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Research Article

The Effect of Microbial Fertilization and Phosphorus Doses on the Chlorophyll Content of Sesame (*Sesamum indicum L.*)

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Abstract

In this aim of this study was to determine the effect of microbial fertilization and P2O5 doses on the chlorophyll content of sesame (*Sesamum indicum* L.) leaf by using chlorophyll meter. Sesame (*Sesamum indicum* L.) crop was cultived for 113 days under the field typifying semiarid properties localed in Konya plain, Turkey. The clorophyll content of sesame (*Sesamum indicum* L.) leaves was measured between flowering period and harvesting stage. The average time to the first capsule formation was calculated as 45 days in the the parcels treated with microbial fertilization, while the one treated with P2O5 doses was calculated as 47 days. At the end of the experiment, although microbial fertilizer application increased chlorophyll content in sesame leaves, it was observed that P2O5 doses increased chlorophyll content with increasing doses. Additionally, it was found that chlorophyll content in sesame leaves fastly increased during capsule formation period with microbial fertilization.

Keywords: Chlorophyll content , Chlorophyll Meter, Semiarid, *Sesamum indicum* (L.).

1. Introduction

Although the gene centre of sesame, which is an ancient culture plant, is not well known, its origin is pointed to Africa because two thirds of its species and economic importance lies in Africa (Nayar, 1984). Secondary gene centre of sesame, which is positioned in Turkey, has been stated to spread in West Asia via India, China and Japan (Yilmaz et al. 2005; Arıoğlu 2007). Sesame, as an annual oil plant belonging to *pedaliaceae* family, has a diploid structure with 26 (2n = 2x = 26) number of chromosomes. Their seeds contain about 50-60 % oil and are mostly used in bagel and pastry processing, as well as production of tahini and halva (Tulukcu 2013). It requires a high rate of manual labour during harvesting and blending. Sesame production has not reached the desired level as there is not enough of its cultivation in Turkey. According to 2016 data, the world produced 6.1 million tons of sesame from 10.6 million ha, yielding around 578 kg per ha.The world sesame production records present 50 % in Asia, 43.7 % in Africa and 3.8 % in the Americas. The largestshare in world sesame production is in India, Sudan, Myanmar, China and Nigeria (Anonymous 2018).

The chlorophyll content in the leaf indicates physiological status of plant. Chlorophyll is necessary to convert the light energy into chemical energy and There are two types of chlorophyll in the leaves of plant namely chlorophyll a and chlorophyll b. The amount of radiation absorbed from the sun is related to the photosynthetic product and activity of the leaf (Tulukcu 2013; Curran et al., 1990). The level of leaf chlorophyll content depends on the stress and ages of the plant (Hendry et al., 1987).

Microbial fertilization is one of the alternative methods for re-establishing the balance between plant and soil microorganisms. Microbial fertilization is the process of natural microorganism's reproduction through being supplied to plants in a suitable formulation (Yonsel and Batum 2007). Some soil microorganisms simultaneously provide various benefits to plants as colonized in both plant roots and rhizosphere (Harley and Smith, 1983). Microorganisms colonized in the roots, especially some fungi, increase the root area of the plant and facilitates the intake of water and nutrients (Sylvia 1999). Because of this increase in nutrient content, the

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plant flourishes better and shows tolerance against biotic and abiotic stress factors such as drought, salinity, heavy metals and soil pathogens (Sylvia and Williams 1992). *Bacillus spp*., *Azotobacter spp*., *Trichoderma spp*., *Rhizobium spp*., *Azospirillum spp*. and *Saccharomyces spp*. are generally known as useful microorganisms. Some microorganisms colonized in the root which induc resistance to abiotic stresses, promote nutrients intake and use as well as increase photosynthesis photosynthesis (Inbar et al., 1994; Yedidia et al. 2001, Harman et al., 2004, Harman 2006). The beneficial bacteria located in the root zone are called Plant Growth Promoting Rhizobacteria (PGPR) (Saharan and Nehra 2011). Among them are *Pseudomonas*, *Azospirillum*, *Azotobacter*, *Klebsiella*, *Enterobacter*, *Alcaligenes*, *Arthrobacter*, *Burkholderia*, *Bacillus* and *Serratia*. PGPRs supports root and shoot growth in early development stage of the plants in which they are inoculated and also enhance the effect of biomass.

In general, fertilization is used to provide the required phosphorus by the plants to having low amount beneficial phosphorus. Difficulties are encountered not only in the use of phosphorus fertilizer but also in its production. Alkaline soil pH and high lime content are among the causes of lower phosphorus availability to plant . In general, plants can benefit from only 5 - 25% of the phosphorus added to the soil.

As an alternative to phosphorous fertilization, some soil-borne microorganisms have the ability to dissolve phosphorus and it was stated that if such microorganisms are produced in pure form and inoculated into plant growth media will meet the plant phosphorus requirement by dissolving phosphorus into highly soluble phosphorus in the rhizosphere. Microbial fertilization with phosphorus dissolving microorganisms is a cheaper and environmentally less risky method compared to the addition of phosphorus fertilizer. In this study, the effect of microbial fertilization with different doses of phosphorus on the chlorophyll content in sesame (*Sesamum indicum* L.) crop was investigated.

2. Material and Methods

The research was established in the Akoren district located in the Central Anatolia within the boundaries of Konya typifying semi-arid features, with four replications according to the Randomized Blocks Trial Design during 2018 vegetation period of sesame (*Sesamum indicum* L.) cultivation. A local sesame (*Sesamum indicum* L.) seeds variety were used as test crops Three different doses of phosphorus P_2O_5 (0, 2, 4 kg / da) were used in the experiment.

Table1. Data for sesame cultivation period for Akoren climate

Months	Maximum and Minimum		Average temperature °C		Precipitation
	Day°C	Night°C	Day°C	Night°C	(mm)
June	34 - 29	23 – 14	28,8	14,9	8
July	34 – 27	21 - 14	28,7	14,7	0
August	34 - 14	19 – 9	25,2	10,8	7
September	26 - 9	12 - -2	18,6	5,5	68

Bacillus spp. microorganism cultures were obtained from the laboratories of Selçuk University, Çumra Vocational School of Higher Education and transmission of bacteria to seed was done in the same centre. 5 kg / da of constant nitrogen dose was applied in all experiment plots. No chemical drugs and watering were applied to the experiment plots. Weed control was made by hand. The observations were made on different leaves of 6 plants marked with a portable chlorophyll meter device (Minolta SPAD-502, Osaka, Japan) which indirectly measures the amount of chlorophyll from flowering to harvest. Akoren soils were observed to be a clayey, clayey sand soil texture, which pH is between 6.9 and 8 average to 7.7. According to the samples, the average salt, lime and organic matter were around 0.2, 12.0 and 1.0 % respectively (Celebi et al. 2011).Data for sesame cultivation period for Akoren climate is shown in Table 1.

3. Results and Discussion

In this study, the effect of microbial fertilization and P_2O_5 doses on the leaf chlorophyll content of sesame was investigated, the time from sowing to the first capsule is recorded in Table 2, SPAD measurements with periodic intervals from flowering to harvest are given in Table 3.

Table 2: The time from sowing to the first capsule (Days)

Application	1.recurrence	2. recurrence	3.recurrence	4.recurrence	Average
0	44	49	46	49	**47**
B0	45	45	43	43	**44**
2	46	46	49	47	**47**
B2	46	46	45	43	**45**
4	46	47	46	49	**47**
B4	47	43	45	45	**45**
Average	**45.66**	**46**	**45,66**	**46**	**45,83**

B: Bacteria Applied

Examining Table 2, the first capsule formation in this study was calculated as 45.83 days on average. Administering the doses of P_2O_5 microbial fertilizers, it was determined that the first capsule formation period was 45 days (below the average) and 47 days (above the average) in other parcels. The time taken to the formation of first capsule in the microbial fertilization was about two days earlier. Saharan and Nehra (2011) reported that the useful microorganisms in soil have a biomass enhancing effect by supporting root and shoot growth in the early stages of the development of plants in which they are inoculated.

Plants need much water during flowering period. In places where the production depends on rainfall without watering, crops sown after spring rainfalls grows faster with a significant improvement in yield to ensure that they pass through the generative period before the summer temperature dries up the soil.

Table 3. SPAD Values According to Microbial Fertilization and P_2O_5 Doses of sesame Growth Period

APL	20/J	23/ J	30/ J	03/A	10/A	17/ A	24/ A	31/ A	7/ S	14/ S	21/ S	25/ S	AVE
0	52,05	54,00	56,80	57,55	56,20	55,23	42,85	38,13	34,30	22,78	22,33	22,23	**42,74**
B0	54,25	54,83	58,53	56,93	54,60	54,13	48,50	39,28	39,50	25,90	21,83	18,13	**43,86**
2	52,08	53,68	56,50	57,03	54,95	54,73	46,95	38,35	35,70	23,88	23,28	22,23	**43,28**
B2	52,88	53,88	59,88	56,23	55,18	53,03	48,88	43,30	40,13	22,08	21,80	19,75	**43,92**
4	52,75	54,33	59,78	55,33	55,15	54,10	46,50	42,95	38,68	26,50	26,40	26,23	**44,89**
B4	53,13	55,65	57,95	54,78	54,13	54,40	47,00	39,73	41,62	29,93	26,15	24,88	**44,95**

APL: Application **B**: Bacteria Applied, **J**: July, **A**: August, **S**: September

Considering Table 3, it was observed that the chlorophyll measurements of the sesame plant at the time of flowering in the plots without P_2O_5 and non-microbial fertilizers application, the SPAD values (52.05 – 57.55) showed an increase in the initial 20-day period during which the capsule formation increased. While maturation of the capsules increase, SPAD values (56.20 - 34.30) decrease. But there is a limited change in SPAD values (22.78 – 22.23) in the last 10 days when the colour of the capsules wrapped by the sesame plant and begin turn brown. The average value of SPAD was 42.74. SPAD values of the plots without bacteria and phosphates application are given in Figure 1.

As can be seen from the analysis of Table 3, in chlorophyll measurements of sesame plant starting at the time of flowering of microbial fertilizers and non-P_2O_5 application, SPAD values (54,25 - 58,53) increased in the first 13 day period when capsule formation increased but when the maturity of the capsules was approaching SPAD values (56.93 - 39.50) decrease. In the last 10 days, when the colour of the capsules engulfed by the sesame plant started browning, SPAD values had a slightly change (25.90 – 18.13). The mean SPAD value was 43.86. The results are given in Figure 2 graphically.

As indicated inTable 3, during the chlorophyll measurements of sesame from the onset of flowering at the plots on which 2kg/da P_2O_5 was applied without microbial fertilizer, there was an initial increase in SPAD values (52.08 – 57.03) and it later started to decrease (54.95 – 35.70) with capsules maturity during the first 20 days of capsule formation. The SPAD values during the last 10 days from the time browning started in the capsules wrapped with sesame plant did not recorded any change (23.88 – 22.23).The mean SPAD value was 43.28. The results are shown graphically in figure 3.

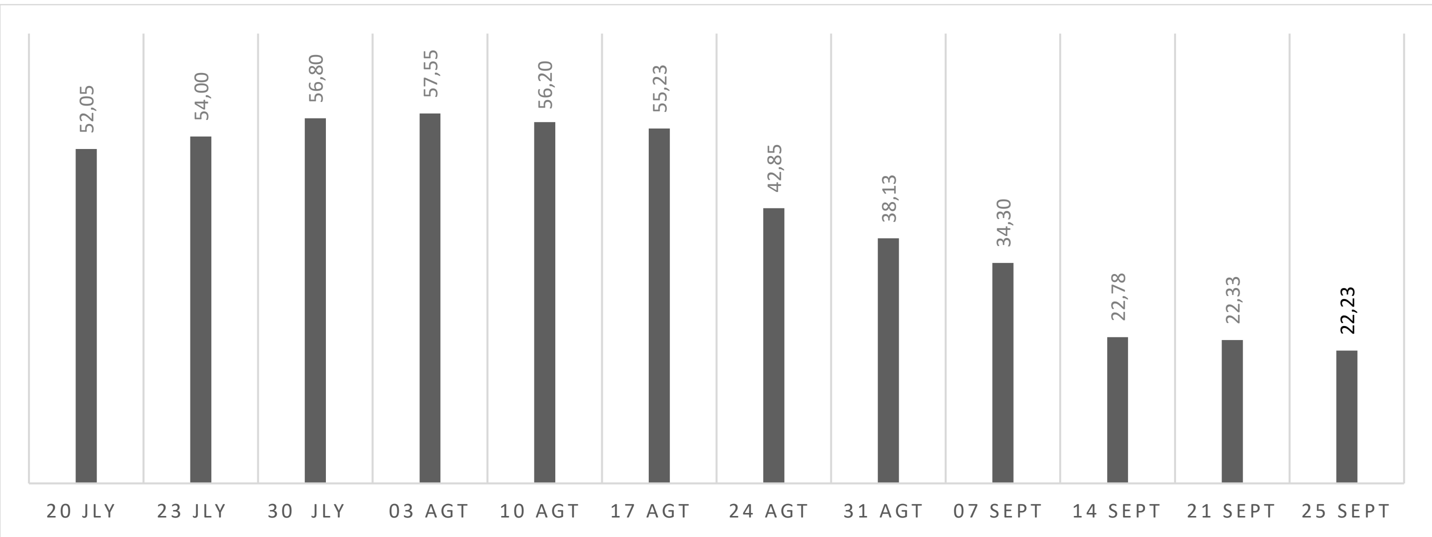

Figure 1. SPAD values of the plots without bacteria and phosphates application

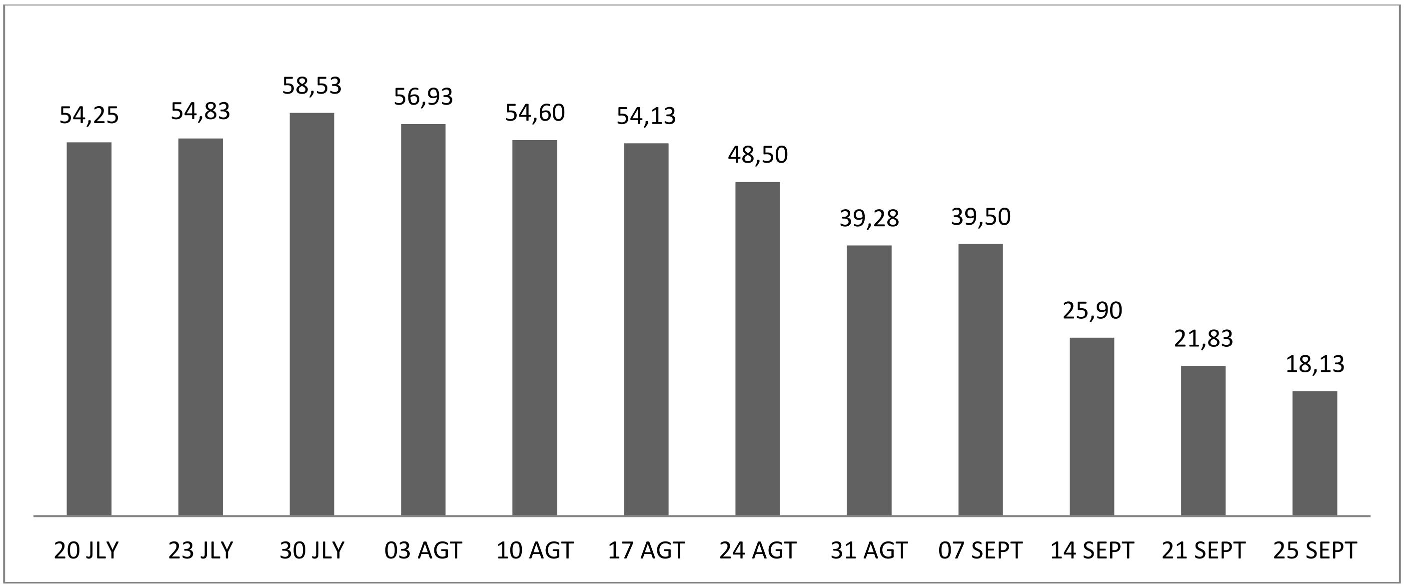

Figure 2. SPAD values of plots with microbial fertilizer and without phosphorus application

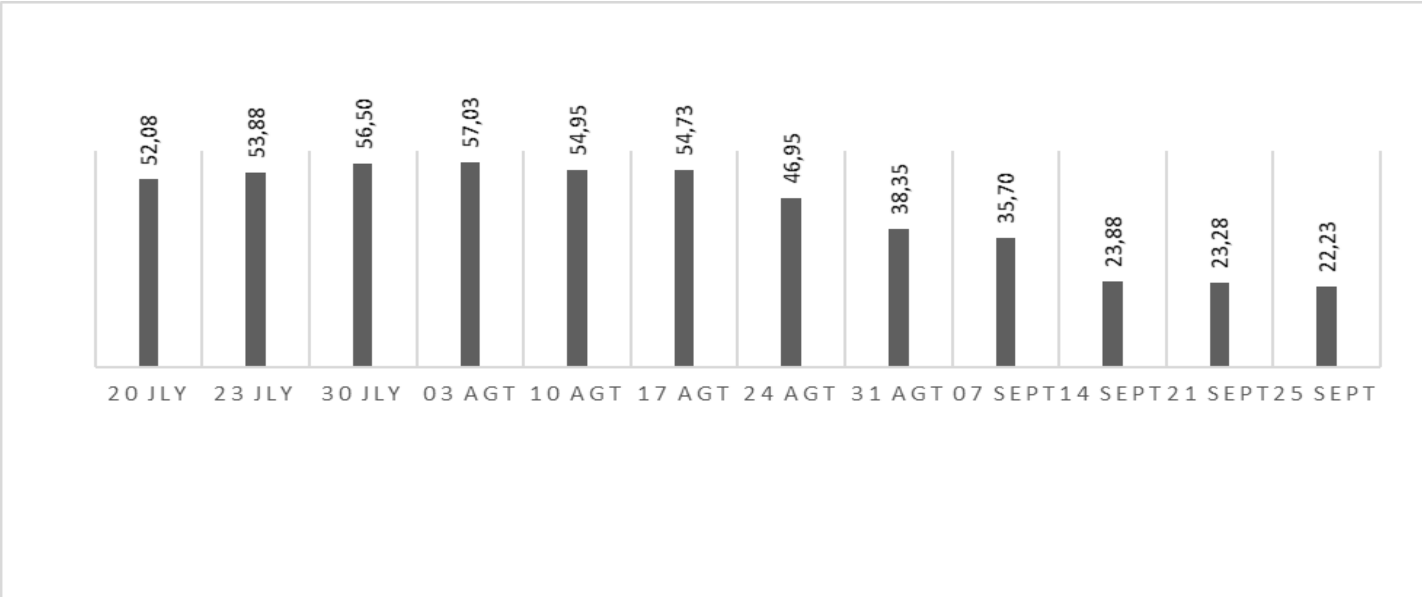

Figure 3. SPAD values of 2kg / da P_2O_5 applied parcels without microbial fertilization

Again in Table 3, the chlorophyll measurements of sesame plants which increased during flowering time in the plots where 2 kg/da P2O5 was used applied together with microbial fertilizer, showed an initial SPAD values increment (52.88 - 59.88) and it started declining (56.23 – 40.13) during maturation at the first 13 days of capsule formation. The SPAD values during the last 10 days from the time browning started in the capsules wrapped with sesame plant had a very little change (22.08 – 19.75). The mean SPAD value was 43.92. The results are shown in Figure 4 graphically.

Showing in Table 3, the results of chlorophyll measurements of sesame from the onset of flowering in the plots on which 4 kg/da P2O5 was applied without microbial fertilizer. There was an initial increase in SPAD values (52.75 – 59.78) and it later started to reduce (55.33 – 38.68) along with capsules maturity during the first 13 days when capsule formation was on the rise. The SPAD values during the last 10 days from the time browning started in the capsules wrapped with sesame plant did not recorded any change (26.50 – 26.23). The mean SPAD value was 44.89. The results are shown in Figure 5 graphically.

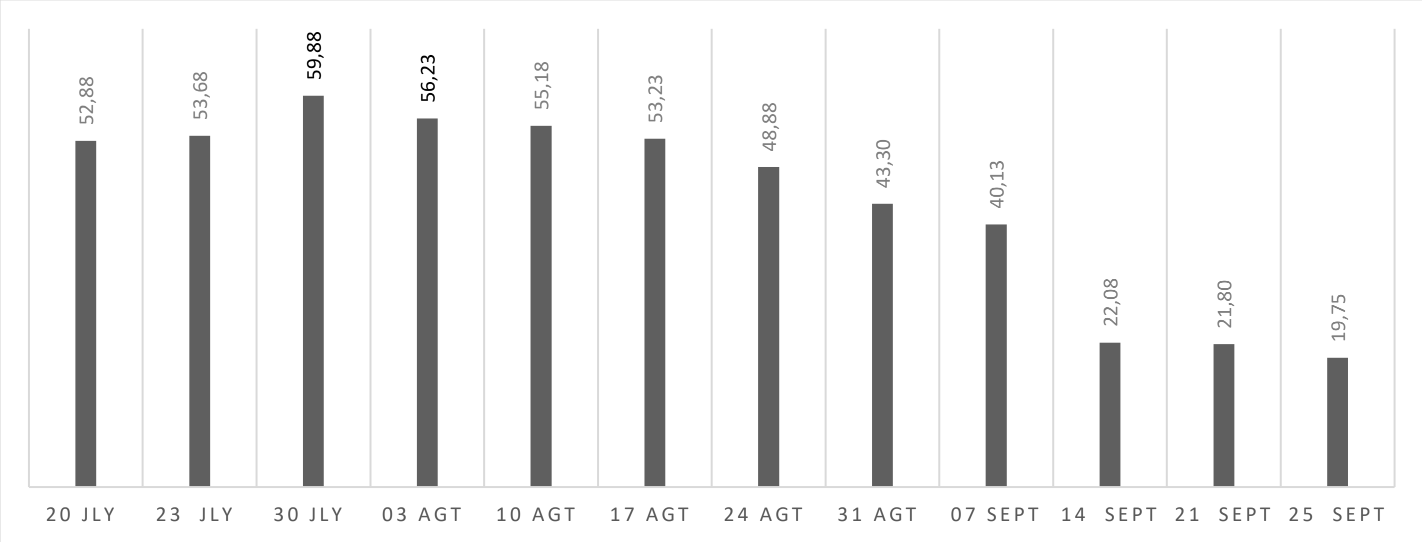

Figure 4. SPAD values of 2kg / da P_2O_5 applied with microbial fertilizer on the plots

Also in Table 3, the chlorophyll measurements of sesame plants which increased during flowering time at the plots where 4 kg/da P2O5 was used together with microbial fertilizer, showed an initial SPAD values increment (53.13 – 57.95) and it started to fall (54.78 – 41.62) during maturation at the first 13 days in which the formation of capsules was increasing. The SPAD values during the last 10 days from the time browning started in the capsules wrapped with sesame plant had a very little change (29.93 – 24.88). The mean SPAD value was 44.94. The results are shown in Figure 6 graphically.

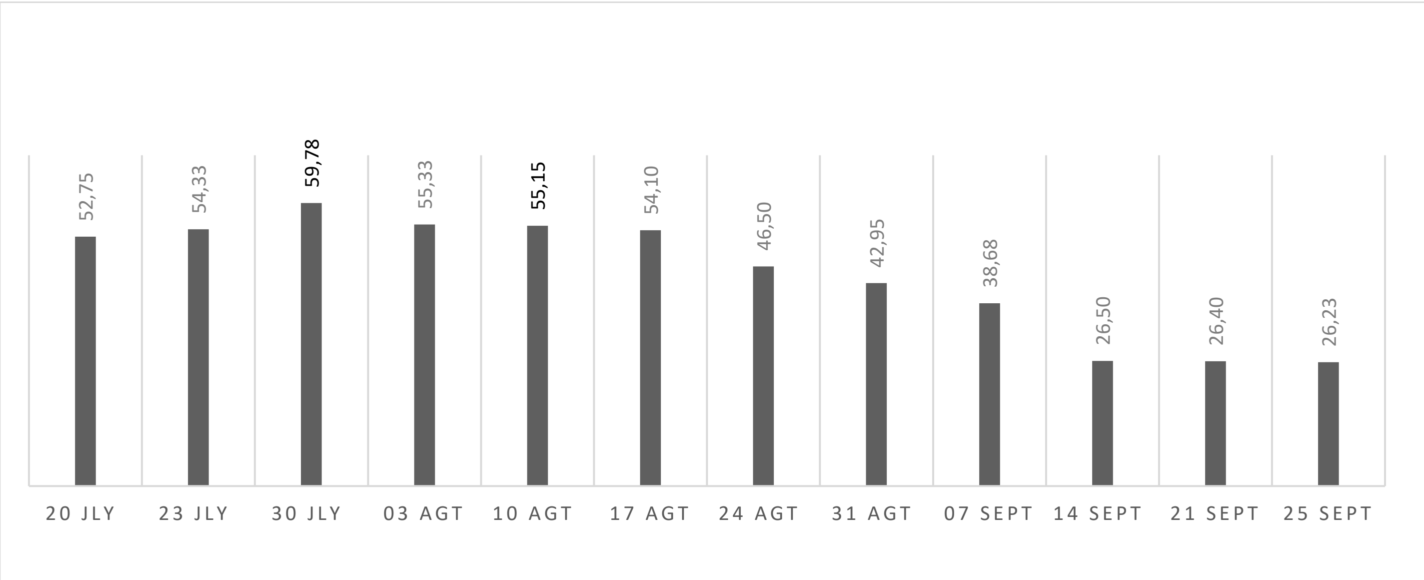

Figure 5. SPAD values of parcels treated with 4kg / da P_2O_5 without microbial fertilization

This study was carried out to determine the effect of different doses of P2O5 on the chlorophyll content in sesame by using natural P2O5 soluble microbial fertilizer application in soil. SPAD values of plots treated with different doses of P2O5without applying microbial fertilizers are shown graphically in Figure 7. In analysing the graph, the highest SPAD value was obtained as 59.78 at the 3rd observation period with the application of 4 kg / da P2O5, while the lowest value was obtained at the last observation (19.75) where P2O5 was not applied.

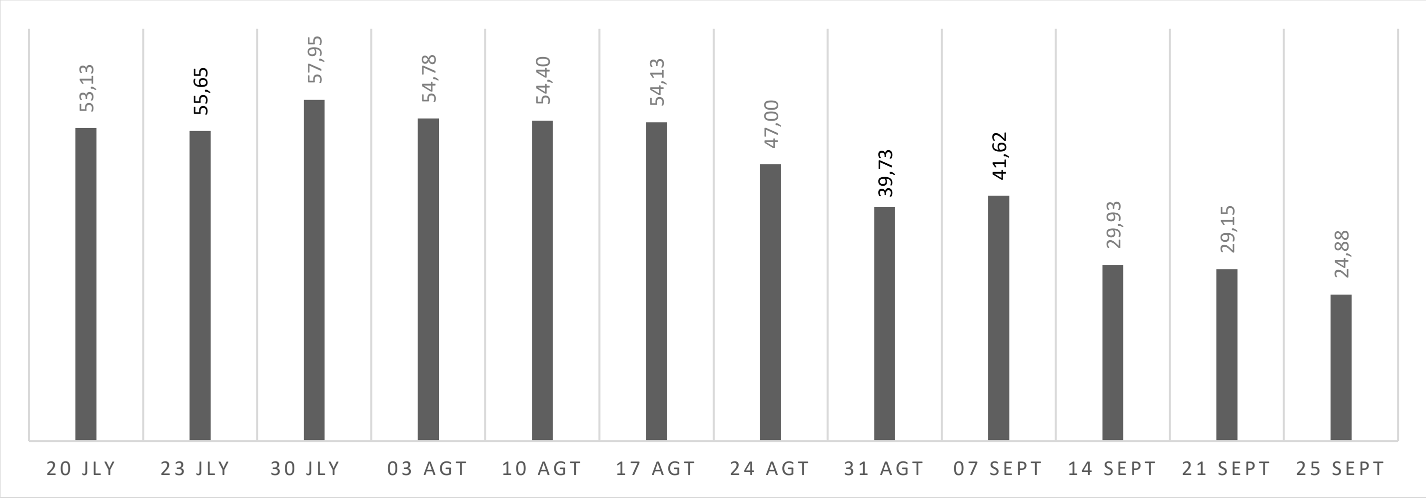

Figure 6. SPAD values of 4kg / da P_2O_5 applied parcels with microbial fertilizer

In this study, it was found that the increasing P2O5 application increased the mean SPAD values as seen in figure 8 (42.74 – 43.28 – 44.89). It has been observed that increasing P2O5 application has limited effect on the chlorophyll content during the sesame flowering and capsule binding period, however, the application of P2O5during the period when the capsules started becoming mature had higher SPAD values as it delayed the breakdown of chlorophyll in the leaves. The fact that the leaves can accumulate organic matter in the grain by making more photosynthesis on the way to harvesting will contribute to increase the efficiency in the unit area.

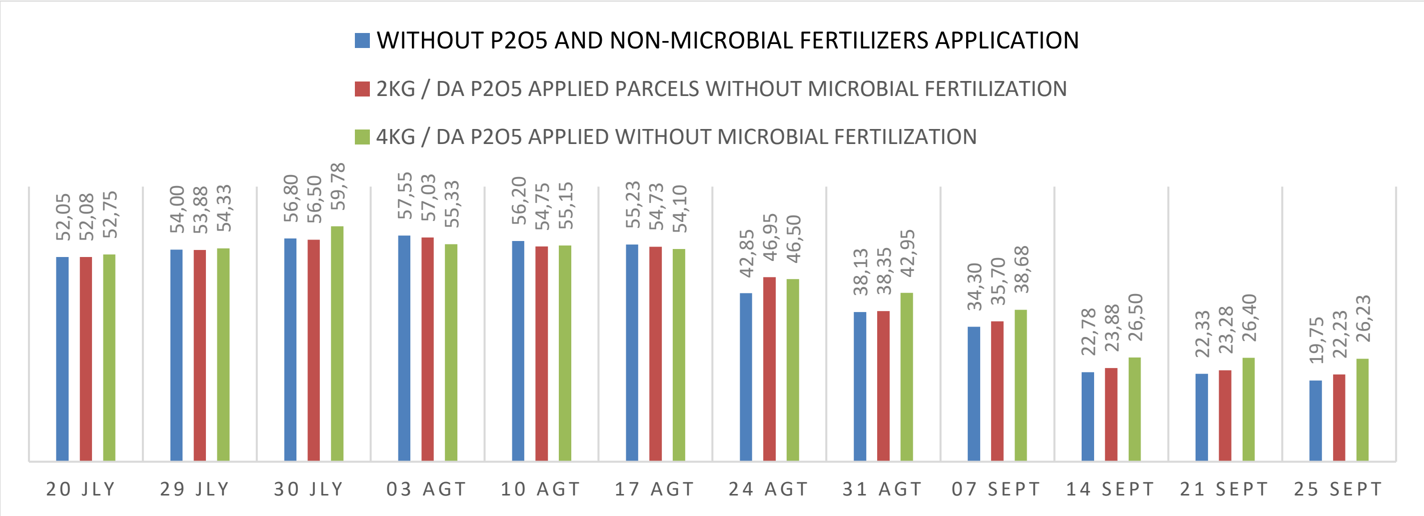

Figure 7. SPAD values of different P_2O_5 doses in non-bacterial plots

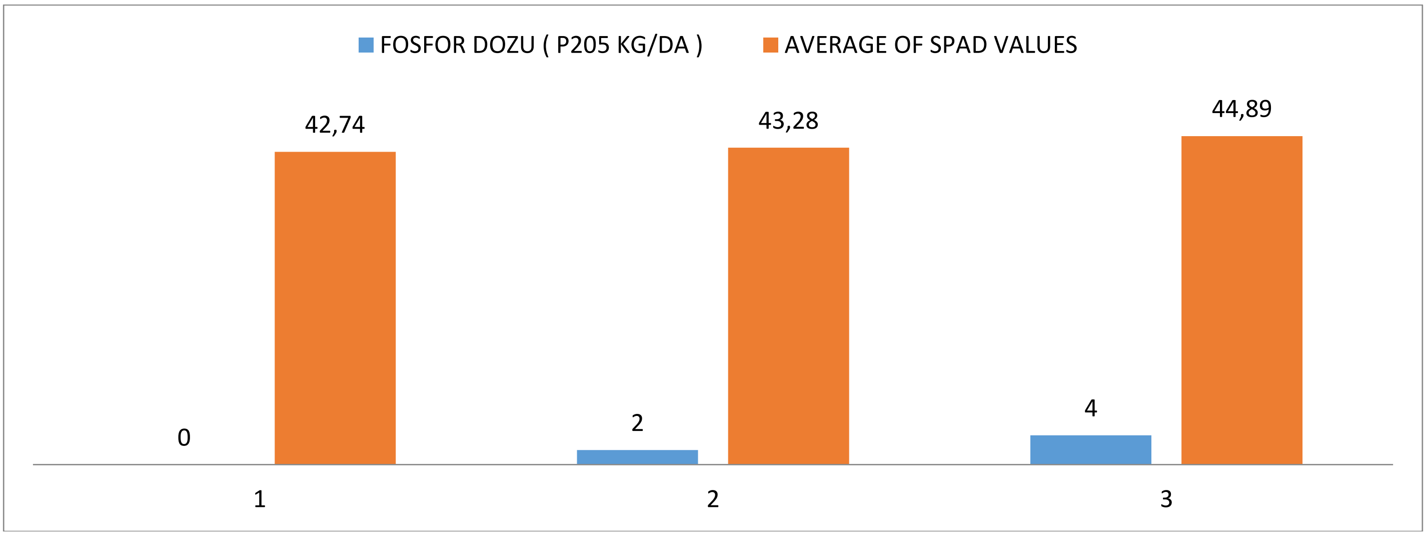

Figure 8. The average of SPAD values of different P_2O_5 doses of non-bacterial plots

This study was to determine the effect of different doses of P2O5 on sesame chlorophyll content with the application of natural

P2O5 dissolving microbial fertilizer present in soil. SPAD values of P2O5 doses in the plots treated with microbial fertilization are given graphically in Figure 9. It can be seen in graph that, the highest SPAD value was 59.88 which was obtained at the third observations of 2 kg / da P2O5 application while the lowest value (18.13) was obtained at the last observation period where P2O5 was not applied. The amount of chlorophyll in plants decreased with aging. Hendry et al. (1987) reported that leaf chlorophyll level is directly related to plant stress and aging. In this study, it was found that increasing P2O5 application led to increase the mean SPAD values as shown in Figure 10 (43.86 –43.92 – 44.95). It was observed that the increased P2O5 application has limited effect on the chlorophyll content during the sesame flowering and capsule binding period, however, during the period when the capsules begin to mature, P2O5 application has higher SPAD values as it delays the breakdown of chlorophyll in leaves.

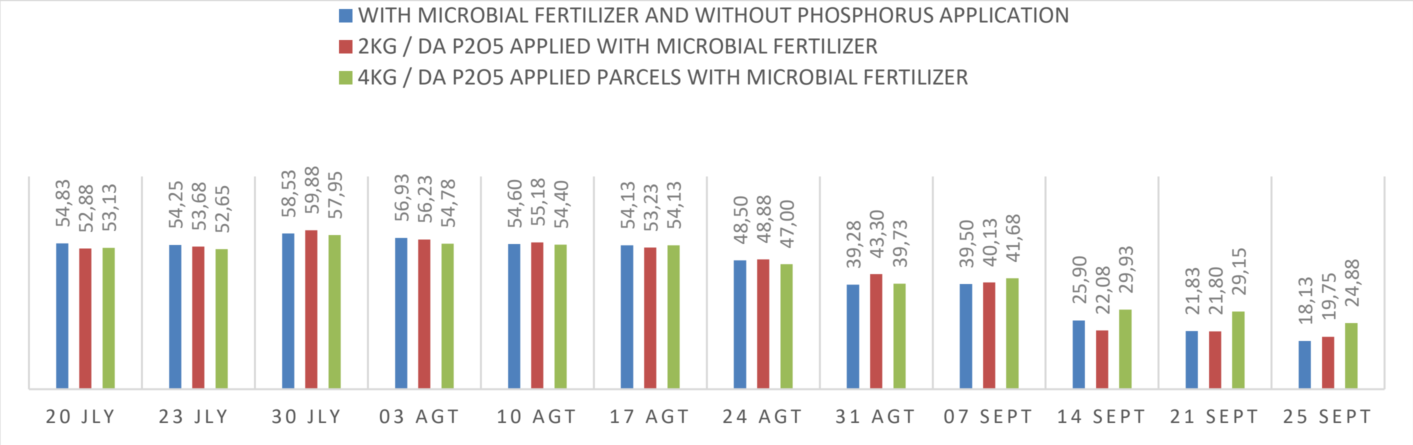

Figure 9. Different P_2O_5 doses of SPAD values in plots applied with microbial fertilizers

In this study, the effect of different doses of P2O5 on sesame chlorophyll content which is natural P2O5 microbial fertilizer dissolver in the soil was determined. SPAD values in the plots applied with microbial fertilization were higher as the P2O5 doses increased. Isame way, the SPAD values in the plots without microbial fertilizer application were higher as the P2O5 doses increased. When Table 4 was examined, the mean SPAD values in the plots with microbial fertilization were higher determined (44.24 – 43.64).

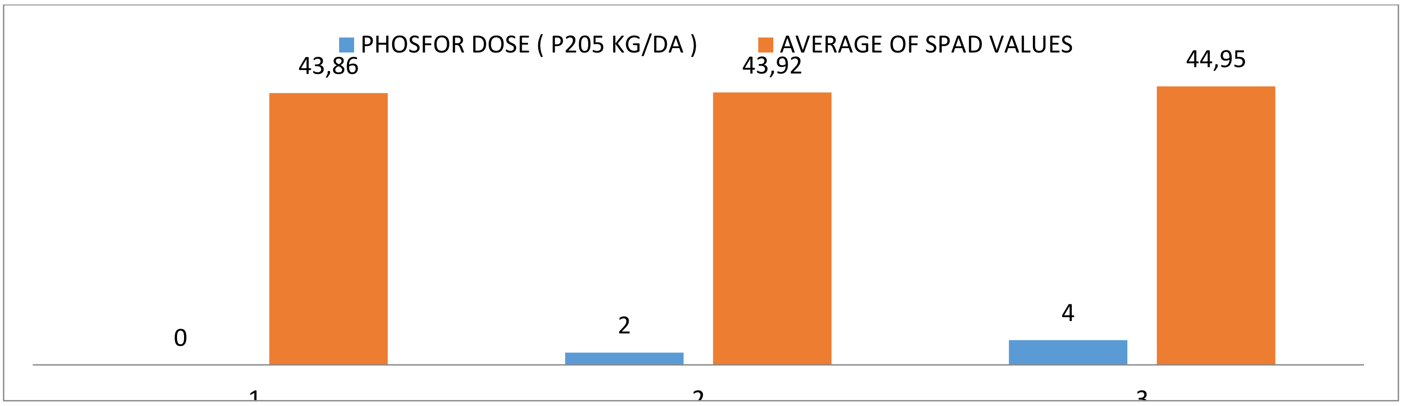

Figure 10. Mean SPAD values of different P_2O_5 doses in microbial fertilized parcels

Table 4. Meaning of SPAD according to Microbial Fertilization and P2O5 doses

P2O5 Dosages Kg / Da	Without Microbial Fertilizer (SPAD)	With Microbial Fertilizer (SPAD)
0	42,74	43,86
2	43,28	43,92
4	44,89	44,95
Average	**43,64**	**44,24**

The average SPAD value of the plots applied with microbial fertilizers was found as 44.24, whereas the untreated plots were found as 43.64 (Table 4). The mean SPAD values of all P2O5 doses were higher in the plots with microbial fertilizer application (Table 4). Some microorganisms that colonize the root stimulate resistance to plant diseases, promote shoot and root growth, increase yield, increase resistance to abiotic stress conditions, promote nutrients intake and use, and increase photosynthesis (Inbar et al. 1994, Yedidia et al., 2001, Harman et al. et al. 2004, Harman 2006). Studies in this area have reported higher chlorophyll content, higher leaf water potential improvement capacity, clear photosynthesis capacity and better water absorption in plants infected with mycorrhizae (Wang 1989; Wang 1998). Mahmood (2015) determined that the main shoot dry weight and total number of leaves per plant has being increased in the trial of applying *Bacillus* subtilis and *Trichoderma harzianum* to Merlot seedlings 2 times. In this study, it was seen that SPAD values of the last observations were close to each other and the range of change was low. These results may help to determine the harvest time in plants with simultaneous problem of death such as sesame. The SPAD values in this study indicate that sesame seeds can be harvested from the 2nd week of September in the autumn season within our region.

4. Conclusion

As a macro element for plants, P2O5 is mixed into soil as fertilizer in case of deficiency. The studies that will increase the usefulness of P2O5 requirements to field cropsand plants should be among the important research subjects especially in the countries where P2O5 are supplied by import. This study was carried out in order to determine the effect of P2O5 and microbial fertilization on chlorophyll content in the seeds of sesame which is useful oil plant in a semi - arid region of Central Anatolia conditions. In this study, portable chlorophyll meter device was used to determine the changes in chlorophyll content from the beginning of flowering to harvest of sesame cultivated on plots that are treated with different doses of P2O5 and microbial fertilizer. As a result, it was determined that increasing P2O5 doses increased the chlorophyll content in sesame. Similarly, microbial fertilization was found to increase chlorophyll content by increasing the usefulness of increasing P2O5 doses in sesame. In addition, SPAD values can be used to determine harvest time in plants that do not exhibit concurrent effects such as sesame. However, there is a need to increase this type of studies for more information.

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