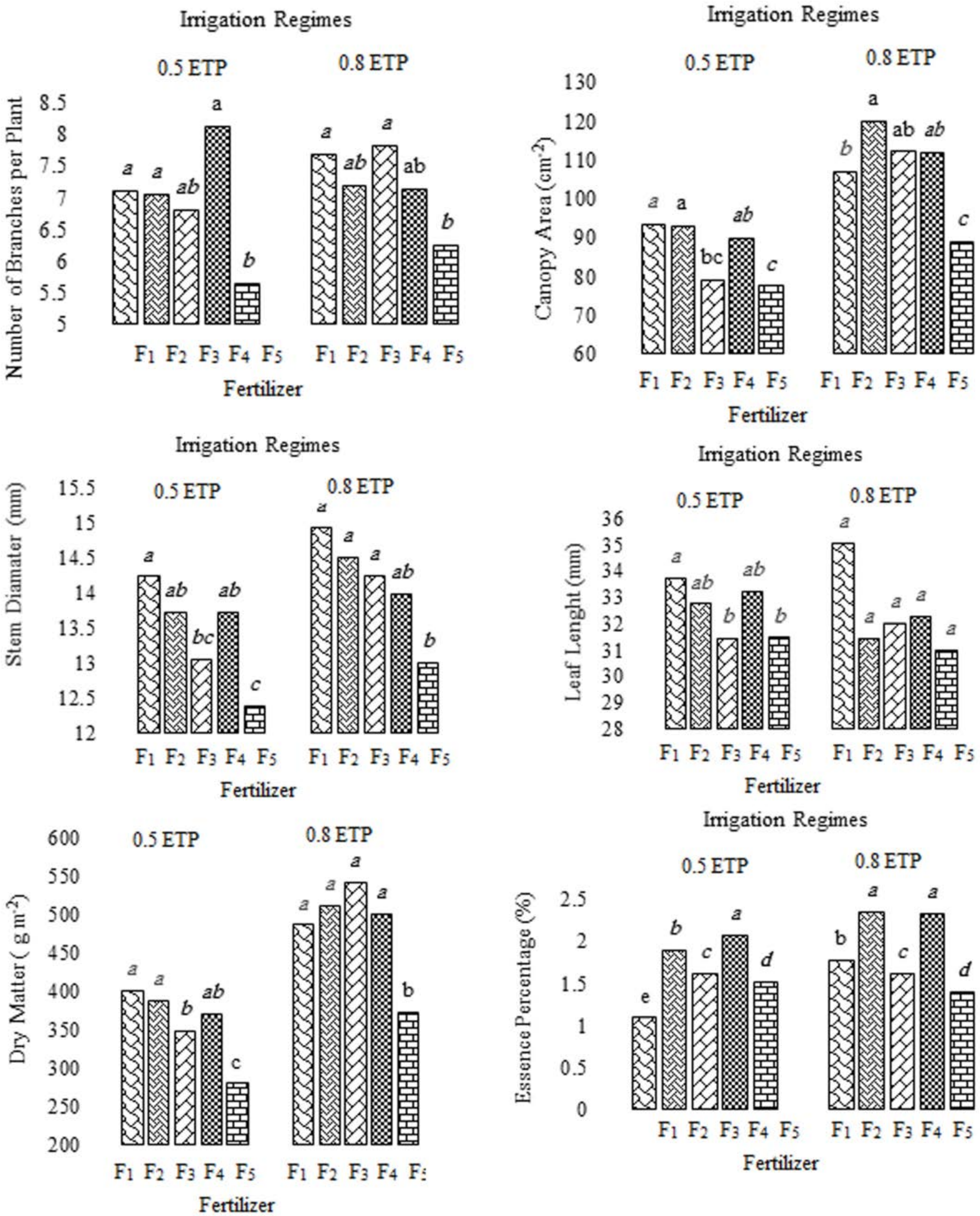

Figure 1. Mean comparisons for the interaction effects of irrigation and fertilizers on some studied traits in rosemary. Organic fertilizer before planting (F_1) – Macromix Gold organic fertilizer as a foliar application (F_2) - NPK chemical fertilizer before planting (F_3) - NPK chemical fertilizer as a Foliar application after planting (F_4) and control without fertilizer (F_5). 0.5 ETP and 0.8 ETP: 50% and 80% of water requirement, respectively.

There was no difference between the two fertilizers in terms of morphological traits and leaf chlorophyll content (Loera et al., 2021).

In terms of canopy area, in 80% water requirement treatment, the difference between all fertilizer treatments and fertilizer-free control (F_5) was significant, in 50% water requirement treatment, all fertilizer treatments except F_3 with fertilizer-free control were significant (Figure 1). Due to the direct relationship between the area of the canopy and the rate of development of leaves and aerial parts, it is expected that with the further development of the leaf area, the area of the canopy will increase, which indicates this issue. In general, the canopy area is a function of leaf expansion, and when the plant is exposed to water stress, the leaf area decreases due to a slower rate of leaf photosynthesis and assimilation transfer to them, and usually, the rate of leaf aging exceeds their rate of expansion and this significantly reduces the canopy area (La Bella et al., 2021).

In terms of the number of sub-branches, in the treatment of 80% of water requirement, fertilizer treatments F_3 and F_1 and in 50% of water supply, fertilizer treatments F_1, F_2, and F_4 had a significant difference from the control (Figure 1). Drought stress had little effect on the number of main branches (about 15.3 branches per plant) but reduced the sub-branches from 17.63 to 10.61 per plant (La Bella et al., 2021). Excessive branching under water stress conditions is considered an undesirable trait because it causes wasteful use of soil moisture and its loss, and limiting branching under water stress conditions is considered an adaptation mechanism by which the plant tries to conserve water for more critical growth stages such as flowering. Stem diameter indicates vascular development in the plant. In terms of stem diameter, in 80% of water requirements, the difference between fertilizer treatments F_1, F_2, and F_3, and in 50% of water requirements, all fertilizer treatments except F_3 were significant (Figure 1).

CONCLUSION

These results suggest that in terms of achieving the maximum percentage of essential oil, in 80% of the water supply only chemical fertilizer treatment as a foliar application (F_4) is recommended, but in 50 of the water supply, Macromix Gold organic fertilizer as a foliar application (F_2) can be a suitable alternative to chemical fertilizer (F_4). In terms of achieving maximum dry matter yield, in both 80% and 50% water supply, pre-planting organic fertilizer (F_1) is an economically and environmentally sound treatment. These results suggest that in terms of achieving the maximum percentage of essential oil, in 80% of the water supply only chemical fertilizer treatment as a foliar application (F_4) is recommended, but in 50% of the water supply Macromix Gold organic fertilizer as a foliar application (F_2) can be a suitable alternative to chemical fertilizer (F_4). In terms of achieving maximum dry matter yield, in both 80% and 50% water supply, pre-planting organic fertilizer (F_1) is an economically and environmentally sound treatment.

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